Trial VI (double dwarf trial). In this yield comparison, line 08 (1.267 kg) had the highest grain yield/plot. Subsequent lines with high grain yields/plot were 06 (1.167 kg), 01 (1.067 kg), 02 (1.017 kg), and 03 (1.017 kg). The check cultivars Sarsabz (0. 983 kg) and Kiran-95 (0.833 kg) had comparatively lower grain yields than all other lines.

Coleoptile length studies.

Eleven genotypes (seven cultivars and four lines) were studied for coleoptile length in three replicates. Genotypes with Rht_1, Rht_2, Rht_1Rht_2, Rht_8Rht_9, and *rht* were compared for their coleoptile length under controlled environmental conditions. The results suggested that the traditionally tall cultivar C-591 (*rht*) had a longer coleoptile than all other cultivars and genotypes. Subsequent genotypes with long coleoptile lengths were Chinese Spring (*rht*) and Rht8-01 (Rht_8). The cultivars Mara (Rht_8Rht_9), Sarsabz (Rht_1), and Soghat-90 (Rht_2) were not significantly different. Line Rht8-02 has the Rht_8 dwarfing genes but was not significantly different than the double-dwarf cultivar Yeccora (Rht_1Rht_2). These results suggest that dwarfing genes probably do not affect the coleoptile length. The genetic background may affect the coleoptile length of individual cultivars.

Participation in an international meeting.

K.D. Jamali participated in an international project planning meeting and presented country report of the IAEA project No. RAS/05/045 held from 25–30 June, 2007, in Kuala Lumpur, Malaysia.

PEOPLES REPUBLIC OF CHINA

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Allelic variation of polyphenol oxidase genes located on chromosomes 2A and 2D and development of functional markers for the PPO genes in common wheat.

Polyphenol oxidase (PPO) activity is highly related to the undesirable browning of wheat-based end products, especially Asian noodles. Characterization of PPO genes and the development of their functional markers are of great importance for marker-assisted selection in wheat breeding. In the present study, complete genomic DNA sequences of two PPO genes, one each located on chromosomes 2A and 2D and their allelic variants were characterized by means of *in silico* cloning and experimental validation. Sequences were aligned at both DNA and protein levels. Two haplotypes on chromosome 2D showed 95.2% sequence identity at the DNA level, indicating much more sequence diversity than those on chromosome 2A with 99.6% sequence identity. Both of the PPO genes on chromosomes 2A and 2D contain an ORF of 1,731 bp, encoding a PPO precursor peptide of 577 amino acids with a predicted molecular mass of ~64 kD. Two complementary dominant STS markers, *PPO16* and *PPO29*, were developed based on the PPO gene haplotypes located on chromosome 2D; they amplify a 713-bp fragment in cultivars with low PPO activity and a 490-bp fragment in those with high PPO activity, respectively. The two markers were mapped on chromosome 2DL using a DH population derived from the cross 'Zhongyou 9507/CA9632', and a set of NT and Dt lines 2DS of Chinese Spring. QTL analysis indicated that the PPO gene cosegregated with the two STS markers and was closely linked to SSR marker *Xwmc41* on chromosome 2DL, explaining from 9.6% to 24.4% of the phenotypic variance for PPO activity across three environments. In order to simultaneously detect PPO loci on chromosomes 2A and 2D, a multiplexed marker combination *PPO33*/*PPO16* was developed and yielded distinguishable DNA patterns in a number of cultivars**.** The STS marker *PPO33* for the PPO gene on chromosome 2A is homologous with *PPO18* that we reported previously, and can amplify a 481-bp and a

290-bp fragment from cultivars with low and high PPO activity, respectively. A total of 217 Chinese wheat cultivars and advanced lines were used to validate the association between the polymorphic fragments and grain PPO activity. The results showed that the marker combination *PPO33*/*PPO16* is efficient and reliable for evaluating PPO activity and can be used in wheat-breeding programs aimed for noodle and other end product quality improvement.

Characterization of a phytoene synthase 1 gene (Psy1) located on common wheat chromosome 7A and development of a functional marker.

Phytoene synthase (Psy), a critical enzyme in the carotenoid biosynthetic pathway, demonstrated high association with the yellow pigment (YP) content in wheat grain. In this study, the full-length genomic DNA sequence of a Psy gene (*Psy-A1*) located on chromosome 7A, was characterized by *in silico* cloning and experimental validation. The cloned *Psy-A1* comprises six exons and five introns, 4,175 bp in total, and an ORF of 1,284 bp, encoding a Psy precursor peptide of 428 amino acids with a calculated molecular weight of ~47.7 kD. A codominant marker, *YP7A*, was developed based on polymorphisms of two haplotypes of *Psy-A1*, yielding 194-bp and 231-bp fragments in cultivars with high and low YP content, respectively. The marker *YP7A* was mapped on chromosome 7AL using a RIL population from cross 'PH82-2/Neixing 188', and a set of Chinese Spring nullisomic-tetrasomic lines and ditelosomic line 7AS. *Psy-A1*, cosegregating with the STS marker *YP7A*, was linked to SSR marker *Xwmc809* on chromosome 7AL with a genetic distance of 5.8 cM, and explained 20 to 28% of the phenotypic variance for YP content across three environments. A total of 217 Chinese wheat cultivars and advanced lines were used to validate the association between the polymorphic band pattern and grain YP content. The results showed that the functional marker *YP7A* was closely related to grain YP content and, therefore, could be used in wheat breeding programs targeting of YP content for various wheat-based products.

