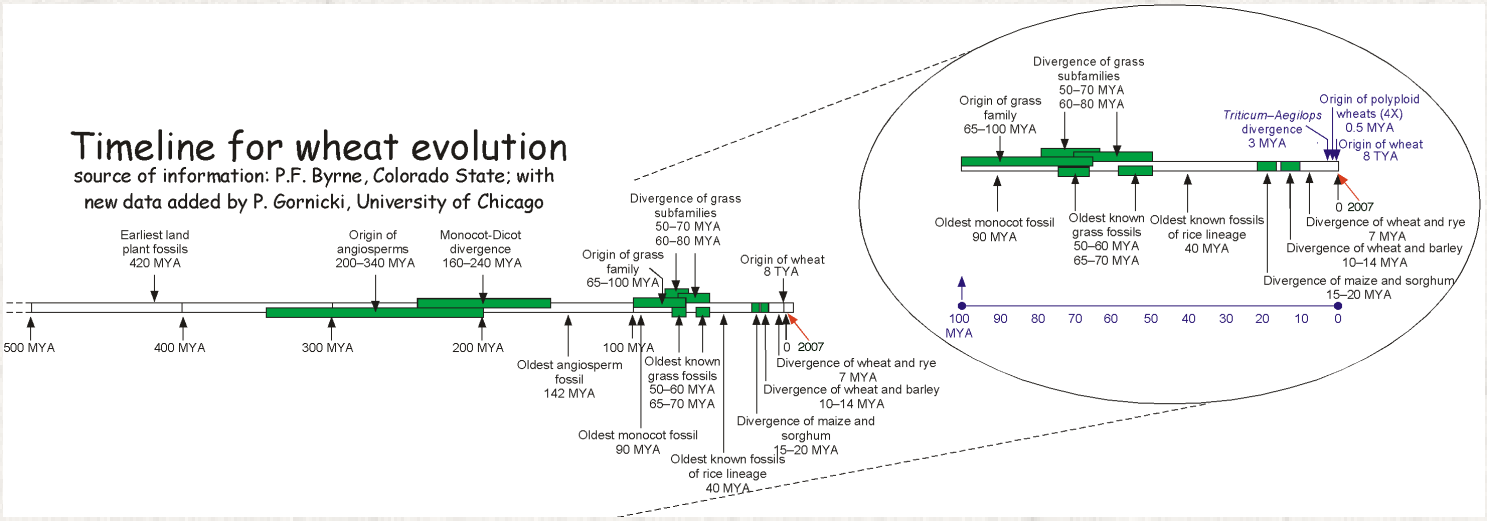
ANNUAL WHEAT NEWSLETTER

Volume 59



Contribution no. 14-049-D from the Kansas Agricultural Experiment Station,
Kansas State University, Manhattan.

ANNUAL WHEAT NEWSLETTER

Volume 59

Edited by W.J. Raupp, Department of Plant Pathology, Kansas State University, Manhattan, KS 66506-5502 USA. Facilities during manuscript editing were provided by the Plant Pathology Department and the Wheat Genetic and Genomic Resources Center, Kansas State University.

9 August, 2013.

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IN DEDICATION TO DR. ROBERT H. BUSCH

Robert Busch, retired USDA Wheat Geneticist and Adjuct Professor Emeritus in the Department of Agronomy and Plant Genetics, age 74, passed away peacefully surrounded by family 29 June, 2012. Survived by loving wife Mavis, with whom he shared 53 wonderful years of marriage; sister Elizabeth Gunion; son, Todd; daughter-in-law Dana; and grandchildren, Patrick, Meaghan, Elliott and Ian. Robert is preceded in death by father, Henry; mother, Lena; and daughter, Shari. Robert was an outstanding scientist and father who will be greatly missed by his friends and colleagues.

Dr. Busch advised many graduate students in agronomy and plant genetics and developed several successful wheat cultivars that have brought millions of dollars to Minnesota farmers and beyond. He was a very kind man with a great sense of humor.

A memorial service was held at 11:00AM, 9 July, 2012, at the Centennial United Methodist Church, 1524 West County Rd. C2, Roseville, MN. Memorials preferred to either the Minnesota chapter of the ALS Society or the Ramsey County Humane Society.

IN DEDICATION TO DR. JAMES P. WILSON

Dr. James T. Wilson passed away quietly at 8:00AM, 31 July, 2012. Jim had suffered a major stroke and had been in intensive care at a local hospital in Wichita.

