

Summary of Genotyping Data from the 2008 Regional Performance Nurseries

Hard winter wheat breeding lines from the 2008 Northern and Southern Regional Performance Nurseries were analyzed for 21 traits using 37 markers. The complete data set is included in the attached spreadsheet. The expected size (in base pairs) of each target band is included in the data set. Sizes preceded with the letter "T" are based on tailed primers and should be 18 base pairs longer than published reports. In the spreadsheet, a "+" indicates that the target allele was present, a "-" indicates that the target allele was absent, and a "?" indicates that the PCR fragment was either missing or due to PCR failure.

Except where noted, protocols used for all assays are listed on the MASWheat website (<http://maswheat.ucdavis.edu/protocols/index.htm>).

Fungal Resistance Traits

1. Wheat Scab (3BS QTL)

Three SSR markers (GWM493 and GWM533, TAG 2003 107:503-508; STS-3B-256, M. Pumphrey and Jim Anderson, personal communication) were used to detect the presence of a QTL on chromosome 3BS that confers resistance to wheat scab. Only two lines, KS970093-8-9-#1 and AP05TW2821, have one of the target bands for GWM493 (211 bp) and GWM533 (159 bp), respectively. No line contained more than one target band in three marker alleles as seen in the controls Sumai 3. The data suggest that none of the lines have the 3BS QTL.

2. Lr21

Newly designed primers for a shorter fragment were used for detecting the Lr21 resistance allele. This set of primers was based on the gene sequence provided by Dr. Li Huang from Dr. B. Gill's lab. Known positive (WGRC07, WGRC27) and negative (WGRC02, Wichita) control lines were tested with the new primers and showed the expected allele. Only entry HV9W03-539R had the band (214 bp) as in the positive controls and likely has the Lr21 resistance gene.

3. Lr24/Sr24

Two STS markers were used to screen for Lr24/Sr24. Sr24#12 and Sr24#50 (Theor Appl Genet (2005) 111: 496–504) are both STS markers closely linked to Sr24 and typically amplify only one band. Resistant germplasm LcSr24Ag was positive for both markers. Sixteen lines were positive for marker Sr24#12 (512 bp) and Sr24#50: Trego, OK03522, KS05HW15-2, KS05HW136-3, HV9W02-942R, TX02A0252, TX03A0563, TX04M410211, SD06W117, SD05118, SD05W030, NE02533, NE02558, NW03666, NE05549, and MT0552. These 16 lines more likely have Lr24/Sr24 resistance gene. TAM107 was positive only for Sr24#50 but not for Sr24#12.

4. Lr34/Yr18

The slow leaf rusting gene Lr34 and yellow rust resistance gene Yr18 are flanked by two markers (SSR marker SWM10, TAG 2006 113:1049–1062; STS marker csLV34-LR34, TAG 2006 114:21–30). Chinese Spring and Thatcher-Lr34 have the Lr34 resistance allele. CS7DS-4 (a deletion line of Chinese Spring) and Thatcher are both susceptible to Lr34. Nineteen lines were positive for both markers: OK00514-05806, OK05737W, OK04505, AP06T3832, HV9W02-942R, KS980512-11-22, TX02A0252, TX03A0148, TX03A0563, TX04A001246, TX01V5134RC-3, TX04M410211, Antelope, SD06W117, SD05210, NX03Y2489, NE02558, NE04490, NE05569 and possibly have Lr34/Yr18 resistance allele. However, Jagger also has the two resistance marker alleles but may not carry the Lr34 resistance gene, therefore, caution needs to be taken for these materials have Jagger in their pedigrees.

5. Lr37/SR38/Yr17

The three rust resistance genes are on a chromosome segment that does not appear to recombine with bread wheat chromosomes. The STS marker (VENTRIUP-LN2) should be therefore completely linked with the resistance genes. The following 31 lines were positive for the marker and likely have the alien segment: TAM-107, Trego, OK00514-05806, OK05737W, OK04505, AP04T8211, AP05T2413, AP05TW2821, AP06T3832, KS05HW15-2, KS05HW121-2, KS05HW122-5, HV9W03-539R, HV9W03-696R-1, HV9W02-942R, NE05425, NE05426, KS980512-11-22, CO02W237, KS980512-2-2, TX01V5134RC-3, TX04M410164, TX04M410211, TX04V075080, SD05W030, NX04Y2107, HV9W03-1379R, NE02533, NE02558, NW03666, and NE04490.

