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ITEMS FROM INDIA

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Molecular breeding, induced mutagenesis, and transcriptome analysis in wheat.

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Deployment of molecular markers for the improvement of some important quality traits and drought tolerance in bread wheat. We are attempting to pyramid one or more QTL/genes for grain quality traits along with the *Lr24* gene for leaf rust resistance into a number of high-yielding Indian wheat cultivars with a view to develop wheat cultivars with improved grain protein content (GPC), leaf rust resistance, and preharvest sprouting tolerance (PHST) using marker-assisted selection (MAS). QTL for grain yield under water stress are also being introgressed into high-yielding Indian wheat cultivars, which are otherwise drought sensitive. The progress made in relation to the above objectives is summarized below.

MAS for grain protein content (GPC) and leaf rust resistance. To introgress two genes, one for high GPC (*Gpc-B1*) and the other for leaf rust resistance (*Lr24*), two high-yielding recipient Indian bread wheat cultivars (Lok1 and HD2967) were crossed with a donor genotype PBW343 (*Gpc-B1+Lr24*). The initial crosses were made at the Research Farm, CCSU, Meerut, during 2009–10 crop season. The two resulting F_1s were grown and backcrossed with the respective recipient genotype in an off-season nursery at the Keylong Research Station during the summer of 2010. Seeds of two BC_1F_1s were planted at Research Farm CCSU, Meerut, during 2010–11. Foreground MAS in the two BC_1F_1s was carried out in two steps; the two BC_1F_1 populations included 938 plants for the recipient parents Lok1 and 1,015 plants for the recipient parent HD2967. These BC_1F_1 plants were screened using a SSR marker (ucw108) specific to the *Gpc-B1* gene for high GPC. As a result, 340 plants involving Lok1 and 149 plants involving HD2967 were selected from the two backcross populations. Foreground selection for leaf rust resistance gene *Lr24* was carried out on the GPC positive plants using SCAR marker SCS73719; 147 BC_1F_1 plants involving Lok1 and 39 BC_1F_1 plants involving HD2967 each carried both the *Gpc-B1* and *Lr24* genes, and were selected for background selection.

For background selection, a set of 127 SSRs were used for plants involving Lok1 and 118 SSRs were used for plants involving HD2967. The SSR markers for background selection were chosen such that these were evenly distributed on all the 21 wheat chromosomes. In this manner, eight BC_1F_1 plants (involving Lok1) with >80% RPG recovery were selected and these plants were backcrossed with the recipient genotype Lok1 again to obtain BC_2F_1 seeds. However, no BC_1F_1 plant involving HD2967 could be selected for further backcrossing, because each of the selected plants contained donor parent allele in homozygous condition for one or more SSR markers. This observation was unexpected and could not be explained without further studies, which we propose to undertake in future.

The BC_2F_1 population involving Lok1 was raised in an off-season nursery at Keylong Research Station during 2011. Foreground selection for *Gpc-B1* and *Lr24* was in a set of 248 BC_2F_1 plants; 43 positive plants with the two genes were selected. Background selection of these 43 plants with the help of SSR markers that were in heterozygous state in BC_1F_1 plants led to the selection of eight plants with >93% RPG recovery, and BC_2F_1 seed was harvested. Eight BC_2F_2 progenies were planted during the 2011–12 crop season at the Research Farm at CCSU, Meerut. Out of 380 BC_2F_2

plants, 57 positive plants with both the genes were selected. The BC_2F_3 progenies of the selected plants are currently (2012) growing in off-season at Keylong Research Station. Foreground selection will be carried out in the above progenies, to identify progenies, which carry *Gpc-B1* and *Lr24* genes in homozygous condition. The selected progenies will be evaluated in BC_2F_4 during the crop season 2012–13 at the Research CCSU, Meerut.

MAS for preharvest sprouting tolerance (PHST). With a view to develop PHS-tolerant, white-grained, wheat genotypes, two Indian wheat cultivars that have white grain but are susceptible to PHS (Lok1 and HD2967) were crossed as recipients with the white-grained, PHS-tolerant, wheat genotype AUS1408 as the donor parent. The crosses were made at the CCSU Research Farm, Meerut, during the 2010–11 crop season.The two F_1s were raised in off-season at Keylong Research Station during 2011, and these F_1s were backcrossed with their respective recipient parent genotypes. The crossed seed was harvested and used for raising two BC_1F_1 populations at the Research Farm, CCSU, Meerut, during the 2011–12 crop season. These two BC_1F_1 populations, one involving Lok1 (216 plants) and the other involving HD2967 (175 plants) were subjected to foreground MAS for the QTL for PHST located on chromosome arm 4AL. In the population involving Lok1, foreground MAS used SSR markers gwm397 and barc170 flanking the QTL for PHST. Foreground selection in the second population involving HD2967 used SSR markers wmc468 and wmc420 flanking the PHST QTL in a genomic region of 31 cM (other 20 SSR markers mapped in the genomic region of the QTL were monomorphic between the two parental genotypes). As many as 78 BC_1F_1 plants involving Lok1 and 43 BC_1F_1 plants involving HD2967 carrying the QTL for PHST in heterozygous condition were selected in the two BC_1F_1 populations. Background selection of 78 positive plants of BC_1F_1 population involving Lok1 used 130 SSRs and 10 BC_1F_1 plants with >75% (range = 75-82.5%) RPG recovery was selected. In the second BC_1F_1 population involving HD2967, all the 43 plants were backcrossed to the recipient parent and also were subjected to phenotypic selection. Eventually, the BC_2F_1 seed of four plants showing higher level of PHS tolerance were harvested.

MAS for grain weight (GW). Introgression of two QTL for high GW (*QGw.ccsu-1A.1* and *QGw.ccsu-1A.3*) from the high grain weight (54.28 g) donor genotype RS111 to two low grain weight recipient genotypes Raj3765 (41.56 g) and K9107 (44.0 g) was attempted by crossing the donor and recipient genotypes. Using the F_1s, one to three backcrosses were attempted with each of the recipient genotypes. Foreground selection in each backcross population was exercised using two SSR markers (wmc59 and wmc24) associated with two QTL for GW on chromosome 1A (Mir et al. 2012). Following foreground selection, 13 BC_3F_2 plants (seven in the Raj3765 background and six in the K9107 background) with significantly higher GW (53.68g to 61.44g) than even that of the donor parent (RS111) were recovered suggesting successful application of MAS in improvement of GW in bread wheat. Moreover, 2.24–17.00% higher GW of the selected progenies over the donor parent further suggested that the two introgressed QTL also had a complimentary interaction with the genotypes of the recurrent parents suggesting the importance of the two QTL for GW in marker-assisted breeding for higher GW. The BC_3F_3 progenies derived from the selected 13 BC_3F_2 plants will be tested for yield and yield-related traits in replicated trials with a view to identify high yielding wheat progenies with bold grains.