Jim was truly a pioneer in the Plains wheat industry. We have all greatly valued Jim's insights, dedication, and friendship. He will be deeply missed. With the passing of Dr. Wilson, we have lost a true pioneer in the wheat industry. Jim leaves a legacy that few can match in breeding and wheat improvement.

Jim Wilson received his B.S. and M.S. degrees from Oklahoma State University, followed by a Ph.D. from Texas A&M University. He began his career as wheat project leader at the Fort Hays Branch Experiment Station, Fort Hays, Kansas in 1957. In Kansas, he initiated among the first studies on cross breeding and cytoplasmic sterility in wheat. Jim Wilson joined DeKalb AgResearch in 1961 with the opportunity to restart private wheat breeding in the U.S. with emphasis on breeding and marketing of hybrid wheat. Jim published several papers on hybrid breeding and restoration systems while with DeKalb. His project released the first commercially available hybrid wheat to growers in 1978. He was also responsible for development and release of the first semidwarf wheat cultivars for production in Kansas.

When DeKalb closed their hybrid wheat project in 1981, Jim founded Trio Research Inc. as a family-owned corporation in Wichita, Kansas. This corporation was initially formed to be a service company for a new biotech company, Agrigenetics Research Associates (ARA). Jim built his own facilities and started breeding on the family farm. The four-year relationship with ARA was followed by a two-year service contract with Lubrizol Genetics. In 1989, as service contracts ended, Trio began its own product development as a private seed company, developing both varieties and hybrids.

Jim's first Trio wheat cultivar, T67, was released in 1993, followed by T81 in 1995. His business was unique in that the wheat cultivars were grown and protected under private contracts. His cultivars, notably T81 and his most recent release T158, have been grown on significant acreage in the Central Plains and contributed as parents to many more. Federal seed authorities recently approved Trio Research for the marketing of the first synthetic wheat cultivars in the United States, developed using a mixture of male and female plants. Trio was acquired by Limagrain Cereal Seeds (LCS) in 2010. Jim then served as a consultant for LCS, continuing his research and breeding on hybrid wheat, until his death. Jim was a strong advocate for wheat breeding, germ plasm exchange, and wheat growers. He was actively involved in regional wheat improvement programs. We have all greatly valued Jim's insights, integrity, dedication, and friendship over the years. He will be deeply missed.

Jim was dedicated to his family and is survived by his wife, Joy, daughter Diane, and sons Dean, Daniel, and David.

RETIREMENT
DR. VASILE P. MOLDOVAN

Wheat breeder Dr. Vasile P. Moldovan has reached retirement age of 65 in March, 2012, after more than 40 years devoted to wheat genetics and breeding research.

Dr. Moldovan was born on 10 March, 1947, in Micestii de Campie, a village located in the heart of the rich agricultural area of the county Bistrita-Nasaud from Romania. After completing his elementary school in the native locality, he continued high school education (1961–65) in the town of Bistrita, where his parents moved later. He began his study with the Faculty of Agronomy of the University of Agricultural Sciences and Veterinary Medicine (USAMV) in Cluj-Napoca in 1966 and graduated in 1971. Later, in 2003 he completed a Ph.D. thesis at the same University and earned title Doctor in Agronomy with *magna cum laude*. In order to widen his professional knowledge, Dr. Moldovan visited some research stations in different countries. One of the most important of these visits was made in September– October 1985 at the University of Nebraska.

The first place of work for Dr. Moldovan was at a Seed Production Farm in Taga, where he spent several months (1 November, 1971 to 1 April, 1972) immediately after leaving the university. From here he moved to the Agricultural Research Station Turda, joining to the wheat breeding team in 1972. He gradually worked his hierarchical scientific way up to head of the Small Grains Breeding Department.

Dr. Moldovan and his colleagues established the main objectives of the wheat breeding program as having the main goal developing wheat cultivars able to satisfy farmers' requirements with reference to productivity, quality, and yield stability. To achieve these complex objectives, it was necessary to carry out a wide range of theoretical and methodological research emphasizing on yield and quality as well as on biotic and abiotic stress tolerance. In close cooperation with his colleagues, Dr. Moldovan was involved in development and release of 10 winter wheat cultivars, two winter triticale cultivars, a spring triticale cultivar, and two spring oats cultivars. He authored or co-authored 125 scientific and technical publications in Romanian and other languages (especially English), and made more than 100 lectures to professional and lay audiences.

