Poster 75. Androgenic response of Nebraskan winter wheat (Triticum aestivum L.) varieties to isolated microspore culture for doubled haploid plant production.

B.K. Das ¹, M. Santra ¹, A. Hazen ¹, P. S. Baenziger ², and D.K. Santra ¹.

Isolated microspore culture based androgenesis is being used for production of haploid (H) and doubled haploid (DH) plants in wheat. This technique can facilitate the efficiency of wheat breeding and genetic mapping as well as research in functional genomics and gene expression. Until now there was no report of microspore culture in Nebraskan winter wheat varieties. The objectives of the present report were to (1) study the androgenic response of Nebraskan winter wheat varieties and (2) establish efficient procedure for green DH plant regeneration. Three Nebraskan winter varieties (Anton, Antelope, and Camelot) were used. The spikes were collected from greenhouse-grown plants when microspores were at mid-late to late-uninucleate stage. For each batch of pretreatment, anthers from 16 spikes were pretreated in solution B at 25°C for 4–5 days followed by microspores isolation (no cold pretreatment). For cold pretreatment, anthers were incubated at 4°C for additional five days. The numbers of embryogenic microspores, multicellular, and embryo-like structures were recorded and analyzed. Compared to no cold pretreatment, a cold pretreatment increased the number of embryogenic microspores significantly in Anton by two fold, but no significant differences between the two pretreatments were observed in Camelot and Antelope. In vitro development of microspores into multicellular and embryo-like structures were quicker in Camelot than Anton and Antelope. The green plants were regenerated in all three varieties following both cold and no cold pretreatment. The number of regenerated green plants per batch of pretreatment was four (no cold) and eight (cold) in case of Antelope. However, for Anton and Camelot, there was one green plant per batch in both the cold and no cold treatments. It seems that higher number of embryogenic microspores due to cold pretreatment in Anton was not regenerated into proportionate number of green plants. An experiment is under progress to determine a similar response in Camelot and Antelope. This is the first report of androgenic response of Nebraskan winter wheat varieties. We believe that this method will be a beneficial tool in our wheat breeding efforts. However green plant regeneration frequency needs to be increased for cost-effective use.

Poster 76. Chromatin state affects the DNA breakage/repair mechanism in wheat.

A. Kumar ¹, F.M. Bassi ¹, Muhammad J. Iqbal ¹, E. Paux ², O. Al-Azzam ³, M. M. de Jimenez ¹, A. M. Denton ³, Y.Q. Gu ⁴, E. Huttner ⁵, A. Kilian ⁵, S. Kumar ⁶, A. Goyal ⁶, V. Tiwari ⁷, M. Dogramaci ¹, H.S. Balyan ⁶, H.S. Dhaliwal ⁸, P.K. Gupta ⁶, G.S. Randhawa ⁹, C. Feuillet ², W. P. Pawlowski ¹⁰, and S.F. Kianian ¹.

¹ Department of Plant Sciences, North Dakota State University, Fargo, ND 58102, USA; ² INRA-UBP 1095, Genetics Diversity and Ecophysiology of Cereals, 63100 Clermont-Ferrand, France; ³ Department of Computer Sciences, North Dakota State University, Fargo, ND 58102, USA; ⁴ USDA–ARS, Western Regional Research Center, Albany, CA 94710, USA; ⁵ Diversity Arrays Technology Pty Ltd, Yarralumla, ACT 2600, Australia; ⁶ Department of Genetics and Plant Breeding, Ch. Charan Singh University, Meerut 25004, India; ⁷ Department of Crop and Soil Science, Oregon State University, Corvallis, OR 97331, USA; ⁸ Akal School of Biotechnology, Eternal University, Baru Sahib 173101, India; ⁹ Department of Biotechnology, Indian Institute of Technology, Roorkee 247667, India; and ¹⁰ Department of Plant Breeding and Genetics, Cornell University, Ithaca, NY 14853, USA.

Meiotic recombination, the basis of genetic mapping is not uniformly distributed across the genome. The regions of high and low recombination that result in uneven map resolution across the chromosomes are evident across many grass genomes including wheat ($Triticum\ aestivum\ L$.). Recombination is believed to be linked to the double-stranded DNA break and repair, a phenomenon highly dependent on the chromatin state. Radiation hybrid (RH) maps have been proposed to provide i) higher, ii) be more uniform resolution than genetic maps, and iii) to be independent from recombination constraints. We generated an RH panel for mapping of wheat chromosome 3B and used it to test these three assumptions. Our RH map contains 541 markers anchored to chromosome 3B BAC contigs. Detailed comparisons with a genetic map of similar quality confirmed that i) the resolution of the RH map was 10.5X higher and ii) six times more uniform. We identified a strong interaction (r = 0.879 at p = 0.01) between the DNA repair mechanism in mitotic cells and the distribution of crossing-over events in meiotic cells. We could explain this finding only by admitting the possibility that the DNA repair mechanism is affected by the chromatin state in a way similar to the effect that chromatin state

¹ Panhandle Research and Extension Center, University of Nebraska, 4502 Avenue I, Scottsbluff, NE 69361, USA, and ² Department of Agronomy and Horticulture, University of Nebraska, Lincoln, NE 68583, USA.