MAS for drought tolerance. Drought stress continues to be a major abiotic stress having adverse effect on the productivity of wheat crop. We anticipate that with climate change, drought will impact many more wheat-growing areas. We recently initiated a MAS program to develop drought-tolerant, prebreeding, wheat material. For this purpose, we are introgressing QTL for drought tolerance in cultivars Dharwad Dry (wmc89 on 4AL) and SQ1 (wmc273.3 on 7AL) (for details, see Quarrie et al. 2005 and Kirigwi et al. 2007) into five high-yielding. drought-sensitive, wheat cultivars. Marker validation for the above QTL used contrasting genotypes for drought tolerance. For the purpose of introgression of the desired QTL using MAS, a total of 10 crosses were attempted between drought-tolerant, donor genotypes (SQ1 and Dharwad Dry) and high-yielding, drought-sensitive, recipient cultivars (HD2967, HUW234, HUW468, DBW17, and K307) at the Research Farm, CCSU, Meerut, during 2011–12 crop season. The resulting 10 F_1s were grown and backcrossed with the respective recipient genotypes in off-season nursery at Keylong Research Station during 2012. In the BC_1F_2 generation, followed by foreground selection, we will estimate the effect of QTL alleles using a bulked segregation analysis to validate the desirable QTL alleles contributed by the two donor genotypes. In case of a validated QTL, foreground MAS, in combination with phenotypic selection will be carried out in the BC_2F_2 populations for selection of drought-tolerant genotypes. Phenotypic selection will be continued in the subsequent generations with the hope that the program would eventually lead to development of drought-tolerant, prebreeding material for wheat breeding.

Marker-assisted pyramiding of genes/QTL for grain quality traits and leaf rust resistance in the background of PBW343. Pyramiding seven QTL/genes including four QTL/genes for grain quality traits and one gene each for leaf, stem, and yellow rust resistance (*Lr24*, *Sr24*, and *Yr36*) in the background of widely grown wheat cultivar PBW343 was undertaken using four genetic stocks developed through MAS by us at our research farms at CCSU, Meerut, and Punjab

Agricultural University (PAU), Ludhiana, India. Using these genetic stocks, two single-cross hybrids were produced; one PBW343 (*Gpc-B1+Lr24+Sr24+Yr36*)/PBW343 (*QPhs.ccsu-3A.1)*) was produced by at Meerut and the other, PBW343 (*Lr24+QGw.ccsu-1A.3+Sr24*)/PBW343 (*Glu-A1*)) was produced at PAU. Using the F_1 seeds of these two single-cross hybrids, our two institutes jointly produced double-cross hybrids during the off-season at the Keylong Research Station during 2009 for MAS-aided pyramiding of the following genes/QTL: (i) *Gpc-B1* for high GPC, (ii) a QTL for high grain weight (*QGw.ccsu-1A.3*), (iii) a QTL for PHS tolerance (*QPhs.ccsu-3A.1*), (vi) a high-molecular-weight (HMW) glutenin (*Glu-A1*) gene, (v) leaf rust resistance gene *Lr24*, (vi) stem rust resistance gene *Sr24*, and (vii) yellow rust resistance gene *Yr36*.

The double-cross F_1 ($DCHF_1$) population comprising 192 plants was grown at the Research Farm, CCSU, Meerut, during 2009–10 crop season and foreground selection used a set SSR/SCAR markers linked to corresponding genes/QTL for GPC, PHST, GW, and leaf, stem, and strip rust. Following foreground selection, four plants carrying all the above QTL/genes in heterozygous condition were selected and self-pollinated by bagging. Progenies of the four selected DCHF1 plants were raised in off-season at Keylong during 2010. A total of 208 DCHF2 plants belonging to the above selected four $DCHF_1$ plants were subjected to foreground selection for the six QTL/genes resulting into selection of 16 $DCHF_2$ plants carrying all six QTL/genes (one or two of the selected QTL/gene(s) still in heterozygous state). Harvested seeds of 16 $DCHF_2$ plants were analyzed using half seed (endosperm) for HMW-glutenin gene *Glu-A1* using SDS PAGE at PAU, Ludhiana, during the 2010–11 crop season. Nineteen (19) seeds belonging to four $DCHF_2$ plants were positive for *Glu-A1*. The remaining half seed (embryo) of the 19 positive seeds were planted at PAU, Ludhiana, during the 2010–11 crop-season. DNA from the 19 positive $DCHF_3$ plants was extracted and foreground selection for other six desired QTL/genes was carried out at CCSU Meerut to select plants with all seven pyramided QTL/genes; three $DCHF_3$ plants with all seven desired QTL/genes were selected (QTL for PHST and GW were in homozygous state; the homozygous/heterozygous state of the genes for GPC and leaf and stripe rust, and glutenin could not be determined because we used dominant markers for these genes). The $DCHF_4$ progenies of three selected plants were sown at Keylong in the off-season during 2011, and foreground selection for seven genes was carried out. Of the three progenies, 36 plants, belonging to a solitary progeny, were nonsegregating for five QTL/genes (*Gpc-B1*, *QPhs.ccsu-3A.1*, *QGw.ccsu-1A.3*, *Yr36*, and *Glu-A1*). Of these 36 plants, 17 were positive for *Lr24* and *Sr24* and four of these 17 plants were homozygous for *Sr24*. $DCHF_5$ seed of the above four $DCHF_4$ plants was harvested. Fifty (50) $DCHF_5$ seeds from each $DCHF_4$ plant were sown in five rows during 2011–12 crop season at the Research Farm, CCSU, Meerut. Foreground selection was conducted in the four $DCHF_5$ progenies, and all plants had all the seven QTL/genes in homozygous condition. The $DCHF_6$ progenies of individual plants of the above four $DCHF_5$ progenies are currently growing in off-season at Keylong Research Station for seed multiplication. Phenotypic evaluation of the $DCHF_7$ progenies will be carried out at Research Farm, CCSU, Meerut, during the 2012–13 crop season.

Meta-analysis for PHST and related traits in bread wheat. A meta-analysis approach, proposed by Goffinet and Gerber (2000), was conducted using Biomercator 2.1 to estimate real number of stable QTL (referred as meta-QTL (MQTL)) for PHST. The analysis was based on the most likely model and minimizing Akaiki criterion (AIC value). The study was restricted to only four chromosomes, 3A, 3B, 3D, and 4A, because QTL for PHST and dormancy on these four chromosomes were reported in several earlier studies. Information on 64 original QTL was collected from 24 individual studies on PHST and its related traits (dormancy, germination index (GI), sprouting index (SI), visible index (VI), and seed coat color (Cl)). As many as 50 original QTL with reliable information were projected onto a consensus map that was created by integrating two highly saturated wheat maps, one with 1,235 markers (Somers et al 2004) and the other with 4,506 markers (wheat composite 2004 map). As a result, eight MQTL with narrow confidence intervals and comprising 37 original QTL were identified. Of the eight MQTL, seven MQTL were located on group-3 chromosomes (3A (2 MQTL), 3B (3 MQTL), and 3D (2 MQTL)) and 1 MQTL was located on chromosome 4A. MQTL1 (chromosome 3A), MQTL 2 (chromosome 3A), MQTL6 (chromosome 3D), and MQTL8 (chromosome 4A)) may be pleiotropic, because these MQTL mapped at the clusters of original QTL for PHST and other traits. However, two MQTL, i.e., MQTL3 and MQTL4 located on chromosome 3B, mapped at the clusters of original QTL for dormancy and PHST respectively, suggesting that these two are independent QTL. Two additional MQTL5 and MQTL7 were such in which original QTL for PHST-related traits (GI, SI, Cl, and VI) were clustered. We note that at least one of the flanking SSR markers for each of the eight MQTL also was reported as the closest marker to QTL reported in one or more of the original studies. We believe that introgression of MQTL for PHST by MAS in an adaptive genetic background may allow development of PHS-tolerant, wheat genotypes.