QTL mapping for flour color components, yellow pigment content and polyphenol oxidase activity in common wheat.

Improvement of flour color is an important breeding objective for various wheat-based end-products. The objectives of this study were to genetically dissect the QTL for flour color components, yellow-pigment content (YPC), and PPO activity, using 240 RILs derived from the cross between the Chinese wheat cultivars PH82-2 and Neixiang188. Field trials were performed in a Latinized α-lattice design in Anyang and Jiaozuo of Henan Province and Taian of Shandong Province in 2005–06 and 2006–07 cropping seasons. One hundred and eighty-eight polymorphic SSR markers, one rye secalin marker *Sec1*, one STS marker *YP7A* for phytoene synthase gene (*Psy-A1*), and four glutenin subunit markers, were employed to genotype the population and construct the linkage map for subsequent QTL analysis. The results indicated that 27 QTL were detected for color components, YPC, and PPO activity, mainly in two clusters on chromosomes T1B·1R and 7AL, respectively. The T1B·1R translocation and phytoene synthase (*Psy*) gene on chromosome 7AL showed great influence on YPC and different color parameters of flour and noodle, explaining 31.9 and 33.9% of phenotypic variance for YPC, respectively. PPO activity was primarily conditioned by the QTL *QPpo-2A* that was closely linked to the SSR marker *Xwmc170* on chromosome 2A and explained 18.3–23.3% of phenotypic variance across three environments. Among different color parameters, flour yellowness index exhibited the highest correlation with YPC (r=0.96, P<0.0001) and very significant correlation with white salted noodle color b* (Nb*) (r=0.76, P<0.0001) and is, thus, a most desirable indicator for yellowness of flour and noodle. The markers *Sec1* and *YP7A* were very closely linked to the QTL for YPC and flour-color components and, therefore, could be used as efficient molecular markers targeting for the selection of YPC and flour-color parameters in wheat breeding programs.

Isolation and expression of novel Viviparous-1 genes in common wheat.

Preharvest sprouting (PHS) of wheat reduces the quality and economic value of grain and increasing PHS tolerance is one of the most important traits in wheat breeding. Two new *Vp-1B* alleles related to PHS tolerance were identified on the B genome of bread wheat and designated as *Vp-1Bb* and *Vp-1Bc*. Sequence analysis showed that *Vp-1Bb* and *Vp-1Bc* had an insertion of 193 bp and a deletion of 83 bp located in the third intron region of the *Vp-1B* gene, and shared 95.43% and 97.89%, respectively, similar to the sequence of AJ400713 (*Vp-1Ba*) at the nucleotide level. Their sequences are deposited in the GenBank under the accession numbers of DQ517493 (*Vp-1Bb*) and DQ517494 (*Vp-1Bc*). Semiquantitative RT-PCR analysis showed that alternatively spliced transcripts of the *Vp-1A*, *Vp-1B*, and *Vp-1D* homologues were present and there were no differences in the splicing patterns or abundances of *Vp-1A* and *Vp-1D* from 35

DAP embryos between PHS-tolerant and susceptible cultivars. *Vp-1Ba, Vp-1Bb*, and *Vp-1Bc* each could produce a set of transcripts, only one of which was correctly spliced and had the capacity to encode the full length VP1 protein. The protein was more highly expressed in genotypes with *Vp-1Bb* and *Vp-1Bc* than in those with *Vp-1Ba*. Comparison of the expression patterns of *Vp-1Ba, Vp-1Bb*, and *Vp-1Bc* at different times after pollination also revealed that the expression of these genes was developmentally regulated. Furthermore, genotypes with different levels of tolerance to PHS showed different responsiveness to ABA exposure and differences in transcript levels of *Vp-1Ba, Vp-1Bb*, and *Vp-1Bc* were observed after ABA treatment. The results indicated that insertion or deletion in the third intron region may affect the expression of the *Vp-1B* gene and its sensitivity to ABA and, thus, resistance to PHS.

Development and validation of a Viviparous-1 STS marker for preharvest sprouting tolerance.

