

Barley Coordinated Agricultural Project Work Plan FY06 (3/1/06 – 2/29/07)
Shiaoman Chao, USDA-ARS, Fargo, ND

1) Describe the research, education, and outreach activities you are planning for the next year (3/1/06 – 2/29/07)

Research : We will set up the Illumina's SNP genotyping system, and work with the Illumina's technical group to establish the SNP genotyping protocols. For genomic DNA extractions, we will try the Qiagen's kit, and, if necessary, automate the extraction process using the kit. The genotyping protocol will be optimized using the test samples before carrying out large-scale genotyping on 960 samples originated from the ten breeding programs.

Education : We will provide the training to students, postdocs or scientists who intend to use the Illumina system and the OPAs generated in the barley CAP for their own research.

Outreach : This will be done through the program set up by the project members at NDSU.

2) List specific outcomes and deliverables that will be accomplished in the first 6 months (3/1 – 8/31). These will be used as benchmarks for your bi-annual progress report.

- Install the Illumina genotyping system by late July, the target date.
- Establish the genomic DNA extraction protocol by June.
- Optimize the SNP genotyping protocol starting in August.

3) List specific outcomes and deliverables that will be accomplished in the second 6 months (9/1 – 2/29). These will be used as benchmarks for the bi-annual progress report.

- Continue to optimize the SNP genotyping protocol if necessary before the arrival of the breeders' samples in October.
- Carry out genotyping on 960 samples after October after samples are received.
- Store and archive data in the database setup in house.
- Send genotyping data to Jennifer Kling in early 2007.

Barley Coordinated Agricultural Project Biannual Progress Report

FY06 (4/1/06 – 9/30/06)

Shiaoman Chao (USDA-ARS, Fargo, ND)

1. Describe the research, education, and outreach activities you completed in the first half of the FY06 (4/1/06 – 9/30/06)

Research: The preliminary work was carried out to evaluate the tissue preparation format and genomic DNA extracted methods. Our lab routinely uses leaf samples prepared in 96 deep-well plate format for high throughput DNA extraction using robot to conduct wheat and barley MAS projects.

However, using the plate format, we can't rule out a low level of cross-contamination between wells, particularly the manual process required to mix well the buffers and solutions added during extraction. To avoid cross contamination for the barley CAP materials, we will collect leaf samples in individual tubes followed by freeze-drying and perform DNA extraction manually. For DNA extraction, the method our lab uses is SDS based. Tim Close advises the use of the Qiagen DNeasy kit for DNA extraction to obtain high quality SNP genotyping data. Thus, we will also explore the method provided by the Qiagen kit.

The Illumina SNP high throughput genotyping equipment has been purchased and shipped in late August, 2006. The equipment will be installed on October 23, and genotyping training is scheduled during the week of November 27 to December 1, 2006. Barley Pilot OPA1 will be used either during the training session or right after the training to assist in optimizing the genotyping techniques. To facilitate SNP genotyping optimization, 96 US barley core germplasm provided by Pat Hayes and Tim Close will be used. At least four of these germplasm have been previously genotyped with the Pilot OPA1 by Tim Close. Genomic DNA will be individually extracted using both the SDS-based method and the Qiagen kit. The results will allow us to cross-check and compare genotyping data generated in different labs, and to determine if both DNA extraction methods are equally adequate to generate high quality of genotyping data.

Education: We have a temporary intern to assist with the DNA extractions.

Outreach: I presented a talk to high school students at Maddock, ND (pop 500) via video conference, and described high throughput genotyping process and the advantage of MAS. A DVD was prepared showing the genotyping lab equipment and facility, and a brief demonstration on how the samples were prepared and the function of the equipment.

2. List specific outcomes and deliverables accomplished in the first half of FY06 (4/1 – 9/30).

- Establish tissue preparation and genomic DNA extraction protocols used for SNP genotyping.
- Prepare samples and lab setups ready for SNP genotyping training and optimization.