In conclusion, our study suggested that whenever hundreds of QTL based on a large number of studies involving a number of different mapping populations are available in the published literature, meta-QTL analysis can be a use-

ful reductionist approach to allow identification of only genuine and real QTL for further study and possible use in MAS. The approach also allows narrowing down the confidence interval) for each QTL and overcomes a part of the difficulty and confusion that often exists due to redundancy in the number of QTL in overlapping genomic regions.

Association mapping for PHS tolerance in bread wheat. PHS is a complex trait governed by several QTL. In bread wheat, all 21 chromosomes are known to carry QTL for PHS tolerance, although the group-3 chromosomes and chromosome 4A are more important. In all the studies conducted earlier to understand genetic architecture of this complex trait, mostly bi-parental populations were utilized, and QTL were identified through linkage analysis. Using bi-parental populations for linkage mapping has certain limitations. Therefore, we carried out association mapping for PHS tolerance using a set of 242 genotypes (230 Indian wheat cultivars and 12 exotic accessions) of wheat to overcome these limitations. The genotypes were assayed using 250 SSR markers and phenotyped for PHS tolerance for 2–4 years on a 1–9 scale, where a score of 1 represents complete tolerance and a score of 9 represents complete susceptibility.

Model-based cluster analysis was performed with the help of STRUCTURE 2.3.3 to infer population structure and to define the number of clusters. This analysis revealed that there were 15 sub-populations in the above set of wheat genotypes. Marker-trait associations (MTAs) were determined using general linear model (GLM) and mixed linear model (MLM) approaches with the help of TASSEL and using a P-value of ≤ 0.05 for determining the significant of MTAs. As many as 30 markers showed association with PHS. These markers were located on 17 different chromosomes (1A, 1B, 2A, 2B, 2D, 3B, 3D, 4A, 4B, 4D, 5B, 5D, 6B, 6D, 7A, 7B, and 7D). Of these 30 markers, eight (gwm425, wmc827, gwm 526, wmc418, gwm131, cfd2, gwm251, and gwm325) were present in the five different genomic regions already known to carry QTL for PHST (Mohan et al. 2009; Fofana et al. 2009; Kulwal et al. 2005). The remaining 22 SSRs could not be associated with any of the QTL reported earlier. Although QTL for PHST perhaps occur everywhere in the genome, these need validation using one or more of the following approaches: linkage analysis, joint linkage analysis, or joint linkage-LD mapping. The application of false discovery rate criteria showed that only three MTAs (*Xgwm16*, *Xcfd16*, and *Xwmc526*) were true, and the remaining 27 MTAs for PHST could be considered only as putative. The information generated in the present study may be used for marker-assisted selection and fine mapping of one or more QTL for PHST.

Induced mutagenesis in wheat. Induced mutation research has witnessed a renewed interest in recent years due to its increasing role in the area of functional genomics. Keeping this in view, we started the induction of mutations in wheat. For this purpose, we treated the seeds of the wheat cultivars PBW550 and Indian with two chemical mutagens; PBW550 was treated with different doses of N-ethyl-N-nitrosourea (ENU) and ethyl methane sulphonate (EMS) and Indian was treated with ENU only.

In the PBW550 M_2 population, following treatment with 2mM ENU, we identified eight different morphological mutants. These mutants were validated in M_3 generation and classified into four classes, (i) stem mutants (axillary branching, reduced number of nodes, and dwarf mutants), (ii) leaf mutants (early senescence of leaf mutant), (iii) spike mutants (spike with altered morphology, including fusiform ear, clubbed shaped ear, and bent peduncle mutants), and (iv) reproductive mutants (nonflowering mutants). Mutants with axillary branching and a reduced number of nodes (1–2) are being reported for the first time in wheat.

In silico studies were conducted on the above two novel mutants using candidate genes reported to produce these mutant phenotypes in other species including rice, maize, and *Arabidopsis*. For the axillary branching mutant, we identified six genomic sequences in wheat that were considered orthologous to the sequences for branching genes of rice and maize (Auldridge et al. 2006; Ishikawa et al. 2005; Takeda et al. 2003; Zou et al. 2006). Similarly, for the mutant with a reduced number of nodes (1–2), we identified 11 unigenes using the EST database, which matched the genes responsible for reduction in number of nodes in maize, each showing >90% similarity. These sequences may prove as important candidates for above mutant phenotypes in bread wheat.

In order to dissect the genetic control of the mutant phenotypes, the mutants were crossed with wheat genotypes with wild-type phenotypes. The axillary branching and early senescence mutants were crossed with wheat cultivar Lok1, whereas the reduced number of nodes (1–2) and dwarf mutants were crossed with the tall cultivars NP114 and K-68. The F_1 hybrids were raised in an off-season nursery at Keylong Research Station during 2011 and the F_2s were grown in the 2011–12 crop season at the Research Farm, CCSU, Meerut. The F_2s and are being subjected to a bulked-segregant analysis to identify marker(s) linked to the mutant trait in all the six populations.

A MAGIC (mutant-assisted gene identification and characterization) program was initiated to identify modifiers/QTL responsible for reduced number of nodes (1–2) in the mutant. During the 2011–12 crop season, the mutant was crossed with 23 different genotypes from a highly diverse germplasm. All 23 F_1 hybrid plants were raised during off-season at Keylong Research Station during 2012 to obtain F_2 seeds. This F_2 seed will be used for raising 23 F_2 populations during the 2012–13 crop season at the Research Farm, CCSU, Meerut, for screening of modifiers affecting the expression of mutant phenotypes.

Construction of integrated physical map using simple sequence repeats (SSRs). Physical mapping of DNA-based markers in wheat has been greatly facilitated due to the availability of deletion stocks, which constitute an ideal resource for mapping these markers to specific chromosomal regions or bins to create physical landmarks. An integrated physical map of bread wheat comprising as many as 1,903 SSR loci was made available earlier in our laboratory (Gupta et al. 2008). During the present study, we integrated a new set of 128 SSR loci into the above map, resulting into a physical map containing 2,031 SSR loci.

