TCAP Quarterly report May 30, 2012

Two-page executive summary

Genotyping: *SNP Genotyping*: The barley iSelect 9,000 SNP barley chip was cross-referenced with previous chips, automatic SNP calling procedures were implemented and the SNP metadata was expanded. The TCAP wheat group mapped 7,517 SNPs from the previous 9,000 SNP wheat chip and played a key role in the development of a new 90,000 SNP wheat chip. Genotyping of the barley and wheat association mapping panels, elite breeding, and genomic selection populations is on target.

New genotyping technologies: The Nimblegen whole exome capture assay targeting 110 Mb of sequence in the wheat genome and 90 Mb of sequence in the barley genome have been designed and tested with promising results (77% of sequence reads mapped to the references and more than 90% of targeted exonic regions were represented). The previous results are being used to design new rebalanced beta versions for wheat and barley. Genotyping by sequencing was expanded into new mapping populations generating thousands of polymorphic GBS tags and high density maps.

The Triticeae toolbox database (T3): The user interface was improved to enable more intuitive searching of the data in T3 and the integration of multiple datasets. We incorporated into T3 several new phenotypic and genotypic datasets. Three online tutorials explaining data submission to T3 have been developed for submitting line names and properties; experiment annotations; and genotype data. Hyperlinks to other databases were created to facilitate users tracking down information about lines or markers. T3 now uses two-dimensional "materialized view" tables to access genotype data. This approach provides quicker access to large blocks of data as well as more compact storage.

Phenotyping: During this period we made significant improvements in the canopy spectral reflectance protocols: improved equipment configurations, developed a more precise measurement protocol and implemented scripts to facilitate the management and analysis of data. This technology is being used to evaluate the NSGC core collections of barley (500 six-row spring accessions) and wheat (540 spring wheat accessions). The best drought resistant lines from the NSGC screen from 2011 have been incorporated into the breeding programs.

Water use efficiency (WUE): In barley, four association mapping populations have been planted in six locations for WUE evaluation and one has been planted for seed increase. In wheat, two association mapping panels have been planted in five (spring) and three (winter) locations and are being evaluated for CSR and other physiological traits. Eight additional specialized mapping populations have been phenotyped for root characteristics, physiological traits associated to WUE, heat stress, and agronomic performance. The chromosome region defining drought tolerance in the 1RS translocation was identified and the beneficial effect of photoperiod sensitivity in early planting rain-fed northern latitudes was validated.

Nitrogen use efficiency (NUE): In barley, NUE is being evaluated using the spring six-row (SP6), spring two-row (SP2), and winter six-row (WN6) association mapping panels in low (70%) nitrogen and normal (100%) nitrogen in three environments. Results from 2011 are being analyzed and incorporated into T3. In wheat, both the hard and soft winter

wheat panels are being evaluated for NUE at two locations (at different N levels) and in four additional locations for yield. All lines have been submitted for genotyping with the iSelect 90,000 SNP wheat chip.

Disease resistance: In barley, the NSGC was evaluated for spot blotch resistance and for spot form net blotch resistance. Nine lines were identified that are highly resistant to current virulent isolates from ND, Australia, New Zealand, and Denmark. The NSGC core collection is currently being evaluated for stripe rust resistance and will be planted in June in Kenya for evaluation for stem rust resistance. In wheat, the analyses of the 2011 data for leaf, stem and stripe rust (1000 spring lines) yielded 35 significant resistance loci. New sources of resistance to the three rust species were identified and are being validated. Seedling screening of the complete core collection for the three rusts will be completed in 2012.

Population development: Nested association mapping populations for barley and wheat were advanced two generations. The development of the wild barley introgression population was completed.

Education: Fifty-nine graduate students have participated in TCAP activities. Thirty-three of these students are funded by TCAP. Twenty undergraduate students are being mentored by TCAP PIs and graduate students, while 15 minority students are being mentored by eight MSI faculty.

Plant Breeding Training Network (PBTN): the online environment was used to deliver and archive three courses, including Plant Breeding Strategies, Entering Mentoring and Quantitative Genetics. To insure sustainability of course offerings, development of an online Plant Breeding Program through Ag*Idea continues with letter of intent approved, business plan developed and Ag*Idea conference attendance. Undergraduate students have been supported in their development through three online meetings with industry representatives and TCAP PIs. The development of three undergraduate educational tools has continued and one tool was submitted to an education journal. The PBTN also has been used as a communication tool for project management both for the TCAP, Ag*Idea Executive Committee, and the NAPB graduate student committee.

Newsletters, films and other communication resources: Information about research and education was shared both internally and externally through six meetings of the TCAP seminar series. Other communication tools include the quarterly newsletters and the face to face meetings at PAG. The TCAP produced film “Holding the future in the palm of your hand” was shown during three recruiting trips to about 200 students. Two of these students applied to TCAP graduate schools. Minority students have been attracted to several internships. The first year evaluation report was received and used to guide second year planning. Evaluation tools were also refined.

Publications and germplasm releases: TCAP participants generated 26 peer reviewed publications during the first 5 months of 2012, which is comparable with the total number of publications for the complete first year. In addition, five new wheat varieties and two wheat germplasm were released.

TCAP May2012 progress report

During the first quarter of the second year the TCAP project progressed as planned without major pitfalls. Progress is described by objectives and deliverables proposed for the second year of the project. The report is organized into seven major sections including a last section on publications and released germplasm at the end of the report. The location of the different sections and subsections in the report are described in the Table of contents below.

Table of contents

A. GENOTYPING

- ***Deliverable: improved barley SNP genotyping platform and maps***

A.1. *Integration of new and old SNP platforms*: To integrate previous barley SNP genotyping with the new iSelect 9,000 SNP barley chip, the Tim Close lab used the information from the design of the SNP Illumina iSelect assay to generate a cross-reference to previous Illumina assays (Close et al. 2009 and HarvEST:Barley assembly #35 unigenes). A new master table of BarleyCAP Core SNP calls in ACGT format using Illumina TopStrand was generated and provided to collaborators and to the T3 database.

A.2. *Improved barley SNP map*: A new barley SNP map containing 2,994 SNPs was provided prior to publication for dissemination through GrainGenes and T3. The new map has higher marker density than the previous map but about 19% higher resolution and improved marker order, and it allocates 100 additional SNPs to chromosome arms.

A.3. *Improved automated SNP annotation*: The project has made advances in the area of automated SNP annotation and automated SNP calling, particularly for barley. In an effort to improve consistency and repeatability of SNP calls in Illumina genotype data, Peter Morrell's lab (UMN) worked with a number of collaborators to implement automated SNP calling. The approach used is implemented in the program Alchemy (Wright et al. 2010). The program was designed by the rice community, and is able to incorporate prior information to improve SNP calls. This information includes inbreeding coefficients for individual samples (to deal with expected heterozygosity), use of known genotypes, and a list of 'bad SNPs'. The accuracy of the SNPs called with Alchemy was tested using multiple approaches, including comparison to genotypes from genetic mapping populations, comparisons to variant calls from Illumina RNASeq data, and comparison to existing SNP calls that were manually curated in the Illumina Genome Studio software. These comparisons suggest that Alchemy SNP calls are comparable to those from manual curation, but that calls are slightly more conservative, resulting in more 'no calls' from ambiguous SNPs. Alchemy SNP calling can be completed in minutes, and results are machine-scored and thus completely reproducible. It will be possible to design future genotyping efforts to include individuals with known genotypes that further improve SNP genotyping accuracy. Barley experience with automated SNP annotation will be very valuable for the analysis of the new wheat 90,000 SNP chip.

A.4. *Improved SNP metadata*: In the absence of a complete reference genome, it is difficult to determine the potential functional consequences of individual SNPs. Potentially, many types of information could be collected for each SNP, including whether the variant is in a genic or non-genic region, in coding or noncoding sequence, and for exon variants, whether the change is synonymous or non-synonymous. This metadata is being compiled for all barley SNPs currently being genotyped, with the goal of submitting all information both to T3 and the GenBank resource dbSNP. The Python tool was used to extract barley annotation from GenBank results for barley SNP contextual sequence compared to barley and wheat BLAST hits and for BLAST hits against the *Brachypodium distachyon* genome. For barley BOPA1 and BOPA2, a total of 2,457 SNPs could be annotated, with 91 SNPs annotated using *Brachypodium* BLAST results. The annotations identified 15% of SNPs as non-synonymous. The ancestral state of mutations can also be inferred relative to an out-group. SNP contextual sequence was

compared to *Hordeum bulbosum* RNASeq data, identifying the putative ancestral state at half of BOPA SNPs. The balance of SNPs will be compared to *Brachypodium* to infer ancestral state. A manuscript reporting the SNP annotation tool is in preparation.

A.5. *Preparation of barley samples for genotyping with 9000 SNP chip*: A total of 960 barley lines will be genotyped using the improved barley custom iSelect 9,000 SNP chip between June and September, 2012. The barley lines to be genotyped include the 384 low-temperature-tolerance AM panel (LTT-AM) and 576 TCAP spring and winter lines from different barley breeding programs. The leaf tissues for the rest of 576 lines were collected at the University of Minnesota and will be sent to Fargo for DNA extraction at the end of May. The seeds for LTT panel will become available in July and will be sent to Fargo for DNA extractions.

- ***Deliverable: improved wheat SNP genotyping platform and maps***

A.6. *7,500 mapped wheat SNPs*: SNPs were mapped in multiple U.S. and Australian segregating populations and a consensus map including 7,517 SNPs has been developed and has been distributed among TCAP participants. A publication including this map is in preparation and the data will be publicly released as soon as the paper is accepted.

A.7. *90,000 iSelect wheat chip*: We have taken advantage of more efficient genotyping platforms and have updated all the genotyping of the wheat association mapping panels previously planned for the 3072-SNP platform to the new 90,000 SNP iSelect wheat chip. A total of 2,500 lines from the spring and winter wheat association mapping panels will be genotyped with the custom iSelect 90,000 SNP wheat chip. Through a coordinated international effort with wheat programs in several countries we were able to increase the initial order of iSelect wheat chips to 46,000 assays, with a dramatic reduction in costs for wheat genotyping. This reduction in cost allowed us to deliver 30-fold more markers than planned in the initial proposal. The Fargo lab will be the central responsible for the genotyping with the iSelect 90,000 SNP wheat chip, which will be completed between June and September 2012. Seeds or leaf tissues for these populations have been sent to the various genotyping labs for DNA extractions.

- ***Deliverable: Marker assisted selection (MAS) with smaller SNP chips***

*Barley GS, 4,032 samples (384 SNP chip)***:** Barley Genomic Selection is scheduled for fall due to unusually warm spring weather and high greenhouse temperatures in MN.

Barley mapping populations, 4032 samples (384 SNP chip): Reagents from Illumina for a 384 SNP barley mapping OPA were received in April 2012. DNA was isolated from 803 wild barley introgression lines and 192 RILs from the Madre Selva/Butta-12 mapping population for CYDV and stem rust resistance. Genotyping with the BeadXpress will begin May 14.