6. Lr39/Lr41

These two resistance genes appear to be the same or closely linked genes and SSR GDM35 was reported to be closely linked markers. More recently, SSR Barc124 is closer marker for the gene. We tested both markers in the study. No line had the expected 183 bp band of GDM35 as that in the controls. Two lines, HV9W96-1271R-1, and KS980512-11-22 had the expected Barc124 band (260 bp). The markers were run twice with DNA isolated from different plants. The data suggest that only HV9W96-1271R-1 and KS980512-11-22 may have Lr39/Lr41.

7. Lr50

Lr50 is flanked by microsatellite markers GWM382 (6.7 cM) and GDM87 (9.4 cM) on wheat chromosome arm 2BL. In the resistant line WGRC36, marker GDM87 produces one distinct band of 124 bp. The following seven lines have the GDM87 124 bp band as found in the positive control WGRC36: CO03W139, CO03W239, OK03825-5403-6, TX04M410211, N98L20040-44, HV9W03-1379R, and MT0495.

None of the lines have the 156 bp band found in WGRC36 for marker GWM382. The seven lines with the GDM87 124 bp band may have the Lr50 gene if they are from a pedigree with Lr50 gene.

8. Sr2

The Sr2 resistance gene has been effective worldwide for more than 50 years. It has recessive inheritance and expresses primarily as adult-plant resistance. It is located on 3BS in the same region as the FHB QTL. The SSR marker, GWM533, produces a 133 bp band in resistant lines (Spielmeyer, 2003. Crop Sci. 43:333–336) and is only 1 to 2 cM away from the gene. The 133 bp band showing in our positive controls (Eagle(USA), Sonalika) presented in 46 lines: Kharkof, Scout 66, Trego, OK03522, KS05HW15-2, KS05HW121-2, KS05HW122-5, KS05HW136-3, HV9W96-1271R-1, HV9W03-696R-1, HV9W02-942R, T153, T154, NE04424, NE05430, KS970187-1-10, KS980512-11-22, KS980554-12--9, CO02W237, CO03064, CO03W054, CO03W139, CO03W239, NE05496, TX03A0148, TX03A0563, TX04A001246, TX04M410164, TX04M410211, Kharkof, Antelope, Jerry, SD06W117, SD03164-1, SD03164-2, SD05210, SD05W030, NW04Y2188, NE02533, NE02558, NW03666, NI04427, NE05549, NE05569, MTS0531, and MT0552. Therefore, these lines may have the Sr2 gene.

Insect Resistance Traits

9. Hessian Fly (H9)

One STS marker was analyzed for the presence of H9 resistance to Hessian fly biotype L. The following six lines had the expected 909 bp band as in 'Iris' and likely have H9 gene: Kharkof, Trego, KS05HW15-2, NE05425, NE05426, and TX04M410211.

10. Hessian Fly (H13)

Three SSR markers (GDM36, CFA2153, and CFD132) were used to test lines for the presence of H13 resistance allele to Hessian fly biotype L. Only one line, HV9W96-1271R-1, has the CFD132 166 bp band. It is likely that none of the tested lines has H13 resistance gene.

11. Russian Wheat Aphid (Dn1, Dn2, Dn5, Dn6, Dnx)

Dn1, Dn2, Dn5, Dn6, Dnx, RWA genes....

We have data on GWM44 and GWM111, but since the controls for all of the RWA genes showed complex band patterns, we can not determine which band is corresponding to the resistance gene based on available information from the publication.

Viral Resistance Traits

12. Barley Yellow Dwarf Virus (Bydv2)

One SCAR marker (BYAgi) was used to detect the presence of the Bydv2 gene. No line had the expected 567 bp band as in P961341. The data suggest that none of the lines have Bydv2.