The 2,031 SSR loci in the integrated map were distributed throughout the wheat genome giving an average resolution of 7.8 Mb between the two SSR loci. A maximum of 765 loci (37.67%) were mapped in the B genome, 651 loci (32.05%) in the D genome, and 615 loci (30.28%) in the A genome. Using 704 SSR loci that were mapped genetically as well as physically, a comparison was made between genetic and physical maps to determine the distribution of recombination frequencies (cM/Mb) in different regions of the wheat genome. Recombination frequencies within the individual bins ranged from 0.01 cM/Mb (low recombination) to 13.16 cM/Mb (high recombination), suggesting an uneven distribution along the chromosomes or chromosome arms. Hopefully, the integrated physical map that was prepared may prove useful in the currently on-going whole genome sequencing of wheat genome through alignment of BAC contigs. A comparison of integrated physical map with genetic linkage maps will also facilitate the on-going and future genomics research.

Transcriptome analysis in a leaf rust resistant wheat stock CSP44 carrying Lr48 gene for adult plant resistance. In wheat, global gene expression analysis was carried out to study adult-plant resistance (APR) to leaf rust conferred by *Lr48* gene carried by wheat cultivar CSP44. Plants of CSP44 were separately inoculated with leaf rust race 77-5 and also mock inoculated (as control). The two sets of plants were used for cDNA-AFLP and qRT-PCR analysis to identify differentially expressed transcripts during incompatible interaction at the adult-plant stage. A total of 298 transcripts expressed differentially following leaf rust inoculation were identified, which included 262 transcripts that were up-regulated and 36 transcripts that were down-regulated. Fifty-two transcripts, which included 44 up-regulated and eight down-regulated transcripts, were eluted from gels. Forty-eight (40 up-regulated and eight down-regulated) transcripts were successfully re-amplified, cloned, and sequenced. Thirty-eight of these 48 transcripts had homology with known genes that are involved in functions like metabolism, energy production, signal transduction, transcription, translation, photosynthesis, transport, development, DNA integration, and disease resistance. The functions of remaining 10 transcripts could not be determined. Apparently, thesetranscripts represented some novel genes reported for the first time in wheat. Specific primers were designed for 18 of the 48 transcripts. These 18 transcripts representing genes with putative and unknown functions were used for qRT-PCR analysis. A majority (15) of these transcripts were induced within 24 h with maximum expression achieved at 96 h in leaf rust inoculated APR stock CSP44. Of the 15 transcripts, 13 showed down regulation during-compatible interaction (susceptible) at the seedling stage and up-regulation during incompatible (resistance) interaction at the adult-plant stage, whereas the two remaining transcripts showed abundant expression at both seedling and adult-plant stages. The expression of the remaining three transcripts did not show any change following inoculation with leaf rust race 77-5. Together the above results indicated that two of the up-regulated 15 transcripts, one each encoding a regulatory protein MarR, and multidrug resistance-associated protein MRP1 (*T. aestivum*) may be responsible for leaf rust resistance in wheat.

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Response to selection for grain and biological yield in early segregating generations of spring x winter wheat.

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The wheat grown in India is of the spring type mainly guided by agro-geographic factors. Winter wheats possess a wide diversity for yield and other traits associated with high yield *per se* and resistance to major biotic and abiotic stresses. In order to enhance the productivity of spring wheats, winter types can be a great source of variability. Because there is no crossability barrier between the two ecotypes, hybridization is easily carried out by synchronizing the flowering time.

The higher productivity associated with winter wheats is due to the larger quantity of biomass they produce. Because the major area under wheat worldwide is occupied by spring wheats, one way to obtain yield enhancement in spring wheat is through introgressing traits for higher biomass production from winter wheats. CIMMYT, Mexico, has successfully utilized 'winter x spring' hybridization for yield enhancement in spring wheats. Furthermore, the recent spring wheat cultivars, which have been released in different countries have higher biomass in comparison to the cultivars of the 1980s and 1990s.

Two, exotic, winter wheat lines (EC 609401 and EC 609402) obtained from the Germplasm Unit at the Directorate of Wheat Research (DWR), Karnal, India, were sown in first week of October, 2010, to synchronize heading with early maturing, spring wheat cultivars that were sown in late December. These winter wheat lines were used as female parents in crosses with the advanced spring wheat lines PBW658, HD3059, WH1125, PBW672, DBW71, and DBW92 to generate F_1s. Five spikes were used as the female parent for each cross. Cross seed was grown in an open field during the winter of 2011 at Karnal and the seed harvested from the F_1 plants was divided into two lots; half was planted in an off-season nursery located at DWR Regional Station, Dalang Maidan (Himachal Pradesh), during May 2012. From the

F_2 plants grown during the off season, only spring type plants were selected. The harvested seed was bulked for generation advancement. During the 2012–13 crop season, seed of the previous season's F_1 plants (F_2 generation seed) and the bulked F_2 seed obtained from off season nursery (corresponding to F_3 generation seed) were planted together. In all, eight F_2s and eight F_3s were space planted at Karnal, in plots of six 5-m rows with row spacing of 20 cm. The seed was placed 5 cm apart in all rows of each plot. All recommended agronomic practices were adopted to raise the crop.

Biological yield (kg/plot) was measured as the total aerial biomass on dry weight basis produced for the inner four rows plus the biomass of individual plants selected in each cross. The grain yield (gm/plot) of the whole plot and selected plants was measured after threshing. The response to selection was determined as the difference between the phenotypic value for the traits biological yield and grain yield in selected population (F_3) and base population (F_2).

The realized gain under selection varied from –1.18 kg (EC609401/PBW658) to 2.95 kg (EC609402/DBW71) for biological yield (Table 1). For grain yield, response to selection ranged from –589.60 g (EC609401/WH1125) to 1,025.00 g (EC609402/PBW672). All eight crosses except one (EC609401/PBW658) showed a positive response to selection for biological yield. In most of the crosses, the positive response for biological yield was associated with a negative response with grain yield. The negative response in grain yield in crosses may be due to rejection of winter types in the F_2 generation in the segregating population grown at off season. However, in segregating populations of 'EC609402/PBW672' and 'EC609402/DBW71', we observed a positive response to selection for both biological and economic yield.

These populations will further be subjected to selection pressure for spring types and, hence, used to study the response to selection in advanced generations. We concluded that biological yield could be used as selection criterion in early filial generations in winter x spring hybridization programs.

Table 1. Response to selection for biological (kg/plot) and grain yield (g/plot) in early segregating generations of spring x winter wheat.

	Biological yield			Grain yield		
Pedigree	F_2	F_3	Response to selection	F_2	F_3	Response to selection
EC609401/PBW658	6.42	5.24	–1.18	821.4	1,692.7	871.3
EC609401/HD3059	6.30	7.02	0.72	2,117.2	1,707.7	–409.5
EC609401/WH1125	4.56	6.84	2.28	1,636.3	1,046.7	–589.6
EC609402/PBW672	5.24	5.77	0.53	1,581.4	2,606.4	1,025.0
EC609402/DBW71	4.68	7.63	2.95	1,472.5	1,737.0	264.5
EC609402/HD3059	6.74	6.78	0.04	2,299.5	1,781.4	–518.1
EC609402/PBW658	6.00	6.33	0.33	1,876.1	1,787.4	–88.7

Increasing water productivity through matriconditioning and seed sprouting in wheat.