Barley and wheat MAS, 9,546 samples (48 SNP - Sequenom): Analysis of the 9,000 SNP results on multiple wheat mapping populations and association mapping panels is providing the information required for the genotyping labs to incorporate SNP assays into the marker assisted selection programs. Considerable progress has been made developing SNP marker assays for the Sequenom MassArray for low to mid-level SNP genotyping of both wheat and barley samples. The Sequenom platform is cost-effective and provides

greater flexibility than the Illumina BeadXpress since labs can vary the content and numbers of markers assayed on each sample. The list of the different lines genotyped by the smaller SNP chips is included below.

Barley – 192 samples completed

1. OR, Pat Hayes. = 96 samples, 35 SNP
2. MN, Kevin Smith = 96 samples, 48 SNP

Wheat – 6,126 samples completed or in progress with varying numbers of SNP.

1. Louise stripe rust HTAP association mapping validation = 384 samples, 16 SNP
2. Spring wheat quality QTL's validation = 384 samples, 15 SNP
3. Winter wheat MAS = 192 samples, 56 SNP
4. *Yr48* high resolution MAS = 3840 samples, 4 SNP
5. Yellowstone X Choteau mapping population = 182 samples, 248 SNP
6. 1,728 eastern and central winter wheat samples are being genotyped with 72 SNP
7. We assembled seven sets of 200 SNP from known FHB resistance QTL regions to increase our ability to determine haplotypes in the elite breeding lines. The primers were ordered and they will be used to analyze 5 mapping populations segregating for FHB resistance.

B. NEW TECHNOLOGIES

B.1. Gene capture

- ***Deliverable: gene capture technology evaluated in barley and wheat.***

The Nimblegen whole exome capture assay targeting 110 Mb of sequence in the wheat genome and 90 Mb of sequence in the barley genome have been designed and tested by re-sequencing the coding regions of 9 accessions of hexaploid wheat (Truman, Chinese Spring, Rialto, Utmost, Opata, Synthetic W7984, Rac875 and pbw343) and 5 accessions of barley (Morex, Barke, *H. spontaneum*, Bowman, Steptoe). Paired-end sequencing was performed using the Illumina HiSeq2000 and MiSeq instruments. The latter platform was mostly used for quick testing and optimizing the conditions of the capture protocol. The capture reactions were performed either with genomic libraries prepared from a single sample or by pooling four barcoded samples in equimolar proportions. Both levels of multiplexing produced similar results. From 22% to 26% of reads in a pool of 4 genomic libraries could be assigned to individual accessions suggesting the similar levels of enrichment achieved for each accession.

Up to 77% of sequence reads could be mapped to a reference using *bowtie* and *bwa* programs. More than 90% of targeted exonic regions were represented in enriched genomic libraries. The average depth of target coverage after removal of duplicated reads was 50x. The distribution of the depth of read coverage at variable sites differentiating wheat genomes from each other suggested that duplicated genes in a polyploid genome can be captured with similar efficiency using the Nimblegen capture assay. The results obtained are currently being used to design a new rebalanced beta version of the capture

assays. The capture experiment is now being scaled up to perform targeted exome re-sequencing of diverse panels of wheat and barley landraces and cultivars.

B.2. Genotyping by sequence (GBS)

- ***Deliverable: first GBS maps generated for barley and wheat***

GBS was used to generate sequence data for one segregating population for barley and two double haploid populations for wheat. Two of these populations have been already published in Poland et al. (2012).

Barley: GBS was used to genotype the Steoptoe x Morex double haploid population consisting of 129 DH lines and the two parents. A 136-plex library was constructed and sequenced at 2x coverage on the Illumina HiSeq. We identified 19,700 SNPs segregating in this population. In collaboration with the IBSC the SxM mapping data is being combined with other reference populations to develop a barley consensus map of SNPs and GBS markers.

Wheat: The first double haploid population includes 268 individuals from the cross between common wheat varieties Duster and Billings (DxB); the second one 192 individuals from the cross of Synthetic wheat with Opata (SynOp), and the third one a slow rusting population derived from CI13227. GBS libraries were constructed using *Pst*I / *Msp*I, *Pst*I / *Mlu*I and *Pst*I / *Mse*I restriction enzyme combinations. Ninety-two barcoded libraries were pooled to sequence in a single lane of HiSeq2000 instrument. A total of 440 million 100 bp-long reads were obtained for the DxB population with an average of 1.6 million reads per individual and 408 million reads were obtained for SynOp mapping population with an average of 2.1 million reads per individual. The data was processed to generate genotype calls using a custom pipeline. The Illumina read clustering performed with CD-HIT program was used to obtain unique reference tags. A total of 1.1 million and 1.2 million unique tags were obtained for DxB population. Similar results were obtained for SynOp mapping population. These clusters have been used as a reference sequence for mapping Illumina reads using the *bowtie* program. SNP genotype calling in the Illumina alignments was performed using the *SAMtools* program. The presence/absence and SNP variations identified between the parents of mapping populations were scored in the mapping populations to call individual genotypes. Both types of variation were tested for 1:1 allele ratio using the binomial distribution.

By allowing up to 80% of missing data, 59,907 potential SNP sites have been identified in the DxB population. Among these variable sites, 1,690 SNPs that showed high quality and low proportion of missing data were selected for constructing the genetic map. The GBS tags generated for SynOp mapping population were mapped to recombination intervals of the SynOp genetic map constructed using DArT, SSR and SNP markers. Mapping to recombination intervals was based on testing of two-marker configurations for independence using binomial distribution with the p-value < 0.001. A total of ~60,000 SNP and 390,000 PAV GBS-tags were integrated with the SynOp genetic map.

B.3. Genome selection

- ***Deliverable: first cycles of crosses completed in barley***

Barley lines from Cycle 1 were generated for seed increase and future evaluation and crossing parents for Cycle 2. The six-row winter barley genomic selection project is based on forty-seven facultative parents tracing to Oregon, Nebraska, Idaho and Minnesota breeding programs. In the fall of 2011, 768 F3 plants tracing to 48 crosses were genotyped by the NC Genotyping Lab with a custom 384 SNP VeraCode assay optimized for polymorphism among the parents and genome distribution. All of this cycle 1 genotyping data has been uploaded to T3.

A training population was assembled using phenotype and genotype data from the Barley CAP. Jean Luc Jannink trained a prediction model using the RKHS method with a Gaussian kernel for the traits: yield, malt extract, low temperature tolerance (LTT), height, heading date, Fusarium head blight, and stripe rust. Little to no negative correlation among the traits was observed and a selection index primarily weighting yield, LTT and malt extract was used to select the best 100 lines to advance to the next generation and to be used as parents in the next cycle of crossing. A random set of 100 lines was also selected. The remnant seed from all 768 F3 plants was planted in single row plots in St. Paul in the fall of 2011 to assess LTT. The correlation between the genomic estimated breeding value (GEBV) for LTT and observed winter survival was 0.6. In the winter greenhouse, crosses among the selected F4 parents were made to generate over 900 F1's for the next round of selection. A slightly delayed planting and a very unseasonably warm spring created a situation where we determined that it would be too risky to make crosses for cycle 2 in the greenhouse. Our plan now is to plant the F_2 generation in the field, which is being done the week of May 15 and genotype 768 F_3's in the fall for cycle 2 selections. We have planted the 100 selected and 100 random $F_{4:5}$ lines from cycle 1 as two row plots in Crookston to generate seed for preliminary yield trials that will be conducted in MN and OR in the fall of 2012.

C. THE TRITICEAE TOOLBOX (T3) DATABASE

- ***Deliverable: improved T3 database***

C.1. *New data released*: New datasets uploaded to T3 since the beginning of 2012 have been primarily new barley allele calls on the University of Minnesota breeding program, and agronomic data spanning three years on a series of height allele NILs in wheat. Wheat disease data on the NSGC core collection and barley elite association mapping panel data are expected in the coming quarter. A schedule for phenotypic data delivery has been assembled to provide milestones for each expected dataset from TCAP participants (http://bit.ly/I9plRt). This schedule will be regularly updated to help ensure timely data submission to T3 and therefore delivery of TCAP data to participants and stakeholders.

C.2. *Improved user interface*: Several improvements in the user interface have been made to enable more intuitive searching of the data in T3 and the integration of multiple datasets. Users can now identify data of interest starting from a focus on the breeding program generating the data; from locations from where the data were collected; from traits measured; or from the identities of the lines themselves. Each initial focus leads in an intuitive way to search restrictions based on other criteria until the user has narrowed

the search to a precise combination or union of criteria. Users may define panels of lines that they expect to work with on a regular basis. Upon login, data for user-defined panels can be queried directly. A BLAST search feature has been implemented allowing users to identify barley or wheat markers with sequence similarity to arbitrary input sequence.

C.3. *T3 training*: Three online tutorials explaining data submission to T3 have been developed for submitting line names and properties; experiment annotations; and genotype data. In conjunction with the tutorials, we created a YouTube channel to show tutorial videos. The line submission video has had 13 views.

A poster and a computer demonstration were presented at the 2012 Plant and Animal Genome Conference. A seminar on uploading data to T3 was presented to the Plant Breeders Training Network on April 12th, 2012. The "sandbox" versions of T3, developed during the fourth quarter of 2011 have been successful in enabling users to test-run data submission prior to transmitting the data to the curator. Seven TCAP participants from five programs have submitted data to T3 via the sandboxes.

C.4. *Coordination and hyperlinks with other databases*: Hyperlinks to other databases can facilitate users tracking down information about lines or markers of interest. The following hyperlinks to other databases now exist in T3: Marker name query on GrainGenes; Rice homologous gene on Gramene; Affymetrix probeset query on PLEXdb; UnigeneID query on HarvEST; Line query to PI number in GRIN

T3's trait descriptions have links to the corresponding Trait Ontology records at www.gramene.org/db/ontology, to allow users to access data about those traits from other sources. We are working with Laurel Cooper of the Plant Ontology Consortium to create links from the Trait Ontology back to T3's data about each trait. For example the link for "beta glucan" would jump to T3's page with all the trials in which that trait has been evaluated.

C.5. *Solutions to the "Big Data" problem***:** T3 now uses two-dimensional "materialized view" tables to access genotype data. This approach provides quicker access to large blocks of data as well as more compact storage. These tables have dramatically decreased query times for retrieval of genotype data used both for internal analyses (e.g., clustering lines by genotype) and for download to external analyses.

T3 developers are maintaining contact with other databases confronted by the "Big Data" problem, in particular with Panzea and MaizeGDB.

A new server dedicated to T3 is scheduled to come online by the end of May 2012. This server was designed with TCAP datasets in mind, in particular aiming to be able to maintain the complete table of allele calls in random-access memory to increase the speed of genotype queries.

C.6. *Expanded use of T3 beyond TCAP*: The T3 database schema and web interface software are potentially useable by any project that generates genotype and/or phenotype data for wheat or barley. With funding from the US Wheat and Barley Scab Initiative, we have created a T3 instance called The Breeders Database, http://malt.pw.usda.gov/t3/bd/, to hold the results of current and historical Uniform Regional Scab Nurseries. USWBSI

participants Dave Van Sanford and Paul Murphy are taking the lead on populating this database, and Dave's student Sandy Swanson has begun adding data.