13. Wheat Streak Mosaic Virus (Wsm1)

STS marker (J15) was used to detect the chromosome segment containing the Wsm1 gene transferred from *Agropyron intermedium*. One line, NW04Y2188, had the expected 431 bp band as in KS93WGRC27 and likely has Wsm1.

Quality Traits

14. 1RS Translocation

One rye SSR marker (SCM9, Euphytica 2003 132: 243–250, <http://maswheat.ucdavis.edu/protocols/drought/index.htm>) was used to detect the presence of the 1RS rye translocation. This marker seems to produce false positive in some line, therefore it was analyzed in both agarose gel and ABI 3730 DNA Analyzer. SCM9 amplified a 225 bp band for the 1B/1R in check cultivar 'Aurora' and in 8 lines. SCM9 amplified a 242 bp band for the 1A/1R in check cultivar TAM107 and in 11 lines. The following 8 lines were positive for the SCM9 225bp band from rye and likely have the 1B/1R translocation: KS05HW15-2, HV9W03-696R-1, HV9W02-942R, NE04424, OK03825-5403-6, TX04M410211, SD05W030, and NI04427. The following 11 lines carry the SCM9 242bp allele and likely have the 1A/1R translocation: TAM-107, AP05T2413, HV9W96-1271R-1, T151, T153, T154, T158, TX04A001246, TX01V5134RC-3, SD05210, and HV9W03-1379R.

15. High Grain Protein Content, HGPC

One STS marker (UCW89) very closely (0.1 cM) linked with the Gpc-B1 gene was used to identify the gene for HGPC. The positive control 'Glupro' produces a band of 138 bp. No line had the distinct 138 bp band, therefore the Gpc-B1 gene may not present in the lines analyzed.

16. High Molecular Weight Glutenins

Three STS markers (Euphytica 2003 134:51-60) were used to determine some of the alleles at the 3 loci controlling high molecular weight glutenins. Marker HMWAx2* produces one band of 1319 bp for Ax2* genotype, or no band for Ax1 genotype. HMWBx produces one band of 669 bp for Bx17 genotype, or 2 bands (630 and 766 bp) for all others (non-Bx17 genotypes). HMWDx5 will produce one 478 bp band for Dx5 genotype, or no band for all others (non-Dx5 genotypes).

These three markers appear to be extremely sensitive to small changes in PCR conditions. Reproducibility of the data using these markers is low to moderate.

Thirteen lines without the HMWAx2* band likely carry Ax1 allele (OK00514-05806, OK05737W, OK03522, AP04T8211, AP05TW2821, AP06T3832, KS970093-8-9-#1, OK03825-5403-6, TX03A0148, Wesley, NW04Y2188, NE02533, MT0495) and the remaining entries likely carry the Ax2* allele. Eighteen lines with the HMWBx 669 bp band likely carry Bx17 allele (OK00514-05806, OK05737W, OK04505, AP04T8211, AP05T2413, AP05TW2821, AP06T3832, T154, CO02W237, TX01V5134RC-3, TX04M410211, TX04V075080, SD05210, HV9W03-1379R, NE02533, NE02558, NE04490, NE05548) and the remaining entries likely carry non-Bx17 allele. Most of the lines tested with HMWDx5 produced a band of 478 bp and therefore, carry Dx5 allele. Only 8 lines (AP05T2413, T151, T153, T154, T158, KS970093-8-9-#1, CO03W043, KS980512-2-2) amplified no band and likely carry non- Dx5 allele.

17. Grain Texture (Pina-D1, Pinb-D1)

One dominant STS marker (Pina-D1) was used to screen for the presence of wild-type (Pina-D1a), soft alleles. The positive control, 'Newana' yielded the expected band size of 348 bp that is associated with soft texture. All entries have the 348 bp band of the Pina-D1a for soft texture except the following five lines that did not carry the 348 bp allele and likely have the null allele (Pina-D1b) for hard texture: OK00514-05806, AP04T8211, KS980554-12-~9, HV9W03-1379R, and NE04490.