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The wheat crop is mainly grown under irrigated conditions. Future projections estimate that by 2050 per capita water availability will decrease to 1,190 M^3/year from present 1,545 M^3/year. In India, 90 % area of the wheat is irrigated, which also includes the area with restricted irrigation; only 10% of the area is rain-fed. In the future, the major challenge will be to produce more food with less water as share of water for agriculture decreases. Therefore, the biggest challenge is to improve water productivity in existing systems. The present water productivity of wheat is 800–1,000 L water/kg wheat grain, which needs to be decreased in order to improve the water use efficiency. This investigation was undertaken to reduce the presowing irrigation requirement of the wheat crop and understand the effect of matriconditioning (seed priming with distilled water) and seed sprouting on crop establishment under different soil moisture regimes.

Materials and methods. A field experiment was conducted in a split-plot design with three replications during the winter of 2010–11 and 2011–12 at the Directorate of Wheat Research, Karnal (Haryana). The experiment was comprised of three main plot treatments; M1, seeding at optimum moisture level (17.5%), M2, seeding at suboptimal moisture (10.9%), and M3, seeding in dry soil followed by irrigation (6.2%); and three subplot treatments, S1, no seed priming, S2, matriconditioning seed priming, and S3, sprouted seed. Soil moisture content was estimated following a gravimetric

method (Black 1965). Rainfall received at the experimental site was 129.7 mm during 2010–11 and 36.3 mm during 2011–12. The climate is subtropical, with the mean maximum temperature ranging in between 34–39°C in the summer and mean minimum temperature ranging in between 6–7°C in winter.

Matriconditioning was in gunny bags as described by Basra et al. (2003). The gunny bags were soaked overnight in distilled water and then spread on a perforated floor. After leaching excess water from the gunny bags, seed was uniformly layered between them. The gunny bags were kept moist for the whole treatment period, i.e., 12 hours. For sprouting, the required quantity of seed was soaked in water for 10 hours, and then they were spread uniformly on a wet bag and covered with another bag for 12–14 h. The soil in the experimental field was sandy clay loam with at pH 7.9 (1:2.5 soil to water). The soil had an organic carbon content of 0.4 %, available N was 190 kg/ha, P was 17.8 kg/ha, and K ws 165 kg/ha at beginning of the experiment. Recommended doses of fertilizer (150:60:30 kg N, P_2O_5, and K_2O/ha) were applied as part of nutrient management. Full doses of P and K and a one-third dose of N through urea and the NPK mixture (12:32:16) were applied at sowing, and the remaining N was applied equally in two parts at the first and second irrigations. The cultivar DBW17 was sown on 18 November, 2010, and 6 November, 2011. The seed was sown in rows 20 cm apart at a seeding rate of 100 kg/ha. For the treatment in dry soil followed by irrigation, a light irrigation (40 mm) was applied soon after seeding. The first irrigation (60 mm) was applied uniformly to all treatments at the crown root initiation stage (22 days after sowing). Subsequent irrigations were applied at all critical growth stages. Irrigation was done with the help of a water pump. Other management practices were adopted as per recommendations of the crop under irrigated conditions in North Western Plains Zone of India. The number of effective tillers/m^2 from the center of each plot was measured at maturity. Plant height (cm) was recorded by measuring the height of 10 random plants from each plot. A random sample of 10 spikes was taken from each plot to determine spike length at maturity. A net plot of size 6 m^2 was harvested manually to obtain biomass and yield data. Grain was randomly selected from each subplot to calculate 1,000-kernel weight. SAS version 10.3 was used to analyze the observations and differences; means were further grouped into significant classes by Duncan's New Multiple Range Test at P = 0.05.

Results. Sowing sprouted seed sowing produced significantly higher grain yield (54.87 q/ha) compared to matriconditioning (53.01 q/ha) and unprimed seeds (50.24 q/ha) (Table 2.). The effect of seeding method was statistically nonsignificant; seeding at optimum moisture level (52.77 q/ha), seeding at sub optimal moisture level (52.02 q/ha), and seeding in dry soil followed by irrigation (53.32 q/ha). Because seed germination is a major limiting factor in crop establishment under moisture-deficient conditions, matriconditioning (priming with plain water) and sprouted seed improved germination, seedling growth, and crop establishment. Priming with plain water and sprouted seed are simple and inexpensive and can increase crop establishment under suboptimal soil moisture conditions. Priming and sprouting can be successfully used in areas of water deficit. Sowing also can be done without presowing irrigation in the North Western Plains Zone.

Table 2. The effect of seed priming and seeding method on crop establishment, growth, and yield of wheat during crop years 2010–11 and 2011–12 (pooled analysis). Treatments were M1, seeding at optimum moisture levels; M2, seeding at suboptimal soil moisture levels; and M3, seeding in dry soil followed by irrigation. Subtreatments were S1, no seed priming; S2, seed priming; and S3, sowing sprouted seed.

Treatment	No. of tillers/ M2	Spike length (cm)	Plant height (cm)	Biomass (q/ha)	Grain yield (q/ha)	1,000-kernel weight (g)
A. Seeding method						
M1	491.73	10.24	87.76	124.87	52.77	38.88
M2	456.94	10.15	88.37	124.17	52.02	38.42
M3	499.53	9.53	88.01	127.86	53.32	38.77
LSD (P = 0.05)	28.51	NS	NS	0.79	**NS**	**NS**
B. Seed priming						
S1	464.98	9.61	87.84	123.69	50.24	38.32
S2	482.48	10.13	88.34	125.81	53.01	38.67
S3	500.73	10.18	87.95	127.41	54.87	39.08
LSD (P = 0.05)	19.06	NS	NS	1.39	1.76	0.41

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The status of host resistance in wheat cultivars of peninsular and central India against the Nilgiri flora of black and brown rusts.

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The Nilgiri Hills in southern India enjoy weather that favors wheat cultivation throughout the year. The weather here also is congenial for maintaining the three rust pathogens on the cultivated wheat crop, volunteer self-sown host plants, and naturally growing grasses. Wheat has been grown intensively in the Nilgiri Hills until the middle of 20th century, but now is restricted to isolated locations where local farmers cultivate old indigenous cultivars with a religious family sentiment of a crop of their ancestors that must be essentially consumed as a diet. Most of the of farmers in Nilgiris now have turned on to more remunerative vegetables and tea crops. The Regional Research Station of IARI situated in the Wellington Valley of Nilgiris has been growing wheat during winter and summer seasons since its establishment in the early 1950s. The wheat materials planted here by the scientists of various Indian wheat programs for the purpose of generation advancement in summer season or for natural screening against rust diseases. Therefore, wheat grows in Nilgiris continuously through the year, which harbors three rusts *in vivo*. These southern hills of India are still playing an active role in contributing primary inoculum of rusts for causing infection of wheat crop during autumn in central and peninsular India.