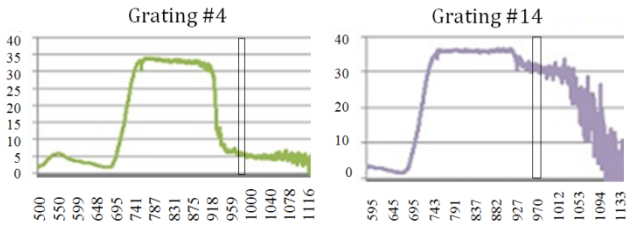
Efforts have begun in individual breeding programs to use T3 itself for their own data management. The University of Minnesota breeding program has been particularly active, submitting line names and properties for over 5,000 lines to T3 Barley. Wheat programs at the University of Nebraska, Lincoln and at Cornell University are also making steps in this direction.

We have packaged T3 and added installation instructions to make it "portable" for installation on any Unix computer, including Mac OS X. It is available for downloading from GitHub (a service similar to Source Forge). A student of Hermann Buerstmayr found it there and has successfully installed it at University of Natural Resources and Life Sciences, Vienna, Austria, for a database of Fusarium Head Blight disease reactions. The modifications necessary to make T3 easily customized for any crop are relatively small, and we are planning to make them.

D. PHENOTYPING

D.1. Canopy Spectral Reflectance (CSR)

- ***Deliverable: Improved CSR methods***



The previous configuration of the Jaz spectrometer resulted in limited efficiency in the NIR wavelengths (970 nm rectangle in the figure). The problem was corrected by replacing grating number #4 by grating #14 and using improved mirrors (see figure). Grating #14 provided improved efficiency in the NIR wavelength used in the water indexes (970 nm). We determined that it is important to use silver mirrors since the basic aluminum coated mirrors absorb near the key wavelength of 970 nm. Silver mirrors have excellent reflectance properties in the VIS-NIR range.

TCAP PhD student Tyson Howell introduced a new scanning protocol that greatly improved the precision of measurements. Previously, we took 3-4 “point” measurements and that resulted in large variability within plots. This problem was solved by writing a new script which allows the user to “scan” along the plot, integrating 400 images into a single plot reading. This method also saves time when collecting data and analyzing data, as the scanning method saves only one file per channel instead of 3-4. CSR data can be collected from 60 plots in 30 minutes with three channels and in ~15 minutes for 2 channels, a great improvement from the previous method. The use of 400 subsamples instead of 4, greatly improved the precision of the measurements of individual plots.

A Perl script has been written to expedite processing of the CSR data. The script parses together information from all plots and merges them into a single tab delimited file. This can then be opened using excel, and several indices can be calculated for hundreds of

plots in a matter of minutes. A web seminar was organized to update the TCAP users of CSR of these improvements (April 26, 2012, 30 participants). The TCAP integration of all US wheat and barley breeding programs in a collaborative project greatly facilitated the nation-wide rapid implementation of these technological advances.

The TCAP coPI from the University of Nebraska has established a collaboration with the Center for Agricultural Land Management and Information Technology (CALMIT) for collecting canopy spectral reflectance (CSR) of their NUE trial. They are using two pole-mounted fiber optics, each attached to Ocean Optics USB2000+VIS-NIR spectrometers. Data collection was initiated May 1, 2012. The system operates as a dual-fiber optic system with two inter-calibrated spectrometers (Rundquist, et al. Computers and Electronics in Agriculture, 43, 173-178). The spectrometers are housed in a backpack, which also contains a 12 volt battery, a voltage converter (12V to 5V), a serial interface, and a wireless router. Reflectance data from the spectrometers is transmitted to the serial interface which converts the serial signal to Ethernet signal for transfer to the wireless router. Data is transmitted to a laptop computer, which uses the CALMIT Data Acquisition Program (CDAP) both to operate the spectrometers and record data. CDAP facilitates control of the spectrometers and visual confirmation of scan quality. The program is capable of automatically adjusting acquisition parameters of both spectrometers and can automatically process the entire raw data directory into an easy to use Excel format. Each scan is entered into an annotatable log in the CDAP software to enable correction of plot identification or notation of errors in scanning procedure. CALMIT also offers aerial spectral monitoring by a small aircraft. We have committed to at least one flyover to collect CSR data on the trial. If data from the flyover proves to have sufficient resolution and to complement the backpack collected data, additional flyovers will be completed.

D.2. Phenotypic characterization of the NSGC core collections

- ***Deliverable: NSGC core collection evaluated for yield, NUE and WUE.***

D.2.1. *Barley:* The focus for year two is spring barley (year one was focused in spring wheat). Over 500 six-row spring barley accessions from the NSGC core collection and five checks used in the elite panel were planted in an augmented complete block design in a drip irrigation nursery at Aberdeen, ID. The design was replicated under three different water/nitrogen treatments: normal water and normal nitrogen, terminal drought and normal nitrogen, terminal drought and low nitrogen.

D.2.2. *Spring wheat:* Although not planned in the original TCAP proposal, we decided to retest the 540 spring wheat NSGC accessions evaluated in 2011 to obtain a more precise evaluation of the phenotypes and to test the reproducibility of the WUE results across years. The 540 wheat lines are being tested under two treatments: normal water and terminal drought (both with normal nitrogen). All wheat and barley accessions are being evaluated for water and N use efficiency (WUE and NUE) using Canopy Spectral Reflectance (CSR) equipment upgraded with a #14 grating system and silver mirrors.

D.2.3. *Winter Wheat:* Over 600 winter wheat accessions were planted in a rain-fed environment in Rockland for a preliminary evaluation of yield and agronomic

performance and for a seeds increase for the 2013 evaluation. A total of 256 elite spring wheat lines were planted under limited water conditions in Aberdeen and will be evaluated for yield and WUE.

D.2.4. *Breeding applications of the 2011 data:* Data collected from 2011 were submitted to T3 and is being used to development a Mini Core of the wheat core. The 2011 evaluation also yielded useful materials for the breeding programs. Out of the 540 spring wheat accessions evaluated in 2011, thirty were selected and used in crossing in several breeding programs. A set of 123 hard white spring wheat accessions has been used in association analysis to identify QTL associated with late maturity alpha amylase, an important quality defect that is widespread in hard spring wheat varieties in the western USA. Three chromosome regions were identified. This set of materials has also been used in association mapping for fusarium head blight and baking quality.

D.3. Phenotyping for water use efficiency

- ***Deliverable: barley populations evaluated for WUE***

D3.1. *Spring 2-row (SP2) and 6-row (SP6) panels***:** Seed for the spring 2-row (256 lines) and spring 6-row panels (256 lines) was distributed by UMN. Randomizations for Type II modified augmented design with repeated checks were generated at OSU. For the SP2 panels, trials were planted at four locations: Fort Collins, CO (dry and normal), Aberdeen, ID (dry and normal) Bozeman, MT (dry) and Huntley, MT (normal). The Fort Collins trial is under the direction of Joshua Butler (Busch Agricultural Resources).The Aberdeen, ID trial is under the direction of Gongshe Hu. The two MT trials are under the direction of Tom Blake. For the SP6 panels, trials were planted at one location in Fort Collins, CO (dry and normal). All trials will be repeated again in 2013. WUE will be determined based on comparing phenotypes under a normal/full irrigation schedule and limited irrigation schedule (50-60% of normal). WUE will be determined based on comparison between the normal and dry treatments for a range of phenotypes based on multiple trials conducted in 2012 and 2013. Genotyping was completed.

D3.2. *Winter 6-row (WN-6)*: Seed was increased for the winter 6-row panel (300 lines) in the Oregon State University greenhouses. The experiment was planted at Corvallis, OR on May 11, 2012 using a Type II modified augmented design. Spring planting is possible due to the facultative growth habit of most accessions. The trial is under the direction of Pat Hayes and Alfonso Cuesta-Marcos and is part of Araby Belcher's Ph.D. thesis research. WUE will be determined based on comparing phenotypes under a normal/full irrigation schedule and limited irrigation schedule (50-60% of normal). The panel will be genotyped with the 9K iSELECT SNP chip in the summer of 2012.

D3.3. *The Barley World Core evaluated for low temperature tolerance (LTT)*: Fall planting is a valid strategy to improve WUE, but it requires genotypes tolerant to low temperatures. A set of 400 accessions from the darley Word Core was planted at St. Paul, MN and Pendleton, OR in the fall of 2011. The St. Paul, MN trial is under the direction of Kevin Smith (UMN). Differential winter injury was observed. Injury was rated as the % of the plot surviving. The Pendleton, OR experiment is under the direction of Pat Hayes and Alfonso Cuesta-Marcos (OSU). Data were received by OSU and are being analyzed. Differential yellowing was observed and plots were rated accordingly. Data are

being analyzed. Genotyping with the 9K SNP chip is completed and data are in T3. Phenotype data are in the queue to be uploaded to T3.

D3.4. *LTT Panel*: Germplasm (384 lines) with potential LTT was solicited from around the world. These accessions will be genotyped with the 9K SNP chip in the summer of 2012. The James Hutton Institute provided an additional 344 accessions, already genotyped with the 9K iSELECT SNP chip. The winter 6-row panel (300) will be added to this set; it will have been genotyped with the 9K SNP chip for the purposes of NUE and WUE. The final panel will be reduced to 1,000 accessions. This panel will be tested for LTT in MN and increased for seed in OR starting fall, 2012. An international consortium has been formed to test the panel for LTT in 2012-2013.

- ***Deliverable: wheat populations evaluated for WUE.***

D3.5. *Hard winter wheat association mapping population for WUE*: The population that was planted in Greeley, CO experienced drought conditions due to a very dry late winter and spring. Through controlled drip irrigations, we were able to create two very distinct soil moisture environments in our wet and dry treatments, with large differences in leaf area, tiller number, and biomass. Canopy spectral reflectance measurements with the JAZ instrument have begun on a weekly basis. We plan to measure relative water content on a subset of lines at two growth stages to correlate with the JAZ readings. At harvest, data will be collected on biomass, yield components, and test weight. The panel has also been planted under dryland and irrigated conditions in Kansas. All traditional yield component data will be obtained and one reading of CSR has been accomplished. Two field trials of the same population are also growing in Etter and Bushland, TX and are likely to experience drought stress this year, though not as severe as in 2011. Data will be recorded at those sites for maturity, plant height, and yield components. Seed produced this year will be used for next year's WUE plots.

Analysis of root traits in the Hard Winter Wheat Association Mapping Panel included evaluation of the 30 Colorado cultivars and advanced lines in a replicated greenhouse trial. Plants were grown in PVC tubes 1 m deep. Starting at the four-leaf growth stage, water was withheld from plants for three weeks, followed by harvest of above- and below-ground plant parts. Roots were cleaned, stained with methylene blue for better contrast, scanned for digital analysis, dried, and weighed. WinRhizo root analysis software (Regent Instruments, Quebec, QC) is being used to analyze digital scans of root biomass from each of three depths in the tubes (top, middle, and bottom thirds). Measurements of root diameter, root length per diameter class, and root volume will be compared among rooting depths and genotypes. In addition to root traits, measurements of water use efficiency were also made.

