has on recombination frequencies. Our RH data support for the first time *in vivo* the hypothesis of noncasual interaction between recombination hot-spots and DNA break/repair. This means that since the initial RH application 37 years ago, we were able to show now for the first time that the third hypothesis of RH mapping (iii) might not be entirely correct.

Poster 77. Molecular characterization and protein-DNA interaction analysis of a WRKY transcription factor in wheat (Triticum aestivum L.) showing differential spatiotemporal expression during leaf-rust pathogenesis.

Dhananjay Kumar, Anjali Kapoor, Kunal Mukhopadhyay, and Manish Kumar. Department of Biotechnology, Birla Institute of Technology, Mesra, Ranchi-835215, Jharkhand, India.

The plant WRKY proteins are one of the largest families of transcription factors involved in physiological processes including biotic and abiotic stresses. These transcription factors have a recognition sequence (T)(T)TGAC(C/T), known as the W-box, found in the promoter region of *WRKY* and other defense-related genes. These transcription factors have a strictly conserved 60 amino acid region containing the WRKYGQK peptide sequence and zinc finger-like motifs. The *WRKY* gene family is subdivided into three different groups (I, II, and III) based on the number of WRKY domains and features associated with the zinc finger like motifs, group I and group II have finger motif C_2H_2, whereas group III contains a C_2HC motif. Very little information on WRKY transcription factors of wheat are available and, hence, the present study was undertaken to characterize a wheat WRKY transcription factor and decipher its role during leaf-rust infection. *WRKY*-specific, wheat consensus sequences were obtained from NCBI to design primers to amplify genomic and cDNA sequences from leaf-rust resistant and susceptible near isogenic wheat lines. The sequences were analyzed using multiple sequence alignment and a conscientious phylogenetic tree was constructed to study the relationship with WRKY transcription factors of other plants. Various catalytic domains were identified using ScanProsite and other bioinformatic softwares. *In silico* docking between a 26-bp oligonucleotide containing the W-box sequence and a 74 amino acid sequence having the conserved 60 amino acids including the core WRKYGQK domain was performed at Haddock server. The optimized peptide and DNA models were able to form a complex that showed interaction between the W-box and the conserved WRKY domain. The protein–DNA binding was validated by observing shifts in electrophoretic mobility shift assay using heterologously produced recombinant WRKY proteins. Temporal and spatial gene expression profiling was done by quantitative real-time PCR using universal probe library based, WRKY-specific probe and primers with RNA isolated at different time points from resistant and susceptible plants that were either mock inoculated or infected with virulent leaf-rust pathogen. During pathogenesis, maximum expression of *WRKY* gene was observed at 24-hours post-inoculation in both compatible and incompatible interaction.

Poster 78. Transferring maize transposable elements to bread wheat through 'wheat x maize' crossing.

Ragupathi Nagarajan [1], Ganna Kondratiuk [1], Gurmukh S. Johal [2], and Kulvinder Gill [1].

[1] Crop and Soil Sciences Department, Washington State University, Pullman, WA 99164, USA, and [2] Botany and Plant Pathology Department, Purdue University, West Lafayette, IN 47907, USA.

Transposon tagging and insertional mutagenesis are important reverse genetic tools in eukaryotes. These are widely used for understanding gene function and targeted manipulation of genes in crop improvement. Although no characterized transposable element system is available in wheat, the maize systems have proven to be very successful for gene tagging not only in maize but also in other plant species including tobacco, *Arabidopsis*, peas, and barley. Normally, genetic transformation is used to introduce these active transposable elements (TE) into nonhost plants. In the case of wheat, however, genetic transformation is technically challenging, variety specific, and the frequency is usually very low. We attempted a simpler method based on direct 'wheat x maize' crossing to transfer well-characterized maize transposable elements into wheat. Wheat doubled haploids are routinely produced by 'wheat x maize' crossing where the resulting zygotes undergo uniparental chromosome elimination giving rise to haploid wheat plants. The maize chromosomes are present during the few initial cell cycles during zygote differentiation, giving enough time for the transposition of TEs. Different maize lines harboring Activator (*Ac*) and Mutator (*Mu*) transposable elements were crossed with bread wheat lines and the haploid embryos were rescued. Regenerated wheat plants were treated with colchicine for chromosome

doubling. These plants were screened by PCR amplification using multiple maize TE specific primers. Three of the 64 plants from the Louise/Maize Activator crosses showed a maize-specific, PCR fragment amplified using *Ac*-specific, PCR primers. Similarly, four of the 40 plants derived from PBW 621/Maize Mutator crosses showed a maize-specific ,PCR fragment amplified using *Mu*-specific PCR primers. Amplified fragments were sequenced to confirm their specificity. All the fragments were matching up with their corresponding maize transposable element sequences. DNA gel blot analysis and inverse PCR are underway to confirm the integration and subsequent transposition activities. The most current status of the project will be presented at the meeting.

Poster 79. The wheat meiotic cohesin gene TtRec8 and its role in haploidy-dependent, unreductional meiotic cell division.

Guojia Ma [1], Wun S. Chao [2], Liwang Liu [1,4], Yong Qiang Gu [3], Steven S. Xu [2], and Xiwen Cai [1].

[1] Department of Plant Sciences, North Dakota State University, Fargo, ND 58108-6050, USA; [2] USDA–ARS, Red River Valley Agricultural Research Center, Fargo, ND 58102, USA; and [3] USDA–ARS, Western Regional Research Center, Albany, CA 94710, USA; [4] Current address: National Key Laboratory of Crop Genetics and Germplasm Enhancement, College of Horticulture, Nanjing Agricultural University, Nanjing 210095, P.R. China.

Meiosis is a specialized cell division that halves chromosomes and generates haploid gametes in eukaryotes. It ensures genomic integrity and generates genetic variability. Variation of the meiotic process leads to aneuploidy and polyploidy. Unreductional meiotic cell division (UMCD) was observed in the polyhaploid of the tetraploid wheat cultivar Langdon (LDN) (*Triticum turgidum* L.) and its interspecific hybrid with *Aegilops tauschii*. This haploidy-dependent UMCD gives rise to unreduced gametes, leading to polyploidy. It has been considered a major mechanism of polyploidization in wheat. The meiotic cohesin gene *Rec8* has been proven to play a significant role in kinetochore orientation and chromosome segregation at meiosis I in model species. In the present study, we attempted to understand the function of the *Rec8*-like gene in the meiotic cell division of wheat and determine the role of this gene in the onset of the haploidy-dependent UMCD. We cloned the *Rec8* homologue in LDN, designated *TtRec8*, and developed the polyclonal antibody against the TtRec8 protein in rabbit. *TtRec8* exhibited an expression pattern similar to *Rec8* in model species as revealed by real-time PCR and Western blotting throughout the meiotic process in anthers. In addition, the TtRec8 protein exhibited analogous kinetics of the meiotic cohesin *Rec8* as revealed by the immunolocalization of the cohesion protein on chromosomes over the meiotic stages. Two homoeoloci of *TtRec8* were identified on chromosome 1A and 1B. We have been determining the map location of this gene on the chromosome in a doubled haploid population of tetraploid wheat. Meanwhile, we have been investigating the expression profiles of *TtRec8* and the kinetics of the TtRec8 protein in an LDN haploid as well as the interspecific hybrid of LDN with *Ae. tauschii*. This will provide new insights into the role of this meiotic cohesin gene in the onset of the haploidy-dependent UMCD in wheat.