In case of leaf rust, race 77-5 (121R63-1) dominated during the period of 2010–12. The other races monitored were 77-7(121R127) and 77-8 (253R31). Frequencies of 77-7(121R127) and 77-8(253R31) being at par differed significantly from the frequency of race 77-5(121R63-1). Only two races of stem rust, 40A(62G29) and 40-1(62G29-1), occurred with equal frequencies. In fact, the urediospores of all three rusts survive in Nilgiris during summer months on the off-season crop/self sown plants. During late October to the end of November, the cyclonic wind circulations formed in the Bay of Bengal, while crossing the Nilgiris, lift the urediospores and carry to peninsular and central India. The accompanying rain washes down the urediospores on 15 to 20 days old wheat crop in peninsular and central India (Nagarajan, 1973 and Nagarajan and Singh, 1975). Rain also provides the needed leaf wetness for infection. In the following two months the pathogen completes several uredo-cycles reaching a high terminal disease severity. Results of surveillance of stem and leaf rusts conducted during 2010, 2011, and 2012, at the IARI Regional Station, Wellington, also corroborated the fact that the Nilgiri Hills act as source of the initial inoculum to the target areas of central and peninsular India covering states of Karnataka, Maharashtra, and Madhya Pradesh. We found that races (77-5 of brown and 40A/40-1 of black rust) dominating in the states of Karnataka, Maharashtra, and Madhya Pradesh constituted the race flora of Nilgiris also. The Nilgiri Hills seems to be the sole source of initial inoculum for these states in case of both black and brown rusts. Races of both the rusts monitored in Himalayas, the another source of initial inoculum for both the rusts could not be traced in states of Karnataka, Maharashtra and Madhya Pradesh except race 77-5(121R63-1), which exists commonly in the Himalayas as well as the Nilgiris.

Applying the avirulence/virulence formulae of races which prevailed upon in Nilgiris, the effectivity of resistance genes present in wheat cultivars released for central and peninsular Indian states was appraised. We note that it is very alarming that all the wheats released so far for cultivation in these states are susceptible to one or more races of leaf rust surviving in Nilgiris, which acts as source of primary inoculum for these states. The *Lr* genes, *Lr10*, *Lr13*, *Lr23*, and *Lr26*, which are commonly available in cultivars of these states are susceptible to all the races existing in Nilgiris. In the present context, cultivars containing gene *Lr24* being resistant to all the races known so far, hold promise of protection from Nilgiri flora of brown rust. However, the singular presence of this gene in few of the cultivars does not benefit their cultivation, especially at inoculum source areas, because it may fall prey to vertical mutability of pathogen bringing in new virulent races. Therefore, to continue harnessing the agronomic benefits of cultivars having gene *Lr24*, we need to fortify their resistance backgrounds with other genes through pyramid breeding. *Lr24* cultivars also need further attention, because *Sr24* is linked to *Lr24* and is infected by the black rust race 40-1(62G29-1) existing in Nilgiris. Pyramiding additional *Sr* genes in such cultivars is needed if envisaged for cultivation in central and peninsular parts of India. Still, another effective gene, *Lr9*, has been reported with virulence of race 77-7 (121R127; Nayar et al. 2003), which was first

traced in Nilgiris only. A population of race 77-7 (121R127) also showed an increasing trend in Nilgiris, therefore releasing cultivars with *Lr9* in central and peninsular India is likely to not serve any useful purpose unless used as a component of gene block with other effective or minor genes pyramided together.

The two races 40A(62G29) and 40-1(62G29-1)of black rust prevailing in Nilgiris with equal frequencies can infect cultivars of central and peninsular India with genes *Sr7b*, *Sr9b*, *Sr9e*, *Sr11*, and *Sr24*. Genotypes possessing these genes, in combination of two or more or in combination with gene *Sr2*, show complete protection from both the races. A majority of the cultivars of central and peninsular zone possess *Sr2*, which is quite desirable for the purpose of preventing black rust epidemics in these zones. The presence of *Sr2* in a majority of cultivars of the Peninsular Zone guarantees aversion of yield losses due to black rust in this zone for the future bacause of the proven durability of this gene. Because of the presence of *Sr2*, rust resistance seems to be stabe in the Central Zone even after 4–5 decades of utilization of cultivars with this gene. *Sr2*, derived from the cultivar Hope, also is responsible for restricting yield losses to only negligible since the late 1960s in southern U.S. This resistance is based on the gene complex *Sr2*, which actually consists of *Sr2* plus 4–5 minor genes pyramided into three to four gene combinations (Rajaram et al. 1988). Because there has been no major black rust epidemics in areas where CIMMYT germ plasm is grown, which may be attributed to *Sr2*, the resistance shows promise to be durable in India also where majority of the cultivars have their origin in CIMMYT germ plasm.

Fortunately, a few cultivars of both central and peninsular India possess gene *Sr31*, which is resistant to both races of black rust monitored in Nilgiris. Therefore, such cultivars show promise of remaining rust free but need to be kept under vigilance, because *Sr31* is susceptible to the stem rust race Ug99 and its variants. Emergence and continued spread of Ug99 races of *P. graminis tritici* now is recognized as a major threat to destabilize wheat production in many countries in its migration path, because most current cultivars are susceptible (Singh et al. 2006). Cultivars such as HI 1531 and DL 788-2, in addition to having *Sr2*, also possess *Lr24*, a gene so far maintaining resistance to all Indian pathotypes of the brown rust pathogen and are capable of providing simultaneous protection from the two rusts. Gene *Lr24* is present in combination with *Lr26* in cultivars such as HW 3094 and HW 5013 of South Hill Zone, which is an inoculum source area, and such a combination may impede the arisal of new races.

A meticulous backcross program is ongoing at the IARI Regional Station, Wellington, which resulted in constitution of black and brown rust resistant genetic stocks. Most of these stocks continuously had adult-plant resistance for five years to black and brown rusts under natural epiphytotics at Wellington (Table 1, pp. 30–31). All these lines have been advanced to the F_9 generation, guaranteeing their genetic stability. Seed can be procured from the IARI Regional

Table 1. Genetic stocks generated through backcrossing for adult-plant resistance to the Nilgiri flora of black and brown rusts (R = resistant, TS = trace susceptibility, MR = moderately resistant, and MS = moderately susceptible) at the Indian Agricultural Research Institute Regional Station, Wellington.