D3.6. *Hard spring wheat association mapping population for WUE*: A final set of 250 elite spring wheat lines from 12 breeding programs was assembled. Seed has been supplied to the USDA Genotyping Lab in Fargo ND for genotyping with the iSelect 90,000 SNP wheat array. Data from the 2011 trial in Montana was entered into the T3 database. Seed was also provided for the 2012 trials in Washington (two locations), California (dry and irrigated), Montana (two locations), Kansas (dry and irrigated) and one location in Idaho. Field experiments have been planted as augmented designs with six checks and five blocks. Seed was provided to a cooperator in Saskatchewan to one

location in Canada and for seed production for additional Canadian locations in 2013. A cooperator in North Dakota has planted the panel to obtain cereal quality data for association analysis. The Spring Wheat AM Panel is also being measured for root traits (Kansas) using plants grown in 1.5 m columns in a green house. Data on plant height, number of tillers, and rooting depth is currently being analyzed. Roots of selected genotypes with maximum, minimum, and intermediate rooting depth will be subjected to complete image analysis (WinRHIZO software). The field evaluation of the AM panel in California was completed. The field was equipped with soil water sensors at 30 and 60 cm depth and soil water content was continuously monitored. Vegetative growth has been monitored by taking early vigor scores, number of elongated nodes, heading dates, and plant height at three consecutive dates. SPAD leaf greenness and CSR have been taken at three consecutive stages.

D3.7. *Mapping of the 1RS drought tolerance gene(s)*: The objective of this experiment is to map the drought tolerance gene present in the rye 1RS.1BL translocation and to separate it from the rye loci that negatively affect bread making quality. Water index NWI3 was determined using the new CSR protocols in four sets of near isogenic lines (NIL) with different combinations of segments of wheat chromosome 1BS introgressed into the 1RS translocation. The lines with two interstitial wheat segments (1RS-MA) and those with only the distal wheat segment were very similar and significantly more susceptible to drought than the sib lines with either the complete 1RS chromosome or only the proximal wheat segment (replacing the rye *Sec1* locus). These results indicate that the rye gene(s) for drought tolerance is (are) located within the distal rye segment replaced by the wheat translocation. We are now initiating the dissection of this distal segment. This result has important agronomic implications since it demonstrates that the rye *Sec1* locus associated to sticky dough can be removed without affecting drought tolerance. This information is being used to engineer a chromosome that combines the distal 1RS segment including the drought tolerance genes, the *Yr15* stripe rust resistance gene, and the strong gluten allele *Glu-B1 7Bx-over-expressor*.

D3.8. *Evaluation of three genes for semi-dwarf growth habit*. The objective of this experiment was to assess the impact of the height-reducing gene *Rht8* relative to the standard *Rht-B1b* and *Rht-D1b* alleles on performance of spring wheat in Montana, Washington and California environments characterized by terminal drought stress. Evaluation of near-isogenic lines developed in four genetic backgrounds showed that *Rht-B1b*, *Rht-D1b*, and *Rht8* caused height reduction of 19, 20, and 6.5%, respectively, relative to wild-type near-isogenic lines over 12 environments (Table 1).

Table 1. Plant height and grain yield for near-isogenic lines with three different *Rht* genes for semi-dwarf growth habit from 10 environments with terminal drought.

Genotype	Plant Height (cm)	Grain Yield (kg/ha)
Recurrent Parents	98 a	3352 b
Rht-B1b semidwarf	80 c	3571 a
Rht-D1b semidwarf	79 c	3561 a
Rht8 semidwarf	91 b	3271 c

Means followed by the same letter are not significantly different ($P < 0.05$).

Significant increases in grain yield were associated with the *Rht-B1b* and *Rht-D1b* genes (Table 1). Lines with *Rht8* yielded less than wild-type. Lines with *Rht-B1b* and *Rht-D1b* tended to have a higher harvest index and more seed per spike than wild-type lines and reduced height lines with *Rht8*. In sum, our results suggest that *Rht-B1b* and *Rht-D1b* are superior to *Rht8* as a source for height-reduction for spring wheat in environments characterized by terminal drought stress.

D3.9. *Relationship of photoperiod response and adaptation to a changing climate*: Spring wheat lines are divided into photoperiod insensitive (PI) or photoperiod sensitive (PS), the latter requiring long days for flower initiation. To assess the impact of earlier spring wheat plantings due to climate change, sets of near isogenic lines for photoperiod sensitivity were tested at three planting dates in 15 environments in Montana, Washington, Saskatchewan and Alberta.

Table 2. Grain yield (kg ha^{-1}) of photoperiod sensitive (PS) and photoperiod insensitive (PI) near-isogenic lines for three recurrent parents tested over 15 environments.

Photoperiod genotype	Planting Date		
	Early	Middle	Late
PI	2832	2638	2341
PS	2959***	2684*	2310

*, **, *** Significant at $P < 0.05$, 0.01, and 0.001, respectively.

Grain yield was significantly greater for PS lines at the first two planting dates, though no difference between PS and PI lines occurred for the latest planting date (Table 2). Our results suggest that PS lines are superior to PI lines for the northern Great Plains. This difference is likely to be enhanced as planting date becomes earlier due to increasing spring temperatures.

D3.10. *Evaluation of near-isogenic lines for stem solidness and stay-green phenotypes*: Previous experiments with recombinant inbred line populations allowed identification of QTL for major genes for stem solidness and long green leaf duration after heading. Both traits have shown association with drought tolerance in previous studies. Near-isogenic lines for these QTL alleles were evaluated at two Montana locations in 2011. Results confirmed the impact of the solid stem QTL, but did not confirm the stay-green QTL. Larger experiments are planted at two locations in Montana and two locations in Washington for 2012. The locations vary for drought potential. The primary goal of these experiments in 2012 and 2013 will be to determine the impact of physiological traits stem solidness and stay-green leaves on drought tolerance.

D3.11. *Evaluation of hexaploid and tetraploid RIL populations from a hexaploid / tetraploid cross*: The goal of this experiment (led by TCAP Ph.D. student Jay Kalous) is to identify QTL for drought tolerance that may be exchanged between hexaploid spring wheat and tetraploid durum wheat. A RIL population containing ~100 lines for each ploidy level was developed by crossing hexaploid Choteau with tetraploid Mountrail. Initial data with microsatellite markers has shown that recombination occurred in the progeny lines such that the A and B genomes are a mixture of DNA that originated from the 4X (tetraploid) and 6X (hexaploid) parents. Table 3 shows mean grain yield of the

tetraploid and hexaploid RIL sets based on 2011 Montana data. The hexaploid RILs tended to be superior to the tetraploid ones, suggesting that A and B genome genes from the hexaploid had a deleterious impact on performance of the tetraploid RIL. All tetraploid RIL yielded significantly less than the tetraploid parent. Conversely, genes from the tetraploid were not deleterious to the hexaploid RILs. In fact, several hexaploid RIL yielded significantly more than the hexaploid parent Choteau. These results suggest that favorable genes from tetraploid wheat will be identified for improvement of hexaploid spring wheat.

The variation in stem solidness scores show segregation at both ploidy levels for a major gene that originated from the hexaploid variety Choteau. This is an indication of recombination, and shows that the gene is expressed similarly at both ploidy levels. Seed harvested from the 2011 experiment has been used to plant larger trials at two Montana sites for 2012. The population will be genotyped with the 90,000 SNP array this year.

Table 3. Grain yield and stem solidness scores for the parents and RIL developed from an hexaploid/tetraploid wheat cross.

Genotype	Grain Yield (g/plot)	Stem Solidness (5-25, where 25 is solid)
Choteau (6X parent)	350	25
6X RIL Mean (n = 134)	258	16
6X RIL range	81-469	7-24
Mountrail (4X parent)	364	11
4X RIL Mean (n=96)	188	18
4X RIL Range	91-310	7-24

D3.12. *Evaluation of wheat chromosomal translocation lines for high temperature tolerance*: *Dasypyrum villosum* and *Aegilops geniculata* Roth are diploid wild relatives of bread wheat that have many agronomically important genes for wheat improvement. The objective of this research was to evaluate selected chromosomal translocation lines for high temperature (HT) tolerance. Sixteen wheat chromosomal translocation lines and four bread wheat varieties were grown at optimum temperature (OT) of 22/14°C day/night and full irrigation in a greenhouse in Kansas. Ten days after anthesis half of the plants were exposed to HT stress of 34/26°C, and the other half remained at OT. The stress period was for 16 d; plants were then returned back to OT. High temperature decreased leaf chlorophyll content by 22%, maximum quantum yield of photosystem II (Fv/Fm) by 39%, individual grain weight by 44% and grain yield by (45%), when averaged across the genotypes. There was genetic variability among chromosomal translocation lines for yield traits. The tolerant genotype TA 5608, identified in this study can be utilized in improving high temperature tolerance of bread wheat cultivars.

D3.13. *Evaluation of Asian spring wheat lines for high temperature tolerance***:** Kansas State University evaluated heat tolerance of 161 Asian Spring wheat lines and 6 checks. These Asian lines were mainly from China and Japan, and the checks are Excalibur,

Krichauff, Halberd, Kukri, Len and Siete Cerros. Phenotypic data were collected from plants grown in a controlled environment. Plants of each line are grown in non-stress condition in growth chambers. At the post-anthesis stage, 50% of the plants from each line were used for heat treatment. Heat tolerance-related traits including chlorophyll degradation rate and grain yield were measured. The results showed that heat stress decreased grain yield by 42% when averaged across all genotypes. Average heat susceptible index (HSI) of all genotypes is 0.937. Seven Asian Spring lines produced greater grain yields in both heat stress and non-stress conditions, and showed lower HSI than the heat tolerant checks. These experiments are being repeated this year, and the best lines will be transferred to the breeding programs.

D3.14. *Genetic variability for root traits in a sample of the world collection*: A study was conducted at Kansas State to quantify genetic variability for root traits in 296 spring wheat genotypes from the Washington State University (WSU) World Wheat Collection. Single plants of all genotypes were grown in 1.5 m columns in a green house. Data on plant height, number of tillers, and rooting depth are currently being analyzed. Roots of selected genotypes with maximum, minimum, and intermediate rooting depth were subjected to complete image analysis (WinRHIZO software) for quantifying traits such as total length, surface area, and volume of total root mass and of roots of various size classes. Partial results shown in Table 4 indicate that considerable genetic variability exists in WSU World Wheat Collection for root traits that can be exploited to improve drought tolerance and/or resource capture in wheat.

Table 4: Root traits of selected wheat genotypes from the WSU World Wheat Collection

Genotypes	Depth (cm)	Total length (cm)	Total surface area (cm^2)	Total volume (cm^3)	Traits of fine roots (diameter, 0-0.25 mm)		
					Length (cm)	Surface area (cm^2)	Volume (cm^3)
KSG 233	155^{ab}	7112^{a}	1126^{a}	14^{ab}	3063^{a}	158^{a}	0.72^{a}
KSG 220	166^{a}	6066^{ab}	923^{abc}	11^{abcd}	2670^{ab}	137^{ab}	0.62^{ab}
KSG 176	46^{c}	1009^{gh}	115^{ij}	1.1^{g}	560^{ghi}	30^{hij}	0.14^{gh}
KSG 31	50^{c}	1009^{gh}	105^{ij}	0.9^{g}	595^{ghi}	30^{hij}	0.13^{gh}

D.4. Phenotyping for Nitrogen Use Efficiency (NUE)

- ***Deliverable: barley populations evaluated for NUE***

D.4.1. *Barley*: NUE is being evaluated using the spring six-row (SP6), spring two-row (SP2), and winter six-row (WN6) association mapping panels in a low (70%) nitrogen and normal (100%) nitrogen environment. All of the field trials are using a Type II modified augmented block design with entries replicated once and 3-5 checks replicated multiple times depending on the dimensions of the field. Randomizations were generated centrally at OSU and distributed to the collaborators. Seed was distributed by UMN.