During 1997–2003, Dr. Moldovan served as Romania's member of Wheat Working Group members of European Cooperative Programme for Crop Genetic Resources Networks (IPGRI–ECP/GR Wheat Working Group Members). He also served as a member of Ph.D. Thesis Committees for some students from USAMV Cluj-Napoca. On many occasions his knowledge and experience have bought a sense of reality to discussions with colleagues and other persons, trying to resolve difficult cereal breeding problems.

We wish to him a long and happy retirement, together with his wife Maria, a former research biologist on wheat disease resistance, and their daughter Luminita, who lives in Bucharest.

WHEAT WORKER'S CODE OF ETHICS

This seed is being distributed in accordance with the 'Wheat Workers' Code of Ethics for Distribution of Germ Plasm', developed and adopted by the National Wheat Improvement Committee on 5 November, 1994. Acceptance of this seed constitutes agreement.

1. The originating breeder, institution, or company has certain rights to the material. These rights are not waived with the distribution of seeds or plant material but remain with the originator.

2. The recipient of unreleased seeds or plant material shall make no secondary distributions of the germ plasm without the permission of the owner/breeder.

3. The owner/breeder in distributing seeds or other propagating material grants permission for its use in tests under the recipient's control or as a parent for making crosses from which selections will be made. Uses for which written approval of the owner/breeder is required include:

   (a) Testing in regional or international nurseries;
   (b) Increase and release as a cultivar;
   (c) Reselection from within the stock;
   (d) Use as a parent of a commercial F_1 hybrid, synthetic, or multiline cultivar;
   (e) Use as a recurrent parent in backcrossing;
   (f) Mutation breeding;
   (g) Selection of somaclonal variants; or
   (h) Use as a recipient parent for asexual gene transfer, including gene transfer using molecular genetic techniques.

4. Plant materials of this nature entered in crop cultivar trials shall not be used for seed increase. Reasonable precautions to ensure retention or recovery of plant materials at harvest shall be taken.

ITEMS FROM BRAZIL

BRAZILIAN AGRICULTURAL RESEARCH CORPORATION — EMBRAPA
Rodovia BR 285, km 294, Caixa Postal 451, Passo Fundo, RS, Brazil.

Wheat in Brazil – 2012 crop year.

Eduardo Caierão.

In the 2012 crop year, Brazilian wheat production was about 4 x 10^6 tons (Conab 2013), which is enough to supply 40% of the domestic demand (Table 1). The deficit in production makes Brazil the largest wheat importer. The southern region, comprised of the states of Rio Grande do Sul, Santa Catarina, and Paraná, accounts for 94.6% of the national production. Nonetheless, due to the characteristics of the cultivation system, average grain yield in this region is not the highest in the country.

Table 1. Area of cultivation, total production, and grain yield of wheat in Brazil in 2012 (Source: CONAB 2013).

Region	Area (ha x 1,000)	Production (t x 1,000)	Grain yield (kg/ha)
North	—	—	—
Northeast	—	—	—
Central–West	24.8	68.2	2,750.0
Southeast	53.5	162.4	3,036.0
South	1,817.1	4,069.8	2,240.0
Brazil	**1,895.4**	**4,300.4**	**2,269.0**

In 2012, the wheat area cultivated was lower than that in 2011 (1,895.4 against 2,166.2). The total production and average grain yield/ha achieved in 2012 were 25% and 15% smaller than 2011, respectively. High rainfall and frost during grain filling were the main factors that justify these data.

Reference.

CONAB. 2012. Companhia Nacional de Abastecimento. Central de Informações Agropecuárias/Grãos/Trigo. Disponível em: http://www.conab.gov.br/conabweb/index.php?PAG=131

Wheat seed market in Brazil – 2009 to 2012.

Eduardo Caierão.

Few companies hold the wheat seed market in Brazil, characterized primarily by private capital. The percent participation of each company in the seed market between 2009 and 2012 in Brazil (Table 2), versus that in the main cereal producing states of Rio Grande do Sul (Table 3, p. 6) and Paraná (Table 5, p. 6) are given. In terms of Brazil, the percent of the market covered by private companies was 71.20%, 74.78%, 75.70%, and 85.09%, respectively, from 2009 to 2012 (Table 2), indicating increased participation of private

Table 2. The market for wheat in Brazil between 2009 and 2012, Passo Fundo, Brazil, 2013 (Source: Kleffmann 2013, bold values indicate the highest percent in each year).