Name	Pedigree	Rust reaction	
		Black	Brown
Lr24 + Sr24 + Sr27			
HW 2091	C 306*3//TR 380-14 *7/3Ag#14/KS [Sr27]	TR	10MS
HW 2093	C 306*3//CS 2A/2M 4/2/KS [Sr27]	TR	10MS
Lr28 + Sr26			
HW 2096	Lok-1*3//CS 2A/2M 4/2/Kite	TS	TR
HW 2099	WH-147*3//CS 2A/2M 4/2/Kite	TS	TR
Lr37 + Sr38 + Yr17			
HW 4022	HD 2285*5/RL 6081	5MR	5MS
HW 4023	HD 2329*5/RL 6081	10MS	5MS
HW 4024	HUW234*5/RL 6081	TR	5MS
HW 4025	Kalyansona*5/RL 6081	5MS	5MS
HW 4026	Lok-1*5/RL 6081	TS	5MS
HW 4028	PBW 226*5/RL 6081	TS	5MS
HW 4029	Sonalika*5/RL 6081	TS	5MS
HW 4030	WH 147*5/RL 6081	R	5MS
HW 4031	WH 542*5/RL 6081	TS	5MS

Table 1. Genetic stocks generated through backcrossing for adult-plant resistance to the Nilgiri flora of black and brown rusts (R = resistant, TS = trace susceptibility, MR = moderately resistant, and MS = moderately susceptible) at the Indian Agricultural Research Institute Regional Station, Wellington.

Name	Pedigree	Rust reaction	
		Black	Brown
***Sr31 + Lr26 + Yr9 + Lr28* (additive effect for yield increase observed)**			
HW 4041	C 306 *2//WH 542/CS 2A/2M 4/2	10MS	10MR
HW 4042	HD 2329*2//WH 542/CS 2A/2M 4/2	10MR	10MR
HW 4043	HUW 234*2//WH 542/CS 2A/2M 4/2	10MR	10MR
***Sr31 + Lr26 + Yr9 + Lr32* (additive effect for yield increase observed)**			
HW 4049	HD2285*2//WH542/C86-8/Kalyansona F4	R	10MR
HW 4050	HD 2329*2//WH 542 /C 86-8/Kalyansona F4	R	10MR
HW 4052	Kalyansona*2//WH 542 /C 86-8/Kalyansona F4	R	10MR
HW 4053	Lok-1*2//WH 542/C 86-8/Kalyansona F4	R	10MR
HW 4055	PBW 226*2//WH 542 /C 86-8/ Kalyan sona F4	R	10MR
HW 4056	Sonalika*2//WH 542 /C 86-8/Kalyansona F4	R	10MR
HW 4057	WH147*2//WH542/C 86-8/Kalyansona F4	R	10MR
***Sr31 + Lr26 + Yr9 + Lr19 + Sr25* (additive effect for yield increase observed)**			
HW 4059	HD 2009*2//WH 542 /Sunstar*6/C 80-1	10MR	TS
HW 4059-1	HD 2009*2//WH 542 /Sunstar*6/C 80-1	10MR	TS
HW 4060	HD 2329*2//WH 542 /Sunstar*6/C 80-1	10MR	TS
HW 4061	HI 1077*2//WH 542/Sunstar*6/C 80-1	10MR	TS
HW 4061-A	HI 1077*2//WH 542/Sunstar*6/C 80-1	10MR	TS
HW 4062	J 24*2//WH 542/Sunstar*6/C 80-1	10MR	TS
HW 4063	Kalyansona*2//WH 542/Sunstar*6/C80-1	10MR	TS
HW 4064	Lok-1*2//WH 542/Sunstar*6/C 80-1	10MR	TS
HW 4065	NI 5439*2//WH 542/Sunstar*6/C 80-1	10MR	TS
HW 4065-A	NI 5439*2//WH 542/Sunstar*6/C 80-1	10MR	TS
HW 4066	WH 147*2//WH 542/Sunstar*6/C 80-1	10MR	TS
Lr19 + Sr25, Sr36 + Pm6			
HW 4202	HD 2009*3//Cook*6/C80-1	TS	10MR
HW 4203	HD 2285*3//Cook*6/C80-1	TS	TS
HW 4204	HD 2329*3//Cook*6/C80-1	TS	TS
HW 4205	HD 2402*3//Cook*6/C80-1	TS	TS
HW 4206	HD 2687*3//Cook*6/C80-1	TS	10MR
HW 4207	HS 240*3//Cook*6/C80-1	TS	10MR
HW 4208	J 24*3//Cook*6/C80-1	TS	TS
HW 4209	Kalyansona*3//Cook*6/C80-1	TS	TS
HW 4210	Lok-1*3//Cook*6/C80-1	TS	TS
HW 4211	MACS 2496*3//Cook*6/C80-1	TS	10MR

Station, Wellington, for use in wheat improvement programs.

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A high-yielding, multiple rust resistant, bread wheat cultivar HW 5216 (Pusa-Navagiri) released for cultivation in the Southern Hill Zone, India.

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A high-yielding wheat cultivar, HW 5216, conferring multiple rust resistance was released for cultivation in the Southern Hill Zone of India. The Southern Hills in India are known as the main foci for leaf and stem rust and acts as source for fresh epidemics of rusts to the plains of India when wheat crop raised in winter. Although the Southern Hill Zone is a small but strategically very important zone for India area wise, it is particularly important for the control of rust. Therefore, continuous efforts in diversifying the genetic basis of rust resistance by the release of cultivars that contain the rust inoculum is of national importance in order to arrest the dissemination of uredospores to the plains of India to avoid any rust epidemics (Table 2).

Table 2. Adult-plant response of HW 5216 to wheat rusts under artificial inoculation. The predominant yellow rust race prevalent in the Southern Hills is only I race for which this cultivar confers a high degree of field resistance. Figures in parentheses are ACI.

Disease	Year	HW 5216	HW 2044	COW(W)1
Stem rust	2008–09	10S (3.5)	10S(2.6)	10S (1.0)
	2009–10	10S (3.2)	20S (2.9)	10MR (1.0)
	2010–11	40MR (4.0)	20MR (3.6)	10S (5.6)
	2011–12	10S (3.5)	20S (2.9)	10S (2.3)
Leaf rust	2008–09	TMS (0.1)	20MR (1.1)	40S (7.7)
	2009–10	30S (5.9)	80S (10.8)	5S (0.5)
	2010–11	30S (5.9)	80S (10.8)	5S(0.7)
	2011–12	10MS (1.2)	20MS (4.9)	10MR (1.0)
Postulated genes	2009–10	*Sr31 + Lr26 + Yr9*, +		
	2010–11	*Sr31 + Lr26 + Yr9*, +		
	2011–12	*Sr31 + Lr26 + Yr9*, +		

Salient features of the released wheat cultivar HW 5216.

HW 5216 has yielded significantly superior over checks, HW 2044 and CoW(w)1 over four testing periods and recorded highest zonal mean grain yield of 45.6 q/ha (Fig. 1).

HW 5216 recorded an overall yield advantage of +11.82% over best check CoW(W)1, which occurred 10 out of 11 times in first nonsignificant group as compared to best check CoW(W)1, indicating wider adaptability and stability in performance across zones.

A significant yield advantage for HW 5216 ranging from 2.7% to 51.0% over the checks, indicats a yielding ability in the proposed zone and has the highest yield potential of 62.4 q/ha

The mean yield of 48.7 q/ha in Tamilnadu (hills and lower hills) and 36.4 q/ha in Karnataka (areas adjoining) over the period of three years of testing was significantly higher than the both checks.