In 2011, the SP6 and SP2 AM panels were evaluated in Crookston, MN under the direction of Kevin Smith and in Bozeman, MT under the direction of Tom Blake. The trials were harvested and all agronomic data was collected. CSR data was collected only at the MT location. The experiment and line list files have been created and uploaded to T3. Agronomic data is being adjusted per the experimental design and is scheduled to be uploaded to T3 by the end of May. We are still working on uploading CSR data.

In 2012, the SP6 AM panel is scheduled for Crookston, MN (Kevin Smith), Prosper, ND (Richard Horsley), and Pullman, WA (Kevin Murphy). At the time of writing this report, the MN location was planted and the ND and WA locations were planning to plant the week of May 14. The SP2 AM panel is scheduled for Bozeman, MT (Tom Blake), Pullman, WA (Kevin Murphy), and Aberdeen, ID (Gongshe Hu). At the time of writing this report, the MT and ID locations were planted and the ID location was scheduled for the week of May 14. The WN6 AM panel was planted last fall in Crookston, MN, Corvallis OR, and Logan UT. The MN location was lost to complete winterkill of the plots. The OR location (Corvallis) was planted Oct. 18, 2011. There was complete winter survival. Based on soil tests, N was applied in fall and spring. CSR measurements are in progress. The UT location was planted Oct 17, 2011. Winter survival and heading notes have been taken and CSR measurements will begin the week of May 21.

- ***Deliverable: wheat populations evaluated for NUE***

D.4.2. *Hard Winter Wheat Elite panel*: The field experiment for NUE and yield evaluation was planted in the fall of 2011 at Tipton OK and Ithaca NE. This panel is also being used for WUE and irrigated, normal N trials were also established for that purpose in KS and CO. Thus, there are two locations for NUE and four for yield. Stands were very good in NE. The OK location narrowly missed an F4 tornado in the fall and stands of perhaps 10% of the plots were damaged at that time. Pesticides have been applied as needed and as prophylactic treatments.

The NE and OK sites were planted in a split plot design for moderate N and low N treatment. N levels were determined based on yield goals and soil N for each location. An equal N amount was applied in the fall at planting and the differential N rates were applied in the spring. Above-ground tissue samples currently are being collected from two 30 cm sections of row from each plot as plots reach anthesis. Dried tissue samples will be analyzed for N and mineral concentration. The complete panel has been submitted for genotyping to Eduard Akhunov (KS) and line information has been uploaded to the T3 database.

D.4.3. *Soft Winter Wheat Elite Panel*: This panel was planted for yield and NUE evaluation in the fall of 2011 in Wooster OH, and Warsaw VA. The panel was also planted for yield using normal N rates in Columbia, MO, KY, and MD. Spring stands appear adequate at all locations. Pesticides have been applied as needed and as prophylactic treatments at all locations. The OH and VA sites were planted in a split plot design so for high N and low N treatment. N levels were determined based on yield goals and soil N for each location. An equal N amount was applied in the fall at planting and the differential N rates were applied in the spring. Dried tissue samples will be collected and analyzed for N and mineral concentration. The complete panel has been submitted

for genotyping to Eduard Akhunov (KS) and line information has been uploaded to the T3 database.

Most program locations (OK, MO, KY, OH, MD, and VA) are collecting CSR readings using the JAZ spectrophotometer. NE is collaborating with the Center for Agricultural Land Management and Information Technology (CALMIT) and the possibility of aerial measurements (see section D1 above). OK and OH collected data at flag leaf and boot stages, VA at Zadoks 31, 41, and 51. In OK at anthesis, whole plant samples were collected from each plot to document biomass and nutrient uptake. VA has conducted an additional field experiment with five soft red winter wheat and 16 sampling patterns as treatments. CSR data was collected 50 cm above the canopy level in eight treatments and at 100 cm for the other eight. Spectral readings are being collected at Zadoks growth stages 35, 45, 55, and 75.

D.5. Phenotyping for disease resistance

- ***Deliverable: Barley NSGC core evaluated for multiple diseases***

D.5.1. S*tem rust resistance*: Brian Steffenson (UMN) conducted the stem rust disease screens. The first half of the barley NSGC core (1,050 accessions) was sent to Greytown, South Africa for adult plant phenotyping to African stem rust. These lines were sown in mid-June and inoculated three times with stem rust. Disease severity assessments were made in mid-November using the modified Cobb scale (0-100% scale), but rust infection was low and not evenly distributed in the nursery, making the data of little value for association mapping. The first half of the barley core collection has now been shipped to Njoro, Kenya for the main season planting in June 2012. Disease assessments will be taken in mid-October and will be uploaded to the T3 website by January 2013.

D.5.2. *Stripe rust resistance*: Patrick Hayes (Oregon State University) is conducting the stripe rust screening of the barley NSGC core. The entire barley core collection (2,062 entries) was planted at the Hyslop farm in Corvallis October 2011. The experiment was arranged in a modified augmented design (type 2) for rectangular plots. All of the accessions survived the winter and were thriving in the early spring. By chance, an epidemic of leaf scald caused by *Rhynchosporium secalis* occurred in the nursery, so notes (1-9 scale) on this disease were recorded. Stripe rust is now developing rapidly in the nursery and severity notes (0-100%) will be taken on accessions three to four times during the season. Data for stripe rust, leaf scald and heading date will be uploaded into T3 by August 2012.

D.5.3. *Spot blotch resistance*: Shaobin Zhong (NDSU) conducted the spot blotch screening of the barley NSGC core. The second half set of the NSGC Barley Core Collection (total of 1,012 accessions) was received in January 2012. Seedling reactions to the spot blotch pathogen (*Cochliobolus sativus*) were evaluated in the greenhouse from March to May 2012 using the highly virulent isolate ND4008. Three experiments (replicates) were conducted. For each experiment, three plants from each accession were planted in cones. Assessments for disease were made according to the 1-9 scale developed by Fetch and Steffenson (1999), with 1 being most resistant and 9 being most susceptible. Susceptible controls showed readings ranging from 6-8, while moderately

resistant controls had readings ranging from 4-5. Among the 1,012 accessions tested, 17 (1.7%) were resistant with readings ranging from 2-4 and 187 (18.5%) showed moderate resistance with readings ranging from 4-5. The remaining accessions (808/79.8%) were susceptible with readings from 6-9. Current data will be uploaded into T3 by July 2012.

D.5.4. *Spot form net blotch resistance*: Robert Brueggeman (NDSU) and Timothy Friesen (USDA-ARS) conducted the spot form net blotch disease screens. In 2011, adult plant evaluations for spot form net blotch were confounded by other diseases in the field; therefore seedling assays will now be conducted. Seedling screening for resistance to the spot form net blotch pathogen (*Pyrenophora teres* f. *maculata*) was completed on the first half of the barley core collection (1,050 accessions). Phenotyping was completed on the 1,050 line set with a highly virulent local North Dakota isolate (FGOB10Ptm1). Lines were scored on a 1-5 reaction type scale with 1 being highly resistant (small dark pinpoint necrotic lesions), 2 moderately resistant (pinpoint lesions with small amounts of necrosis and chlorosis surrounding the penetration point), 3 moderately susceptible (necrotic or chlorotic lesions 2-3 mm in size with little coalescence of lesions), 4 susceptible (coalescing necrotic or chlorotic lesions >3 mm across), and 5 highly susceptible (necrotic or chlorotic lesions coalescing and covering greater than 70% of the leaf area). Barley lines showing an average disease reaction of 2 or less (81) to isolate FGOB10Ptm1 were evaluated with three additional isolates including SG1 (from Australia), NZKF2 (New Zealand), and Den2.6 (Denmark). Of the 81 lines that showed good levels of resistance (≤2) to ND isolate FGOB10Ptm1, only nine lines showed the same level of resistance to the other three isolates indicating high levels of diversity in both resistance and virulence. Currently, the second half of the barley NSGC is being phenotyped in the same manner to identify additional barley accessions with the highest potential for durable resistance. Using single seed descent, we developed genetically clean stocks of several of the resistance sources for use in future crosses. The crossing block has been established and TCAP students will assist in the development of genetic materials for resistance characterization during the 2012 field season. Current data will be collated for uploading to the T3 website by June 2012.

- ***Deliverable: wheat NSGC core collection evaluated for multiple diseases***

D.5.5. *Analysis of the NSGC wheat core 2011 data*: Preliminary association mapping analyses have been conducted using the leaf, stem, and stripe rust data collected in 2011, on 1000 spring wheat lines from the NSGC core collection. These lines have been genotyped with the iSelect 9000 SNP wheat chip. In total, ~35 resistance loci that are significant after accounting for experiment-wide error rates have been detected. Briefly, significant loci for seedling resistance to wheat stem rust races were identified: BCCBC (7 SNP markers, likely representing 3 loci), TTTTF (24 SNP markers, likely representing 6 loci), a bulk of North American isolates (33 SNP markers, likely representing 4 or more loci), TTKSK (49 SNP markers, likely representing 4 or more loci), and TRTT (9 SNP markers, likely representing 3 loci). Association mapping with data from stem rust evaluation of adult plants in the field in Minnesota resulted in the identification of 2 SNP markers that represent a single locus. Association mapping of leaf rust resistance in the seedling stage with Race 1 resulted in the identification of 2 loci (4 SNP markers). Surprisingly, seedling testing with a North American bulk of isolates and adult plant

phenotyping of leaf rust in the field did not identify any significant loci. Association mapping of stripe rust reaction in field nurseries in CA and WA (2 locations) under conditions of natural infection resulted in the identification 14 significant resistance loci linked to ~30 SNP markers.

D.5.6. *Selection of promising lines from the NSGC wheat core collection*: From the first 1,000 NSGC spring wheat core accessions evaluated in 2011, 29 were selected that showed low (≤10%) severity to leaf rust. The 29 lines were screened with 10 leaf rust races to postulate major leaf rust resistance genes and were crossed to the susceptible line Thatcher to develop bi-parental mapping populations. The 29 lines were also screened for adult plant resistance in the greenhouse using a mixture of races. DNA was collected from each line and screened with SSR markers for nine known leaf rust resistance genes (*Lr16, Lr19, Lr21*, *Lr24, Lr26, Lr32, Lr34, Lr37,* and *Lr46*). Fifteen of the 29 crosses were selected based on adult plant, seedling, and marker screenings to be planted in the field in 2012 as F_2's for advancement. Ten populations will be evaluated in the fall for seedling resistance and five will be targeted for adult plant resistance and advanced by the SSD.

