

### **Poster 6. Survey of the wheat 5AS chromosome synteny in *Triticum* species with different ploidy levels.**

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In the frame of the project 'Physical mapping of wheat chromosome 5A', we have analyzed markers collinearity in the short arm of chromosome 5A (5AS) considering different species of the *Triticum* genus (*T. aestivum*, AABBDD, *T. turgidum* subsp. *durum* (AABB), and *T. monococcum* (AA) characterized by different ploidy levels and evolutionarily separated on a time scale in order to get insights into possible chromosomal rearrangements occurred during evolution. In detail, we relied on four mapping populations: (1) Chinese Spring (CS, *T. aestivum*) x Renan (*T. aestivum*), (2) CS x CS disomic substitution line for chromosome 5A (*T. turgidum* subsp. *dicoccoides*), (3) Latino (*T. turgidum* subsp. *durum*) x MG5323 (*T. turgidum* subsp. *dicoccum*), and (4) DV92 (*T. monococcum*) x G3116 (*T. monococcum*). Several categories of molecular markers, including SSRs (simple sequence repeat), SSR–EST (SSR-expressed sequence tags), TE junction (transposable elements), and COS (conserved ortholog set) were used to obtain high density genetic maps for the short arm of wheat chromosome 5A. The specificity of each marker for chromosome 5AS was assessed with nulli-tetrasomic lines derived from the reference cultivar Chinese Spring, and deletion lines were used to assign the physical position of the developed markers to deletion bins of 5AS. The evaluation of syntenic blocks and nonconserved regions among the homologous segments of different *Triticum* species is reported, while the mapping of EST-based markers allowed identification of syntenic regions in the rice genome. Identification of possible rearrangements in the different 5AS genetic maps of wheat is providing valuable information for the subsequent steps of BAC contigs anchoring, while the consensus map deriving from the integration of these four maps will provide a fundamental tool to link the genetic and physical maps.

### **Poster 7. Physical mapping resources for large plant genomes: Radiation hybrids for the D-genome of Chinese Spring and *Aegilops tauschii* accession AL8/78.**

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Due to the lack of recombination in certain regions of the chromosomes, genetic mapping alone is not sufficient to develop a high quality marker scaffold for sequence ready physical maps. Radiation hybrid (RH) mapping, which uses radiation induced chromosomal breaks to identify physical marker linkages, has proven to be a valuable tool for generating physical maps for complete genome assembly in animal systems. In order to construct high-resolution, RH-based physical maps of the wheat D-genome chromosomes that integrate the BAC based contigs, we developed D-Genome Radiation Hybrid (DGRH1) panels for the wheat D-genome donor *Aegilops tauschii* accession AL8/78 (1,510 DGRH1) and the reference hexaploid wheat cultivar Chinese Spring (2,565 DGRH1). Characterization of these DGRH1s with a set of molecular markers evenly spanning the entire genome indicates a homogenous marker loss (2.1%) across the chromosomes. Four different marker systems used in this study, mostly detected unique deletions suggesting that a combination

of marker systems will yield a better contiguous map. The mapping resolution of these RH panels estimated on the basis of markers spanning known distances was <140 kb. Two sets of informative lines carrying breaks in multiple D-genome chromosomes were selected from *Ae. tauschii* DGRH1s (399 lines) and Chinese Spring DGRH1s (300 lines). First generation RH maps based on 178 lines and 676 markers (641 DaRT and 35 SSR) showed a 17:1 map ratio cR/cM when compared with the genetic maps. A NimbleGen array has been designed and tested for high-throughput mapping, and a total of ~30,000 retro-junction markers and ~6,000 gene-based markers, specific to the D genome were identified. The selected DGRH1 lines currently are undergoing genotyping with this array and, once analyzed, will provide a very dense scaffold for the assembly of the D genome of wheat. This research also provides valuable resources for fine mapping and map based cloning studies of genes present on the D genome along with an unprecedented view into the evolution of grass genomes (<http://avena.pw.usda.gov/RHmapping/>).

### ***Poster 8. Construction of a radiation hybrid map for chromosome 6B of common wheat.***

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It is well known that recombination events in wheat are not evenly distributed along the length of chromosomes. This is the case with chromosome 6B, which is the target chromosome assigned to Japan by the International Wheat Genome Sequencing Consortium. The genetic maps of chromosome 6B so far constructed are heavily populated by markers in the pericentromeric region and scarcely in the telomeric regions. This represents that recombinations mostly take place in the telomeric ends of the chromosome. Therefore, we would face problems in determining orientation of contigs in the pericentromeric region if we solely depend on the genetic maps. Our objective of the current study is to establish a radiation hybrid (RH) mapping panel that can be useful in determining marker orders in the recombination-poor, pericentromeric region of chromosome 6B. We crossed nullisomic 6B-tetrasomic 6A plants of Chinese Spring (CS) wheat with the pollen freshly irradiated by  $\gamma$ -ray (15 Gy). We sowed 2,171 M0 seeds and extracted genomic DNA from 461 (21.2%) surviving plants. Additionally, we used 12 6B deletion lines (five deletions in 6BS and seven deletions in 6BL; obtained from NBRP-Wheat, Japan). We scored the presence or absence of 21 previously reported 6B-specific SSR markers and four newly developed EST-based markers. We analyzed the data by CarthaGene software to construct a RH map with the default setting. The resulting RH map consisted of two linkage groups, corresponding to the short and long arms. The gap between the linkage groups may be due to the absence of markers in a pericentromeric bin (6BS-CEN-0.25). The marker order is consistent with that of bin mapping and largely with a previously reported genetic map. All the RH-map markers occupied individual loci. Two pairs of markers genetically mapped to the same loci in the pericentromeric region of 6B were completely separated from each other in the RH map. This result indicates that RH mapping with our panel has better resolution in proximal region of 6B.

### ***Poster 9. A NimbleGen comparative genomic hybridization array for high-throughput physical mapping of genome specific repeat junction and gene-based markers in the D genome of hexaploid wheat.***

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Mapping and map-based cloning of agriculturally and economically important traits remain a great challenge in complex highly repetitive genomes such as the grass tribe, Triticeae. This limitation is primarily based upon the availability of polymorphic markers and frequency of genetic recombination. Most markers are gene-based, derived from polymorphisms within coding regions. Non-gene-based markers, such as repeat junction markers, are derived from the noncoding intergenic space. Repeat junction markers take advantage of the repetitive nature of the wheat genome, providing random and equal distribution of these markers throughout the genome and can facilitate mapping efforts. Repeat junction markers are designed upon the junction of nested repetitive elements, and approximately 90% of these markers have been determined to behave as a single copy locus and are genome specific. Repeat junction markers were designed from 454 genome sequences of the wheat D-genome progenitor, *Aegilops tauschii*. Mapping of *Ae. tauschii* repeat junction