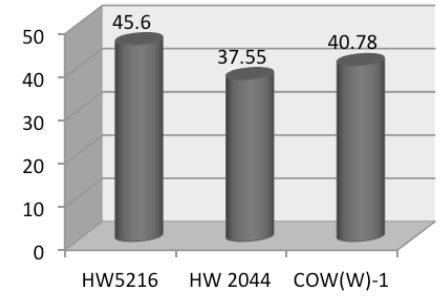

Fig. 1. Comparative mean grain yield of *T. aestivum* cultivar HW 5216 and checks over four years of testing at multiple locations.

When tested for varied irrigation levels, HW 5216 recorded no significant yield loss.

HW 5216 exhibited a high degree of seedling resistance to most stem, leaf, and yellow rust pathotypes under artificial epiphytotic conditions and for all the occurring races in the zone for which it is relased when tested under natural epiphytotic conditions

HW 5216 produced good appearing grains with higher test weight (81.47 kg/hc) and better grain quality (>12% protein and 44.75 sedimentation value).

Marker-assisted selection for the introgression of rust resistance genes (Yr10 and Sr26) in some select Indian wheat cultivars using PCR-based markers.

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Introduction. Wheat, the 'golden grain' or 'king' of the cereal, plays a pivotal role in human nutrition and is a fair source of protein, vitamin B, and minerals around the world. In India, wheat occupies the second slot in production next only to rice, with around 27 x 10^6 ha, yet producing >90 x 10^6 tons and a productivity over 3,000 kg/ha. The consumption of wheat in India has increased over the years, and the demand will continue to grow faster than that of any crop.

There are twelve different wheat diseases in India (Sharma and Mishra 1999) that cause considerable economic loss, but none is more serious than that of the rusts that attack wheat. A combination of cultural and control practices with disease resistance and fungicide applications is the most effective means of control of rust (Singh et al. 1998). Chemical control of rust diseases has been successful, but environmental concerns about chemicals in food grains have limited their use (Rowel 1985). Therefore, the only alternative way of protecting wheat crops is by developing disease-resistant cultivars employing the modern breeding tools such as marker-assisted selection (MAS). Dekkers and Hospital (2002) recently reviewed some of the potential limitations of MAS strategies and concluded that the use of MAS will be determined by the economic benefit relative to conventional selection. This study detected the transfer of two rust resistance genes. *Sr26* and *Yr10*, in different genetic background through molecular markers (AFLP-Sr26#43SSR–XPSP3000).

Materials and methods. All lines were produced employing conventional backcross selection at IARI, Regional Station, Wellington, which is a hot spot for wheat rust, and by taking three crop cycles of wheat in a single year. Simultaneous selection for the targeted genes using MAS is at the TamilNadu Agricultural University, Coimbatore. The entire lot was shuttled between the centers for selection of the best-yielding phenotype with specific genes with wider adaptability. Segregating material was regularly screened at the IARI Regional Station in Wellington for host-pathogen interaction (phenotyping) and confirmation of the resistance offered by each targeted gene(s).

The yellow rust resistance locus *Yr10* is located on short arm of chromosome 1B in Moro and originated from the Turkish line Pl178383. Polymorphism in 12 segregating lines of two different cultivars, 'WH147/*Yr10*' in the BC_1 generation and 'LOK-1/*Yr10*' in the BC_3 generation, confirmed the transfer of *Yr10* resistance gene using a molecular marker (SSR–XPSP 3000). The microsatelite marker *Xpsp3000* is inherited in a codominant manner and linked with the yellow rust resistant gene *Yr10*, with a distance of 1.2 cm, and is useful in MAS.

The *Thinopyrum elongatum*-derived *Sr26* gene is located on the long arm of chromosome 6A. This gene was confirmed at the molecular level by using AFLP marker Sr26#43 in the segregating lines from four cultivars along with five different sources. The plant materials used for molecular confirmation of rust resistance gene *Sr26* included BC_1, BC_2, and F_2 plants from various cross combinations.

Results and discussion. Molecular markers act as DNA signposts for locating gene(s) for a trait of interest on a plant chromosome. In 50 lines of different cross combination screened for the presence of stem rust resistance gene *Sr26*, 46 lines revealed the presence with the primers specific to *Sr26* at 200 bp and 207 bp through PCR analysis.

The dominant gene *Yr10* showed polymorphism for the microsatelite locus *Xpsp3000* in 12 segregating lines of 'WH147/*Yr10*' (BC_1) and 'LOK-1/*Yr10*' (BC_3). Because *Yr10* was located on the short arm of chromosome 1B, only nine SSR markers distributed on the short arm of chromosome 1B were chosen for screening for polymorphism between the resistant and susceptible cultivars. The unique genetic associations of *Yr10*-specific alleles of *Xpsp3000* will be useful in MAS and gene pyramiding. *Yr10* showed a close genetic association with alternate alleles at *Xpsp3000* and/or *Gli-B1* and could be used in MAS for pyramiding *Yr10* with other stripe rust resistance genes (Bariana et al. 2007).

Conclusion. Public breeding programs that have the expertise to utilize successfully MAS technologies have both a challenge and a fantastic opportunity. With the availability of detailed information regarding the location and function of gene(s) encoding for useful traits, scientists are now well equipped for efficiently creating cultivars with the exact combinations of desirable traits. However, genetic transformation will remain a significantly important tool for understanding gene functions and testing the utility of new sequences. In near future, cultivars could be tailor-made to meet both local

consumer preferences and the demands of particular environment or niche. The new tools of biotechnology not only have the potential for increasing the effectiveness and efficiency of wheat breeding programs and but also provide insights into the genetic control of key traits to be used for genetic manipulation. The coming years will undoubtedly witness an increasing application of biotechnology for the genetic improvement of wheat trait-specific products and with better genome recovery.

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ITEMS FROM ITALY

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CONSIGLIO PER LA RICERCA E LA SPERIMENTAZIONE IN AGRICOLTURA (CRA-QCE)
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Faenza, Italy.

CENTRO RICERCHE PRODUZIONI VEGETALI (CRPV)
Imola, Italy.

Response of 31 durum wheat cultivars to cereal soilborne mosaic virus in 2012.

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Thirty-one durum wheat cultivars were grown during the 2011–12 season in a field with natural inoculum source of cereal soil-borne mosaic virus (CSBMV) at Cadriano, near Bologna, and evaluated for resistance on the basis of symptom severity, DAS-ELISA value, and agronomic performance. The cultivars, planted on 24 October 2011, were grown in 10-m^2, solid-seeded plots distributed in the field according to a randomized block design with three replicates. Symptom severity was evaluated on four dates (9, 16, 20, and 27 March) using a 0–4 scale. DAS-ELISA was performed on extracts from a bulk of the basal half of the second and third youngest leaves of 10 randomly chosen plants/plot collected on one date only (19 March, 2012). The trial included 10 cultivars THAT had not been tested before.