From the same 1,000 NSGC core lines, 30 were selected based on their resistance to stem rust, and other agronomic traits and were crossed with susceptible parent LMPG-6 in fall 2011. Seedlings of all lines were screened with 10 North American stem rust races, 3 Ug99 races, and one Yemeni and one Pakistani race. These selected lines were also screened with available SSR markers for 17 *Sr* genes. Based on the information gathered from seedling screening, marker screening and their parental pedigrees, we identified four lines as potential new sources of resistance to North American stem rust races, two lines resistant to Ug99 races and 2 lines resistant to Yemeni and Pakistani races. Bulk segregant analysis will be carried out on $F_{2:3}$ families of all 8 populations in fall 2012. Also, RIL populations will be generated from 15 F_2 populations with potential novel APR genes. These 15 F_2 populations were planted in St Paul in summer 2012 and subsequent generations will be advanced via SSD. After additional evaluation, one or two RIL populations (F_6 or further inbred) will be selected for extensive phenotyping and genotyping.

Similarly, development of validation and mapping populations is underway for stripe rust resistance loci identified by association mapping results obtained in 2011. Depending on polymorphism at specific SNP loci, new resistance donors are being crossed to Avocet S, or another susceptible parent. Loci on chromosomes 1BS, 2BS, 2BL, 3BS, 4BL, 4DL, 5AL, 6BS, 6D, and 7B were initially selected.

D.5.7. *2012 phenotypic evaluations of the NSGC collection*: Additional phenotypic evaluation is currently underway on the complete NSGC Core. All 1,417 winter wheat core accessions were evaluated for seedling leaf rust resistance with two different races and are currently being evaluated for seedling stem rust resistance with three races. These lines were recently evaluated in the field in TX and KS (3 locations) and as adult plants in a greenhouse test for adult plant leaf rust resistance. The lines will be evaluated for field stem rust resistance in MN this summer, and are also being evaluated at two locations in WA for stripe rust resistance. All 1,000 spring wheat accessions evaluated in 2011 are being re-evaluated in 2012 in CA and WA (2 locations) for stripe rust resistance in the field. In addition, the final 2000 spring wheat core accessions are currently being

screened for leaf, stem and stripe rust under field conditions in MN (2 locations), CA and WA (2 locations). Seedling leaf, stem, and stripe rust screening on the complete core collection will be completed by the end of 2012.

D.5.8. *Evaluation of Winter Wheat Association Mapping panels*: Approximately 310 hard winter wheat lines from the Winter Wheat Association Mapping Panel were evaluated for leaf rust resistance at the adult stage in the greenhouse, and in Castroville, TX, Hutchinson, KS, and Manhattan, KS in 2012. These data will be added to data collected in 2011 in Manhattan, KS. The panel has been genotyped with the iSelect 9,000 SNP wheat chip and data will be analyzed to identify resistance loci for leaf rust.

Approximately 160 additional elite breeding lines from another winter wheat panel were phenotyped for leaf rust resistance in the greenhouse, Castroville, and Manhattan, KS. These lines have also been phenotyped and will be used to augment the analysis of the main AM panel. The Winter Wheat AM panel was phenotyped for resistance to the new race of stripe rust identified at Hays and Hutchinson in 2012. These data will be added to data from 2010-2011 and analyzed to identify important loci for resistance. A manuscript is in preparation.

D.5.9. *Lakin/Heyne RIL mapping population*: this population was analyzed for resistance to stripe rust and QTL were found on 2AS (*Yr17*), 2BS, and 1A in Heyne as well as a QTL on 3A for Lakin. A manuscript is in preparation.

D.5.10. *KS05HW14*2/Kingbird*: A BC_1F_5 population of KS05HW14*2/Kingbird was scored for resistance to a new race of stripe rust at Manhattan, KS. The resistance of KS05HW14 was substantially defeated by the new race, but resistance derived from Kingbird segregated in the progeny. This population will be genotyped in the coming year. This population was also scored for resistance to the QFCSC race of stem rust at Manhattan, KS. KS05HW14 was partially resistant to QFCSC, possibly due to *Sr7b*. Resistance derived from Kingbird segregated in the progeny as well. Additional greenhouse testing with more virulent races is planned.

D.5.11. *Spring wheat stripe rust resistance AM panel*: this panel, which includes 510 entries, is currently being evaluated in field nurseries in WA. This panel is a collection of various sources of resistance identified over the past 30+ years of germplasm evaluation at Washington State University. DNA of this panel has been prepared, and it will be genotyped with SNP to identify new sources of resistance. Avocet S was crossed to 70 spring wheat accessions with unknown resistance to stripe rust. F_2 and BC_1F_1 populations for all accessions are currently being advanced in 2012 field nurseries in WA.

D.5.12. *Winter wheat stripe rust AM panel*: this panel, which includes 486 entries, was planted in the field in fall 2011 for stripe rust evaluation at two WA locations in 2012. Stripe rust has developed heavily at Mt. Vernon, WA, and multiple ratings have already been taken. This panel will be genotyped with the iSelect 90K SNP wheat chip in 2012/2013 for association mapping.

D.5.13. *Yr48 high density mapping*: TCAP student Josh Hegarty developed a high density map of *Yr48* that placed this gene 0.03 cM distal to AK336105 and completely linked to markers *cfa2149* and *gpw2149*. A physical map has been initiated and the first chromosome walk step has been completed orienting the selected BACs relative to the

gene. Additional 3000 segregating lines have been phenotyped and flanking markers are being run in the genotyping laboratories to further reduced the target region. An EMS mutagenized population derived from a plant carrying only *Yr48* was grown in the field and 5 putative susceptible mutants have been identified and will be crossed to validate segregation.

D.5.14. *YrLouise high density map*: this locus on chromosome 2BS is a strong and consistent source of adult plant stripe rust resistance. To develop diagnostic markers for this source of resistance, two populations of 1,536 progeny each have been advanced (Avocet S*2/Louise and Penawawa*3/Louise). Backcrossed-derived plants heterozygous for the QTL region were identified and self-pollinated, and over 3,000 F_3 families produced. We have completed SNP-based genotyping of 1,536 Avocet S*2/Louise F2's, and detected ~170 recombinants in the QTL region. These 170 recombinant families along with 1,536 Penawawa*3/Louise BC_2F_3's are currently growing in a field stripe rust screening nursery. Homozygous recombinants will be selected while phenotyping for adult plant stripe rust resistance this summer. A high-density SNP-based map is in progress.

E. POPULATION DEVELOPMENT

- ***Deliverable: Advanced barley spring NAM populations***

E.1. *Barley Six-row NAM population*: Kevin Smith (UMN) is developing the 6-row NAM population. A total of 97 crosses were made between the NSGC parents and Rasmusson. The F_1's from these crosses were planted in January in the greenhouse and backcrosses were made to Rasmusson. In addition, nine NAM parents were replanted to cross to Rasmusson that were missed in the fall. The target is to produce 15 BC_1 and 85 F_2's from each of the 100 NAM crosses. We are still assessing the number of successful crosses and backcrosses.

E.2. *Barley two-row NAM population*: Rich Horsley (NDSU) is developing the 2-row NAM population. A total of 128 crosses were made between the NSGRC parents and Conlon. The F_1's from these crosses were planted in January in the greenhouse and backcrosses were made to Conlon. At the time of writing this report, backcrosses are still being made. Presently, we have obtained 15 or more seeds from backcrosses to over 80 of the F_1 lines. All of the BC_1 and F_2 seed produced this spring from both NAM populations will be advanced by single seed decent in the Fall of 2012 in the greenhouse.

- ***Deliverable: Advanced wheat NAM populations***

E.3. *Soft and hard winter wheat NAM populations*: Thirty F_1 hybrids between accessions from the NSGC Core collection and 'Overland' have been made in the greenhouse to initiate the Hard Winter Wheat NAM population. Thirty six F_1 hybrids between accessions from the NSGC Core collection and 'Branson' have been made in the greenhouse to initiate the Soft Winter Wheat NAM population. Crosses toward development of an NUE male-sterile facilitated Hard Winter Wheat recurrent selection population utilizing the NAM donor parents also have been made.

E.4. *Spring wheat NAM populations*: A total of 38 land-race accessions from around the world and 15 elite spring wheat lines from Montana, California, CIMMYT and Australia

were crossed to the common parent Berkut. From initial populations of about 800 F_2 individuals for each landrace cross, approximately 300 F_4 individuals with semidwarf growth habit have been selected. These are currently being grown in a growth chamber under 12 hour days to select for photoperiod insensitivity. The goal is to develop approximately 100 semidwarf photoperiod insensitive RILs per cross. These will have suitable agronomic properties for evaluation for WUE throughout our growing region beginning in 2013. The seven elite populations from California are one generation behind and are being advanced at Tulelake as F_3 progenies. Those populations will be ready for evaluation in 2014.

- ***Deliverable: Completed wild barley introgression population***

E.5. *Wild barley introgression population*: TCAP graduate student Liana Nice (UMN) has developed a wild barley introgression population consisting of 25 wild barleys that represent 90% of the diversity in the 318 individuals of the wild barley diversity collection. These 25 wild barleys have been backcrossed twice to the elite six-row malting barley Rasmusson. An average of 32 BC_2 individuals have been derived from each of the 25 wild barleys for a total of 803 lines. Four generations of single seed descent have been conducted to derive inbred lines. A customized 384 SNP VeraCode assay has been designed and is currently being used to genotype the population.

798 individuals of the population are being grown in Crookston and St. Paul, MN in a type II augmented design. There are 60 entries of the primary check (Rasmusson) and 16 secondary checks (Harrington and Conlon, late and early heading, respectively) randomly arranged in the field as well as 26 repeated line entries to fill in the field design. To permit maturity-appropriate harvesting, the field was blocked using heading date (early, mid, late, and unknown) data collected from head rows planted in 2011. This is necessary due to the wide heading date window (over three weeks) of the diverse population. Multiple traits will be scored on the population including: height, heading date, yield, productive tiller number, grain protein, and spike phenotypes (seed/hull color, awn texture, shattering, nodes per spike, etc.).

F. EDUCATION

The education activities for the first quarter of 2012 are summarized by **goal** and *output*.

- ***Deliverable: Integrated Education and Research programs***

F.1. *PAG Planning meeting:* The TCAP held its annual meeting in San Diego, CA on January 15. TCAP co-PIs, students, stakeholders, the scientific advisory board and USDA administrators attended the meeting (~100 participants). The morning was devoted to reporting progress and the afternoon was devoted to planning future activities. Jamie Sherman (Montana State University) provided an overview of the progress achieved in the different education objectives. Then S. Chao (USDA-ARS, Fargo, ND), Gina Brown-Guedira (USDA-ARS, Raleigh, NC) and Eduard Akhunov (Kansas State University) updated the group on the genotyping efforts. Eduard Akhunov also discussed new developments in wheat and barley gene capture and genotyping by sequencing (GBS) efforts. There were short updates on the phenotyping efforts on water use

efficiency by Luther Talbert (Montana State University) and Pat Hayes (Oregon State University), on nitrogen use efficiency by Kevin Smith (University of Minnesota), and Clay Sneller (The Ohio State University), and on diseases by Brian Steffenson (University of Minnesota) and Mike Pumphrey (Washington State University). Jianli Chen (University of Idaho) updated the group on the evaluation of the wheat NSGC core for water and nitrogen use efficiency. Kevin Smith and Luther Talbert presented progress of developing nested association mapping populations in barley and wheat, respectively. Tyson Howell (UC, Davis, graduate student in the Dubcovsky lab) gave a summary of his characterization of drought resistance derived from the wheat 1RS:1BL translocations using canopy spectral reflectance. The afternoon portion of the meeting was spent planning for the coming year. The meeting ended with comments and suggestions from the scientific advisory board.