Preharvest sprouting of wheat reduces the quality of wheat grain, and improving PHS tolerance is a priority in certain wheat growing regions where environments favor happens of PHS. Two new *Viviparous-1* allelic variants related to PHS tolerance were explored on B genome of bread wheat and designated as *Vp-1Bb* and *Vp-1Bc*. Sequence analysis showed that *Vp-1Bb* and *Vp-1Bc* had an insertion of 193 bp and a deletion of a 83-bp fragment, respectively, which are located in the third intron region of the *Vp-1B* gene. The insertion and deletion affected the expression level of Vp1 at the mature seed stage. More correctly spliced transcripts were observed from the genotypes with either insertion or deletion than that of the wild type. Based on these insertions and deletions, a codominant STS marker of *Vp-1B* gene was developed and designated as Vp1B3, which in most cases could amplify an 845-bp or a 569-bp fragment from the tolerant cultivars and 652 bp from the susceptible ones. This Vp1B3 marker was mapped to chromosome 3BL using a set of Chinese Spring NT and Dt lines. A total of 89 white-grained, Chinese wheat cultivars and advanced lines were used to validate the relationship between the polymorphic fragments of Vp1B3 and PHS tolerance. Statistical analysis indicated that Vp1B3 was strongly associated with PHS tolerance in this set of Chinese germ plasm, suggesting that Vp1B3 could be used as an efficient and reliable codominant marker in the evaluation of wheat germ plasm for PHS tolerance and marker-assisted breeding for PHS tolerant cultivars.

Characterization of CIMMYT bread wheats for high- and low-molecular-weight glutenin subunits and other quality-related genes with SDS-PAGE, RP-HPLC, and molecular markers.

Two hundred seventy-three CIMMYT bread wheat cultivars and advanced lines were investigated with gene-specific markers for PPO, phytoene synthase (Psy), and waxy genes. We tested 142 lines with SDS-PAGE, RP-HPLC, and molecular markers for the characterization of HMW-glutenin and LMW-glutenin subunits. The over-expression of Bx7 (Bx7OE) and subunit By8* were detected by RP-HPLC. Quality parameters for SDS-sedimentation volume (SDS-SV), flour protein content, mixing time, and Alveograph parameters, such as W and P/L, were investigated in the quality laboratory at CIMMYT. Results showed that in the 273 lines tested by marker PPO18 the frequencies of alleles *PPO-A1a* and *PPO-A1b* were 79.1% and 20.2%, respectively, and no PCR fragment was amplified in two lines (3.5%), whereas 227 lines (83.2%) contained the allele *Ppo-D1a* and 46 lines (16.8%) had *Ppo-D1b* detected by markers PPO16 and PPO29, respectively. In the test with the marker YP7A, 142 lines (52.0%) were assumed to have the allele *Psy-A1a* and 131 lines (48.0%) contained the allele *Psy-A1b*. Using the marker YP7B for the gene *Psy-B1*, the alleles *Psy-B1a* and *Psy-B1b* were detected in 55 (56.8%) and 43 (15.8%) lines, respectively, and 75 (27.5%) lines possessed the allele *Psy-B1d* detected by the marker YP7B-3. All of 273 lines contained the alleles *Wx-A1a* and *Wx-D1a* tested by the markers MAG264 and MAG269, respectively. Using the marker Wx-B1, 204 lines (74.7%) were assumed to have the *Wx-B1a* allele and 69 (25.3%) possessed the allele *Wx-B1b*. The lines with subunits Ax2*, By8, By9, Bx17, Bx20, Dx5, and Glu-B3j were 90, 16, 57, 5, 46, 118, and 33, respectively, in the 142 lines detected by molecular markers. They were consistent with the results tested with SDS-PAGE, except that one line with the T1A·1R translocation and SDS-PAGE could not discriminate the subunits By8 and By8*. Eight lines with Bx7OE were determined by RP-HPLC. Subunits Ax1 and Ax2* at the *Glu-A1* locus showed significantly better effect on all quality parameters than the subunit Null. The possession of subunits 17+18 and 7+8 at the *Glu-B1* locus showed superior value than subunits 7 and 20 on SDS-SV and Alveograph W. Subunits 5+10 represented significantly better effects for all parameters. Subunit Glu-A3b showed more positive effects than its counterpart allelic variation on SDS-SV and SDS-sedimentation volume/protein content index (SPI) at the *Glu-A3* locus. The allele *Glu-B3g* showed the best effect on quality parameters SDS-SV and Alveograph W, whereas *Glu-B3j*, associated with T1B·1R translation, exhibited a strongly negative effect on all quality parameters.

Development of two multiplex PCR assays targeting improvement of bread-making and noodle qualities in common wheat.