A codominant PCR-CAPs marker (Pinb-D1) was used to screen for Pinb-D1 alleles. After PCR amplification and restriction using Bsr BI, a 320 bp band indicates the soft allele (Pinb-D1a). A band of 200 bp indicates the mutant allele (Pinb-D1b) for hard texture. Sixteen lines had the 320 bp band of soft allele Pinb-D1a (OK00514-05806, OK04505, AP04T8211, KS05HW121-2, KS05HW122-5, HV9W03-539R, KS980554-12-~9, TX01V5134RC-3, Kharkof, SD05118, SD05210, NX03Y2489, HV9W03-1379R, NE02533, NE02558, NE04490). The remaining lines had the 200 bp of Pinb-D1b allele for hard texture.

Combined analysis of two grain texture loci shows that 11 lines have soft alleles of both markers Pina-D1a and Pinb-D1a: Kharkof, OK04505, KS05HW121-2, KS05HW122-5, HV9W03-539R, SD05118, TX01V5134RC-3, SD05210, NX03Y2489, NE02533, and NE02558. Those lines are either soft wheat, or other genes may be involved in determination of grain hardness in these lines.

18. Waxy Mutants

One STS marker (Waxy4) was used to detect null mutants at all three loci controlling granule-bound starch synthase (GBSS) or waxy protein. Almost all entries had all three bands and are non-mutants or non-waxy lines. Two lines (SD06165, NE02533) showed mutation in the Wx-A1 locus (273 bp) on 7AS, 9 lines (KS05HW121-2, KS05HW122-5, KS05HW136-3, KS980554-12-~9, TX04A001246, TX04M410211, TX04V075080, NX03Y2489, NX04Y2107) showed mutation in the 4AL locus (243 bp). These 11 lines are likely partial waxy wheat.

Abiotic Stress and Agronomic Traits

19. Aluminum Tolerance

Two SSR markers WMC331 and ALMT1-SSR3A (Mol Breeding 2006 18:171–183) were used for screening of 4DL Al-resistance QTL. Sixteen entries (OK04505, AP04T8211, HV9W03-539R, KS970093-8-9-#1, TX01V5134RC-3, TX04M410211, Wesley, SD06165, SD05118, NX04Y2107, HV9W03-1379R, NE02533, NE02558, NE04490, NE05569, and MT0552) were positive for both SSR markers and are most likely have the 4DL Al tolerance QTL. Additional five lines that were positive for only the WMC331 SSR marker (KS05HW121-2, KS05HW122-5, NE05425, NE05426, KS980554-12-~9) and one line (TX04V075080) was positive for only the ALMT1-SSR3A SSR marker may also carry the Al tolerance QTL on chromosome 4D.

20. Plant Height Genes (Rht1, Rht2, Rht8)

Two gene-specific STS markers were used to detect the Rht1 and Rht2 genes (TAG 2002 105:1038-1042). One linked SSR marker (GWM261, TAG 1998 96:1104-1109) was used to detect Rht8. All but fourteen lines (OK04505, KS970093-8-9-#1, KS980554-12-~9, CO03W239, OK03825-5403-6, TX03A0148, Kharkof, Wesley, SD06W117, SD05210, SD05W030, HV9W03-1379R, NE02533, and NE02558) had the 255 bp band indicating the presence of the Rht1 gene. Only two lines (KS970093-8-9-#1 and KS980554-12-~9) had the 270 bp band diagnostic for the Rht2 gene. Six lines (OK03305, TX03A0148, Wesley, SD06069, SD06163, and NE05549) had the 212 bp band linked with Rht8 and may carry the Rht8 gene.

21. Pre-harvest sprouting tolerance (PHS)

Two SSR markers (Barc321 and Barc12) were used to screen pre-harvest sprouting tolerance QTL on 3A that we have recently been mapped in Rio Blanco. Nine lines (HV9W96-1271R-1, T153, T154, KS970187-1-10, KS980554-12-~9, TX03A0563, TX04M410211, NE02533, and NI04420) had the expected bands (187 bp for Barc321 and 218 bp for Barc21) as found in Rio Blanco. Therefore, the nine lines may carry 3A pre-harvest sprouting tolerance allele.