F.2. *PAG Graduate Student meeting*: To help build a graduate student community, 19 graduate students, six faculty (including a MSI representative) and an industry representative met at the PAG meetings. To begin working on industry's request that we improve all plant breeding human capital, we brainstormed a list of skills needed by professionals. Next the group broke into small groups and the students discussed their specific research topics. Graduate students expressed an interest in organizing the meeting next year.

F.3. *Newsletters*: The winter 2012 Newsletter was produced and distributed among TCAP participants, stakeholders and members of the scientific advisory board *http://passel.unl.edu/Image/CrowDeAnna1129929130/Winter%202012%20Newsletter.pdf*. Stakeholder comments about the first newsletter were integrated into a more stakeholder friendly format for the winter newsletter. The release of the 2012 spring newsletter is planned for the end of May.

F.4. *TCAP seminar series*: To encourage communication about the TCAP project a seminar series was held online. Six seminars were presented by TCAP faculty, students and collaborating faculty from minority serving institutions (Table 5).

Table 5. T-CAP seminars

Presenter	Title	Date	Attendance
S. Baenzinger	Understanding Grain Yield: It's a journey, not a destination	2/23/12	47
D. Namuth-Covert	Cyberspace: New Frontiers in Learning and Networking	3/15/12	15
E. Akhunov	Usage of genome wide genotyping approaches to understand the genetics of agronomically important traits in wheat.	3/29/12	41
J. Jannink, V. Blake, C. Birkett & D. Matthews	Introduction to T3	4/12/12	31
T. Howell and J. Dubcovsky	TCAP Canopy Spectral Reflectance updated methods for 2012	4/26/12	30
M. Matute	Plant Parasitic Nematodes- The Farmer's Hidden Enemy	5/3/12	16

Participants view the seminar series as an important tool to ensure the success of the project. All the seminars were archived at http://passel.unl.edu/communities/pbtn. The

seminars facilitated the implementation of new tools e.g. T3 database or CSR by sharing on real time problems and solutions among the participating groups. Graduate students are designing the TCAP seminar series for fall.

F.5. *Improvements of online environment*: The online environment is being used as a communication tool for project management as well as to provide educational opportunities through classes and seminars. Other groups are using the site for communication e.g. Ag*Idea Plant Breeding Program design committee (meets monthly), UNL Graduate Student Plant Breeding Symposium (80+ attendees) and NAPB Student committee (meets monthly to plan student activities at NAPB). In collaboration with NSF project (Namuth-Covert), computer programming was added to generate reports of how often materials, communities, etc. are being accessed (Table 6).

Table 6. Usage statistics of educational materials.

	Dates	Total visits	Aver. Time on site	Pages/visit
Entering Mentoring (Spring class)	2/1/12-4/30/12	35	19 m 10 s	4
PBTN (Entry into all offerings)	2/1/12-4/30/12	527	5 m 9 s	3
	9/1/11-11/30/11	758	5 m 37 s	4
PB Grad Student community (available for use fall and spring)	2/1/12-4/30/12	35	19 m 10 s	4
	9/1/11-11/30/11	3	3 m. 43 s	16
Plant Breeding Strategies(Fall class)	9/1/11-11/30/11	124	10 m 6 s	6
Q genetics (Spring)	2/1/12-4/30/12	95	9 m 36 s	37
TCAP Graduate courses (lists offerings)	2/1/12-4/30/12	24	8 m 43 s	11

Several improvements have been implemented this quarter. An auto-generated members list was created so each community now has a tab, with lists of members who wish their profiles to be public. We hope this will help people feel more comfortable in contacting each other for collaborations.

A major overhaul to improve the quizzing system is underway. Quizzing will be more user friendly for instructors and allow for more types of questions (matching, short answer, fill in the blank, etc.) than the current multiple-choice format. We are currently looking at either programming our own code or adopting code from Moodle. We are outlining the key features we want in quizzing and grade book. If we adopt Moodle, we will also use Moodle's discussion forum platform, which appears to be more user friendly than the one we have in place.

We are researching the best option for long term storage of large video files, such as narrated power points and archived webinars. Their current location is a temporary space at UNL. We are also researching the possibility of a mirrored site at Montana State to

serve as back-up in case of a failure of the UNL servers and backup, or to also help with traffic.

- ***Deliverable: Recruit a diverse group of undergraduates to plant breeding***

F.6. *Education/recruitment films*: The TCAP film, "Holding the future in the palm of your hand", was finished and posted at http://vimeo.com/43017010. The film was used successfully in recruitment at University of Arkansas Pine Bluff, Rust College and Chicago State University to about 200 students. A second film on Plant breeding for disease and pest resistance and a short film about the wheat stem sawfly will be completed this summer.

F.7. *Brochure*: A recruitment brochure as a companion to the film was created and 200 copies were distributed in spring 2012 during recruitment trips. It has also been posted at http://passel.unl.edu/communities/pbtn. Participants are encouraged to print and use in recruitment efforts.

F.8. *MSI / TCAP bridge*: Eight MSI faculty/TCAP faculty collaborations were established. Fifteen undergraduates have been engaged in research with these eight faculty. Each MSI faculty will report on first year progress by end of May. Renewal of Support decisions will be made by end of June.

F.9. *Increase minority student interest and awareness*: Recruitment efforts so far have raised minority student interest. Ten students have contacted Sherman with interest in internships. Two minority students from University of Arkansas, Pine Bluff (UAPB) have applied to graduate school and 1 student from Woodland Community College has begun internship in drought tolerance at UC Davis. A short-term research experience at a TCAP institution for students from collaborating MSIs will be implemented this summer. Four minority students from UAPB will travel to Washington State University for a two month research experience in coPI Arron Carter's laboratory. Students will conduct research related to disease and drought tolerance in wheat, participate in field days and visit additional labs. The TCAP will contribute to travel and housing costs. In addition to providing research experience in plant improvement for undergraduate students, it will serve as a pilot for collaborative research/training activities between TCAP and minority serving institutions.

- **Deliverable: support faculty in innovative teaching methods**

F.10. *Development of educational materials*: Nitrogen Use Efficiency Lesson is in a third draft stage and under peer review. Discussion of additional inquiry-based activities connected to nitrogen use and nitrogen use efficiency in crop plants are in progress. Activities and materials that can be used in class and online are being planned.

A storyboard is being developed for an interactive quantitative traits lesson. The lesson will utilize actual data from the Oregon Wolfe Barley.

The Wheat Breeding Activity developed as a year one deliverable was submitted to the Journal of Natural Resource and Life Science Education. *Introduction to Plant Breeding Learning Activity: Wheat.* Amy Kohmetscher, Don Lee, and Deana Namuth-Covert. Available at: http://passel.unl.edu/animation/Wheat_Breeding2/Wheat%20Breeding%20Activity%202.swf

F.11. *Teaching workshop*: Planning for an educational workshop on the use of inquiry-based approaches is in progress. The purpose of the workshop will be to create a knowledge base, generate ideas, encourage collaboration and initiate development of inquiry-based activities for use in plant science courses at the undergraduate and graduate levels. Plans are to provide travel stipends for educators from TCAP and collaborating MSIs to participate in the 2-day workshop.

- **Deliverable: trained new plant breeders**

F.12. *Graduate students training*: A total of 59 graduate students have participated in the plant breeding training network. Of these, 33 graduate students are fully or partially funded by TCAP and involved in field and lab research with TCAP PIs. The remaining students are for the most part affiliated with a TCAP PI. However, several students have participated in TCAP classes and have no affiliation with TCAP. During 2012 PhD students supported from the project published three peer reviewed publications (two of them as first authors) and made eight presentations in scientific meetings and symposia, all of them as first authors.

F.13. *Graduate courses*: Three courses that have been offered through the TCAP are listed below. Courses currently in development will cover association mapping and developing plant breeding human capital. TCAP PIs have contributed lectures that were used in the graduate classes. These lectures are available for use online under the lecture tab at http://passel.unl.edu/communities/pbtn.

Table 7. Graduate course.

	Semester	Total Registered	Total completed
Plant Breeding Strategies (5 lectures)	Fall 2011	26	14
Entering Mentoring	Winter 2012	10	5
Q Genetics (7 lectures)	Spring 2012	33	6

Students completing Entering Mentoring and Quantitative Genetics received a certificate of completion. However, completion rates are below our goals likely because students are not getting official credit for these courses. We are continuing to develop the Plant Breeding Program through Ag*Idea so that interested students can receive credit for our offerings (see below).

F.14. *Undergraduate students training*: Twenty undergraduates are being mentored by TCAP faculty and graduate students. Five graduate students completed Entering Mentoring to support their mentoring efforts.

F.15. *Undergraduate online meetings*: There are currently 35 undergraduate students at TCAP institutions and MSIs who are involved in research related to plant improvement and are funded in full or in part by the TCAP. An online community was established to support these students through meetings in which students interact with plant breeding

scientists in academia and industry and discuss their own research experiences with other undergraduate students. Jamie Sherman met with students in January and provided background on the TCAP. Donn Cummings (Monsanto) discussed the future of biotechnology in plant improvement and careers in that field in March. Kevin Smith will discuss preparation for, and application to, graduate school in the area of plant sciences on May 9. Plans are to strengthen the community through asynchronous discussions and self-guided reflection and evaluation materials.

F.16. *Undergraduate student presentations*: Kelsey Salvo and Sarah Grogan. Characterizing glaucousness in wheat. Celebrate Undergraduate Research and Creativity Symposium, Poster 170. Colorado State University, April 17, 2012.

Mariam Kaleem and Yvonne Manning presented Initial soil profiling for plant-parasitic nematodes before a *Triticum aestivum* crop on March 7, 2012 at 25th Annual Student and Faculty Research Forum, University of Arkansas, Pine Bluff, Arkansas, winning Overall Winner for Best undergraduate presentation and First Place Award in Undergraduate Level in Scholarly Research in the Area of Biology.

Jasmine Gaston presented Potential plant-parasitic nematode constraint to profitable *Triticum aestivum* production on April 20, 2012 at Twentieth Annual Arkansas Space Grant Symposium, The Winthrop Rockefeller Institute, U of A System, Morrilton, Arkansas.

Ryan Graebner (our TCAP undergrad) and Araby Belcher (our TCAP grad) at Oregon State explain the principle of CSR at a field day held for "Leadership Corvallis".

F.17. *Sustainable online program*: Namuth-Covert, Baenziger, Sherman (Co-Chair) and others outside the TCAP project are meeting regularly to design a plant breeding program through AgIdea, an online course sharing consortium. AgIdea officials have accepted the letter of intent to create a Plant Breeding program and the group is working on a business plan. The goal is to have online course sharing available by Fall of 2013.

