Using BSMV–VIGS for functional genomics in wheat.

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Our laboratory has gained significant experience using the barley stripe mosaic virus system for virus-induced gene silencing (BSMV-VIGS) as a tool for functional genomics in wheat. This presentation will summarize our experiences using the BSMV-VIGS system to functionally identify genes with essential roles in disease resistance pathways and, in particular, it will focus on the design considerations for successful VIGS experiments.

Development of resources for reverse-genetic analysis in Triticum monococcum.

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Reverse genetics is a powerful tool to discover gene function by identifying modifications in specific genes. Mutagenized populations are generated using either chemical, physical, or biological methods and screened for lesions for the desired gene of interest to identify its functional role. The goal of this project is to develop a mutagenized population of *Triticum monococcum* and screening systems for lesions in genes of interest using DNA pooling and PCR-based approaches as a reverse-genetic resource for the scientific community. We have generated mutagenized populations using 1,2,3,4-diepoxybutane (DEB) or trimethylpsoralen along with a UV treatment (TMP/UV) as a pilot study with chemical concentrations that lead to 20–25% survival rates. Experiments were conducted to identify the relative efficiency of these chemicals in a) creating mutations and b) detecting deletions/lesions using forward and reverse-genetic approaches.

We have generated approximately 1,000 M_2 families from each chemical treatment. Initial observations from five germinating seeds per M_1 plant indicated 2% albinos in 250 M_2 families per chemical treatment. DNA was isolated using a filter-based method from individual plants and, currently, is pooled to 1:8 and 1:16 times and used for screening using different methods. A total of 424 DEB-treated, M_2 families were advanced to the M_3 generation to observe visible mutant phenotypes and determine forward mutation frequency for this chemical treatment. Many phenotypic mutants, such as dwarf, early and late flowering, bushy, oligoculm, small spike, purple plants, and disease mimic were observed in this M_3 population. The percent of phenotypic mutants in the M_2 as observed in the M_3 families was 0.94% to 8.02 % for ten different phenotypes. The results of the screening for lesions in the mutagenized population will be presented, which will strengthen the use of this approach for developing reverse-genetic resources in wheat.

Poster 1. A detailed, comparative sequence analysis on the HMW-glutenin locus regions of eight genomes from diploid and polyploid wheats.

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Wheat is the most widely grown crop worldwide and feeds one-third of the world's population. As a result, wheat is foremost among the world's crops both in regards to its importance as a staple of mankind and its economic significance. Despite several successes in sequencing several plant genomes, the complex wheat genome might still represent challenges in genome-sequencing projects. Compared to most other cereals, bread wheat (*Triticum aestivum*) has an extremely large genome (~16,000 Mb); more than 30-fold greater than the rice genome. Furthermore, bread wheat is an allohexaploid species consisting of three related subgenomes (A, B, and D). To study the structural organization of wheat genomes, we sequenced large genomic regions harboring HMW-glutenin genes from eight Triticeae genomes including the D genome from diploid *Ae. tauschii*, the A^m genome from diploid *T. monococcum* subsp. *monococcum*, the A and B genomes from tetraploid *T. turgidum*, the A, B, and D genomes from hexaploid wheat, and the H genome from barley. The in-depth sequencing of the HMW-glutenin locus regions allowed us to compare sequence changes among the three

homoeologous A, B, and D genomes and analyze the types and rates of sequence evolution between homologous wheat genomes. Our detailed comparative sequence analyses of HMW-glutenin regions among the different wheat genomes provided molecular mechanisms underlying the rapid sequence changes among the A, B, and D genomes and revealed extensive sequence conservation between homologous HMW-glutenin genomic regions. The results from this study also provided useful knowledge on designing effective strategies to decipher the complex wheat genome.

Poster 2. Meta-analyses of QTL associated with Fusarium head blight resistance.

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Quantitative trait loci (QTL) associated with different types of Fusarium head blight (FHB) resistance have been identified from various sources. Because of differences in genetic backgrounds, experimental factors, and analysis methods, the marker loci orders on chromosomes and significance are not consistent across studies. Such discrepancies in the proposed chromosome location and the effect of putative QTL on FHB as well as differences in the amount of variation explained by markers associated with a QTL make it difficult to select common flanking markers that will be most diagnostic when applied in marker-assisted selection (MAS) and breeding. Meta-analysis has been used to estimate the confidence intervals (CI) of identified QTL in plant and animal genomes. The objective of this study is to estimate the CIs of 63 QTL associated with different types of FHB resistance and align them onto the consensus ITMI map to determine if different QTL on the same chromosomes from different studies overlap. Forty-seven QTL associated with FHB resistance types I, II, III, and IV from various sources were classified into 15 clusters on 10 chromosomes. Thirty-nine QTL are significant QTL (LOD > 4.0). Two clusters on 3BS and 5A contain confirmed QTL from Sumai 3 and Wangshuibai. Markers flanking a QTL cluster may help breeders to pyramid QTL more efficiently in marker-assisted selection.

Poster 3. Whole genome mapping and QTL analysis in a doubled-haploid population derived from the cross between a synthetic hexaploid wheat and hard red spring wheat.

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Quantitative trait loci analysis allows the identification of genomic regions associated with quantitative traits, which provides an estimation of the number and chromosomal location of genes involved and leads to the identification of molecular markers suitable for marker-assisted selection. In this research, we used the 'wheat × maize' method to develop a doubled-haploid population derived from the synthetic hexaploid wheat line TA4152-60 and the North Dakota hard red spring wheat line ND495. The population consisted of 213 lines, and a subset of 120 lines was randomly selected and used to construct linkage maps of all 21 chromosomes. The maps consisted of 626 markers, including 408 SSRs and 218 TRAPs, and spanned 3,811.5 cM with an average density of one marker per 6.1 cM. Telomere, sequence-based fixed TRAP primers were used to define the ends of seven linkage groups. Novel tan spot resistance QTL were identified on chromosomes 2A, 5A, and 5B. In addition to *Tsn1* and *Snn1*, a new *Stagonospora nodorum* blotch toxin-sensitivity gene identified on chromosome 3D was found to be significantly associated with the disease. Major QTL for days to heading, plant height, coleoptile color, glume toughness, and seed threshability also were identified. The DH population and genetic map will be a useful tool for the identification of other disease resistance QTL and agronomically important loci and aid in the identification and development of markers for marker-assisted selection.