Wheat quality properties are genetically determined by the compositions of high- and low-molecular-weight glutenin subunits, grain hardness, PPO activity, and starch viscosity. Two multiplex PCR assays were developed and validated using 70 cultivars and advanced lines from Chinese autumn-sown wheat regions. Multiplex PCR I includes molecular markers for genes/loci α-secalin, *Glu-B1-2a* (By8), *Glu-D1-1d* (Dx5), *Glu-A3d*, *Glu-B3* (for non T1B·1R type), and *Pinb-D1b* targeting improved gluten parameters and pan bread quality. Multiplex PCR II comprises markers for genes/loci *Ppo-A1*, *Ppo-D1*, and *Wx-B1b* targeting improved noodle quality. The results were consistent with those achieved by SDS-PAGE and RP-HPLC, indicating that the two multiplex assays were highly effective, with good repeatability and low costs enabling their use in wheat-breeding programs. In total, nine alleles (subunits) at locus *Glu-B1*, four at *Glu-D1*, and five at *Glu-A3* locus were identified, and the alleles (subunits) *Glu-B1b* (7+8), *Glu-B1c* (7+9), *Glu-D1a* (2+12), *Glu-D1d* (5+10), *Glu-A3a*, *Glu-A3c*, and *Glu-A3d* were most frequently present in the cultivars and lines tested. The T1B·1R translocation was present in 28 (40.0%) lines, whereas the *Wx-B1* null allele for better noodle quality was present in only seven (10.0%) cultivars and advanced lines, and 37 (52.9%) lines had *Pinb-D1b* associated with hard grains. The allele *Ppo-A1b* on chromosome 2AL associated with lower PPO activity was present in 38 (54.3%) genotypes, whereas the less effective allele *Ppo-D1a* on chromosome 2DL, also associated with low PPO activity was present in 45 (64.3%) of genotypes. These two multiplex PCR assays should be effective in marker assisted selection targeting improved pan bread making and noodle qualities.

Allelic variation at the vernalization genes Vrn-A1, Vrn-B1, Vrn-D1, and Vrn-B3 in Chinese wheat cultivars and their association with growth habit.

Information on the distribution of vernalization genes and their association with growth habit is crucial to understand the adaptability of wheat cultivars to different environments. In this study, 278 Chinese wheat cultivars were characterized with molecular markers for the vernalization genes *Vrn-A1*, *Vrn-B1*, *Vrn-D1*, and *Vrn-B3*. Heading time of the cultivars was evaluated in a greenhouse under long days without vernalization. The dominant *Vrn-D1* allele showed the highest frequency in the Chinese wheat cultivars (37.8%), followed by the dominant *Vrn-A1*, *Vrn-B1*, and *Vrn -B3* alleles. Ninety-two winter cultivars carried recessive alleles of all four vernalization loci, whereas 172 spring genotypes contained at least one dominant *Vrn* allele. All cultivars released in the North China Plain Winter Wheat Zone were winter type. Winter (53.0%), spring (36.1%), and early heading (10.9%) cultivars were grown in the Yellow and Huai River Valley Winter Zone. Most of the spring genotypes from this zone carried only the dominant *Vrn-D1* allele, which was also predominant (64.1%) in the Middle and Lower Yangtze Valley Winter Zone and Southwestern Winter Wheat Zone. In three spring-sown wheat zones, all cultivars were early heading spring types that frequently possessed the strongest dominant *Vrn-A1a* allele, and combinations with other dominant *Vrn* gene (s). The *Vrn-D1* allele is associated to the latest heading time, *Vrn-A1* the earliest and *Vrn-B1* intermediate values. The information is useful for understanding the adaptation of Chinese wheat cultivars, and also important for breeding programs in other countries with an interest in using Chinese wheats.

Development of a STS marker specific to Yr26 conferring resistance to wheat stripe rust using the resistance gene-analog polymorphism (RGAP) technique.

The gene *Yr26* confers resistance to all races of *P. striiformis* f. sp. *tritici*. Here, we report development of the molecular markers specific to *Yr26* using a resistance gene-analog polymorphism (RGAP) technique. A total of 787 F_2 plants derived from the cross between resistant cultivar Chuanmai 42 and susceptible line Taichang 29 were used for linkage analysis. Eighteen NILs, 18 Chinese wheat cultivars, and advanced lines with different stripe rust-resistance genes were employed for the validation of the STS markers. In all, 1,711 RGAP primer combinations were chosen to test two parents and the resistant and susceptible bulks. Five polymorphic RGAP markers were used for genotyping the F_2 plants. Linkage analysis showed that the five RGAP markers were closely linked to *Yr26* with genetic distances ranging from 0.5 to 2.9 cM. These markers were then successfully converted into STS markers, of which CYS-5, with the genetic distance of 0.5 cM distal to *Yr26*, was specific to the resistance gene in the validation of 18 NILs and 18 Chinese wheat cultivars and lines. The results indicated that CYS-5 can be used in a MAS program targeting for the pyramiding of *Yr26* with other resistance genes to wheat stripe rust.

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