F.18. *Workshops and symposium*: TCAP is supporting student attendance this summer at two workshops and the National Association of Plant Breeders meeting. The first workshop is 'Plant Breeding for Drought Tolerance" (June 11-22), and is led by Pat Byrne at Colorado State University. The second workshop is "Rust Research Methodology" (July 8-10, 2012) at the University of Minnesota and is led by Brian Steffenson and Pablo D. Olivera. The 2012 NAPB annual meeting "Sustaining Life through Plant Improvement" will be held August 6-8 in Indianapolis, IN. Jamie Sherman will be an invited speaker discussing TCAP online education. A presentation about plant breeding was given to about 45 FFA students March 30th at the University of Nebraska Lincoln East Campus (Lee and Kohmetscher).

- ***Deliverable: an independent evaluation of the education activities***

F.19. *First year evaluation results*: a report was submitted by the Education Evaluation team in March 2012. The report presents findings and recommendations based on surveys of TCAP PIs, graduate students and collaborating PIs at minority serving institutions and from interviews with a sample of faculty and students at TCAP and minority serving institutions. The data provide a baseline of PI and graduate student perceptions of TCAP

educational activities, attitudes toward knowledge and skill areas involved in graduate student training, and characterization of professional networks. PI perceptions of obstacles to TCAP – MSI collaborative research efforts were also assessed. Evaluation of the education component of TCAP has provided data that is being used to inform decision making. It has revealed activities that are highly valued by TCAP faculty and graduate students, perceptions of initial educational activities, factors that are perceived to be important in building collaborative research between TCAP and minority serving institutions, factors that can enhance minority student success in science programs and the need for communication among all groups within TCAP.

G. PUBLICATIONS AND GERMPLASM RELEASES

G.1. Publications

Summary

2011. 25 peer reviewed publications were published in peer reviewed journals during the first year of the TCAP grant.

2012. 26 peer reviewed additional publications were published in the first five months of the second year of the TCAP grant

Total: 50 publications

List of new publications in 2012. Total 26, three of them by TCAP Ph.D. students

1.- Anderson, J.A., J.J. Wiersma, G.L. Linkert, J.A. Kolmer, Y. Jin, R. Dill-Macky, J.V. Wiersma, G.A. Hareland, and R. H. Busch. 2012. Registration of 'Tom' Wheat. J. Plant Registrations. 6: 2: 180-185

2.- Anderson, J.A., J.J. Wiersma, G.L. Linkert, J.A. Kolmer, Y. Jin, R. Dill-Macky, J.V. Wiersma, G.A. Hareland, and R. H. Busch. 2012. Registration of 'Sabin' Wheat. J. Plant Registrations. 6: 2: 174-179

3.- Baenziger, P.S., R. A. Graybosch, t. Regassa, L.A. Nelson, R. N. Klein, D. K. Santra, D.D. Baltensperger, L. Xu, S. N. Wegulo, Y. Jin, J. Kolmer, Ming-shun Chen, and Guihua Bai. 2012. Registration of 'NE01481' hard red winter wheat. J. Plant Reg. 6:49-53.

4.- Baenziger, P.S., R. A. Graybosch, T. Regassa, L.A. Nelson, R. N. Klein, D. K. Santra, D.D. Baltensperger, J. M. Krall, S. N. Wegulo, Y. Jin, J. Kolmer, Ming-shun Chen, and Guihua Bai. 2012. Registration of 'NI04421' hard red winter wheat. J. Plant Reg. 6:54-59.

5.- Bernardo A. N., H. Ma, D. Zhang, and G. Bai. 2012. Single Nucleotide Polymorphism in wheat chromosome region harboring *Fhb1* for Fusarium Head Blight resistance. Mol Breed. 29:477–488

6.- Chen, J. L. ; C. G. Chu; E. J. Souza; M. J. Guttieri; X.M. Chen; S. Xu; D. Hole; R. Zemetra. 2012. Genome-wide identification of QTL conferring high-temperature

adult-plant (HTAP) resistance to stripe rust (*Puccinia striiformis* f. sp. *tritici*) in wheat. Mol. Breeding 3:791-800.

7.- Edwards, J.T., R.M. Hunger, E.L. Smith, G.W. Horn, M.-S. Chen, L. Yan, G. Bai, R.L. Bowden, A.R. Klatt, P. Rayas-Duarte, R.A. Osburn, J.A. Kolmer, Y. Jin, D.R. Porter, K.L. Giles, B.W. Seabourn, M.B. Bayles, and B.F. Carver. 2012. 'Duster' wheat: A durable, dual-purpose cultivar adapted to the southern Great Plains of the USA. J. Plant Reg. 6:1-12.

8.- Heslot, N., H.-P. Yang, M.E. Sorrells, and J-L. Jannink. 2012. Genomic selection in plant breeding: A comparison of models. Crop Sci. 52:146-160.

9.- Leng, Y. and S. Zhong. 2012, Sfp-type 4′-phosphopantetheinyl transferase is required for lysine synthesis, tolerance to oxidative stress and virulence in the plant pathogenic fungus *Cochliobolus sativus*. Mol. Plant Path. 13: 375–387.

10.- Liu, Z.-H., Zhong, S., Edwards, M.C., and Friesen, T.L. 2012. Virulence profile and genetic structure of a North Dakota population of *Pyrenophora teres* f. *teres*, the causal agent of net form net blotch of barley. Phytopath. 102:539-546.

11.- Morrell, P.L., Buckler, E.S., Ross-Ibarra, J. 2012. Crop genomes: advances and applications. Nat. Rev. Genet. 13:85-96

12.- Naruoka, Y., J. D. Sherman, S. P. Lanning, N. K. Blake, J. M. Martin, and L. E. Talbert. 2012. Genetic analysis of long green leaf duration in spring wheat. Crop Sci. 52: 1: 99-109.

13.- Pradhan G.P., P.V.V. Prasad, A.K. Fritz, M.B. Kirkham, and B.S. Gill. 2012. Response of *Aegilops* species to drought stress during reproductive stages of development. Functional Plant Biology. 39:51-59.

14.- Pradhan G.P., P.V.V. Prasad, A.K. Fritz, M.B. Kirkham, and B.S. Gill. 2012. Effect of drought and high temperature stress on synthetic hexaploid wheat. Functional Plant Biology. 39:190-198.

15.- Pradhan G.P., P.V.V. Prasad, A.K. Fritz, M.B. Kirkham, and B.S. Gill. 2012. High temperature tolerance in *Aegilops* species and its potential transfer to wheat. Crop Sci. 52: 292-304.

16.- Poland J.A., P.J. Brown, M.E. Sorrells, and J.-L. Jannink. 2012. Development of high-density genetic maps for barley and wheat using a novel two-enzyme genotyping-by-sequencing approach. PloS ONE 7:e32253.

In press (10)

17.- Hazard B., X. Zhang, P. Colasuonno, C. Uauy, D.M. Beckles, and J. Dubcovsky. 2012. Induced mutations in the *Starch Branching Enzyme II* (*SBEII*) genes increase amylose and resistant starch content in pasta wheat Crop Sci. *In press*.

18.- Kumar S., S.K. Sehgal, U. Kumar, P.V.V. Prasad, A.K. Joshi, B.S. Gill. 2012. Genomic characterization of drought tolerance-related traits in spring wheat. Euphytica. *In press*. *doi:10.1007/s10681-012-0675-3*.

19.- Lorenz, A.J., K.P. Smith, and J.-L. Jannink. 2012. Potential and optimization of genomic selection for Fusarium head blight resistance in six-row barley. Crop Sci. *In Press*.

20.- Mengistu, N., P. S. Baenziger, K. M. Eskridge, I. Dweikat, S. N. Wegulo, K. S. Gill, and A. Mujeeb-Kazi. 2012. Validation of QTL for grain yield-related traits on wheat chromosome 3A using recombinant inbred chromosome lines. Crop Sci.: *In press*.

21.- Rutkoski, J., J. Benson, Y. Jia, G. Brown-Guedira, J.-L. Jannink, and M.E. Sorrells. 2012. Evaluation of genomic prediction methods for Fusarium head blight resistance in wheat. The Plant Genome. *In Press.*

22.- Tao Li, Guihua Bai, Shuangye Wu and Shiliang Gu. 2012. Quantitative trait loci for resistance to fusarium head blight in a Chinese wheat landrace Huangfangzhu. Euphytica. *In press. DOI 10.1007/s10681-012-0631-2.*

23.- Yu, L-X, A. Morgounov, R. Wanyera, M. Keser, S. Kumar Singh, and M.E. Sorrells. 2012. Identification of Ug99 stem rust resistance loci in winter wheat germplasm using genome-wide association analysis. Theor. Appl. Genet. *In press*.

24.- Zhang X.H., H.Y. Pan and G.H. Bai. 2012. Quantitative trait loci for fusarium head blight resistance in U.S. hard winter wheat cultivar 'Heyne'. Crop Sci. *In press*. doi: 10.2135/cropsci2011.08.0418

25.- Zhang X.H., H.Y. Pan and G.H. Bai. 2012. Quantitative trait loci responsible for fusarium head blight resistance in Chinese wheat landrace Baishanyuehuang. Theor. Appl. Genet. *In press DOI: 10.1007/s00122-012-1848-0*

26.- Hale I., X. Zhang, D. Fu, and J. Dubcovsky. 2012. Registration of wheat lines carrying the partial stripe rust resistance gene *Yr36* without the *Gpc-B1* high grain protein content allele. J. Plant Reg. *In press*.

G. 2. Varieties and germplasm releases

2012 Variety releases

1. UI Stone (IDO599) soft white spring wheat was release in March 2012. UI Stone has high yield under both irrigation and water limited conditions as well as excelleng end-use quality. UI Stone has good resistance to FHB and FHB1 gene based on molecular marker UMN10. Two additional lines are in the process of being released: IDO671 (SWS), IDO694 (HWS)
2. Hard red spring variety 'Rollag'. Jim Anderson, Jan. 2011, MN. Rollag has a unique combination of highest available resistance to Fusarium head blight and strong straw. Rollag contains the *Fhb1* QTL for Fusarium head blight resistance and *Lr34*.
3. Hard red spring variety 'Norden', Jim Anderson, Jan. 2012, MN. Norden is a competitive yielder with high test weight, good straw strength and leaf rust resistance not based on *Lr21*. Norden contains the *Fhb1* QTL for Fusarium head blight resistance and *Lr34*.

4. WB9879CLP is a two-gene Clearfield variety with resistance to imidazolinone herbicides developed by Montana State University and licensed to Westbred LLC for commercialization. The genes for herbicide resistance were backcrossed into the widely grown variety Choteau using marker-assisted selection. WB9879CLP is being grown in areas of Montana and North Dakota with wheat stem sawfly pressure.
5. Patwin-515 hard white spring wheat released by J. Dubcovsky (University of California, Davis) which includes stripe rust resistance genes *Yr5* and *Yr15*.

2012 Germplasm releases

1. Recombinant inbreed lines with *Yr36* but without the closely linked gene *GPC-B1* in both tetraploid (PI 656793) and hexaploid wheat (PI 664549). The hexaploid recombinant line is particularly useful for soft wheat breeding programs.