Company	Type	2009	2010	2011	2012
Biotrigo	Private	0.00%	0.00%	0.12%	6.75%
Coodetec	Private	**25.79%**	22.08%	19.21%	18.38%
Embrapa	Public	25.27%	23.62%	22.11%	13.42%
CCGL TEC	Private	21.17%	12.36%	9.79%	6.72%
OR Sementes	Private	24.23%	**40.34%**	**46.58%**	**53.24%**
Outros	Public	3.53%	1.60%	2.18%	1.49%
Total	Private	71.20%	74.88%	75.70%	85.09%
	Public	28.80%	25.22%	24.30%	14.91%

companies in the seed market. Coodetec (2009) and OR Sementes (2010, 2011, and 2012) were the market leaders. Analyzing the information by state, there are some differences. In Rio Grande do Sul, for example, the participation of private companies in the seed market is even higher, over 90% in 2011 and 2012. However, the leadership still prevailed OR Sementes. In Paraná, public enterprises occupy an area more significant than in Rio Grande do Sul, reaching 37% in 2011, primarily due to the contribution of Embrapa. In this state, Coodetec led the market in 2009, 2010, and 2012. Embrapa had greater participation in 2011. The use of certified seed, saved seed, or breeder seed for each region (Brazil, and the states of Rio Grande do Sul and Paraná) are given (Tables 5, 6, and 7). The highest use of certified seed can be found in Paraná, ranging from 79.45% to 89.67%. The usage profile in Rio Grande do Sul is much lower, reaching a level close to 50% in 2012.

Table 3. The market for wheat in the state of Rio Grande do Sul between 2009 and 2012. Passo Fundo, Brazil, 2013 (Source: Kleffmann 2013. Bold values indicate the highest percent in each year).

Company	Type	2009	2010	2011	2012
Biotrigo	Private	0.00%	0.00%	0.00%	9.18%
Coodetec	Private	1.28%	1.95%	3.02%	0.53%
Embrapa	Public	14.08%	18.26%	8.40%	7.46%
CCGL TEC	Private	**54.39%**	30.66%	21.35%	12.11%
OR Sementes	Private	29.70%	**49.13%**	**67.13%**	**69.74%**
Outros	Public	0.55%	0.00%	0.10%	0.97%
Total	Private	85.37%	81.74%	91.50%	91.56%
	Public	14.63%	18.26%	8.50%	8.44%

Table 4. The market for wheat in the state of Paraná between 2009 and 2012. Passo Fundo, Brazil, 2013 (Source: Kleffmann 2013. Bold values indicate the highest percent in each year).

Company	Type	2009	2010	2011	2012
Biotrigo	Private	0.00%	0.00%	0.02%	3.27%
Coodetec	Private	**45.52%**	**37.29%**	34.30%	**40.94%**
Embrapa	Public	31.76%	26.32%	**34.41%**	20.44%
CCGL TEC	Private	0.21%	0.36%	0.64%	0.63%
OR Sementes	Private	18.54%	34.38%	27.65%	33.27%
Outros	Public	3.98%	1.65%	2.98%	1.45%
Total	Private	64.27%	72.03%	62.61%	78.11%
	Public	35.73%	27.97%	37.39%	21.89%

Table 5. Percent of certified and saved seed in the wheat market in Brazil. Passo Fundo, Brazil, 2013 (Source: Kleffmann 2013. Bold values indicate the highest percent in each year).

Type of seed	2009	2010	2011	2012
Breeder Seed	1.41%	0.06%	2.14%	1.86%
Certified	**79.18%**	**78.82%**	**76.42%**	**64.47%**
Saved	19.41%	21.11%	21.44%	33.67%

Table 6. Percent of certified and saved seed in the wheat market in the state of Rio Grande do Sul, Brazil. Passo Fundo, Brazil, 2013 (Source: Kleffmann 2013. Bold values indicate the highest percent in each year).

Type of seed	2009	2010	2011	2012
Breeder Seed	0.92%	0.00%	4.49%	2.61%
Certified	**69.25%**	**63.49%**	**65.14%**	**51.59%**
Saved	19.41%	21.11%	21.44%	33.67%

Table 7. Percent of certified and saved seed in the wheat market in the state of Paraná, Brazil. Passo Fundo, Brazil, 2013 (Source: Kleffmann 2013. Bold values indicate the highest percent in each year).

Type of seed	2009	2010	2011	2012
Breeder Seed	1.32%	0.00%	0.22%	0.81%
Certified	**87.36%**	**89.67%**	**86.33%**	**79.45%**
Saved	11.32%	10.33%	13.45%	19.74%

Reference.

Kleffmann Group. 2013. Wheat seed's market in Brazil - 2009 to 2012. Available at: http://www.kleffmann.com.

Use of Embrapa germ plasm in Brazilian wheat breeding programs.

Eduardo Caierão, Pedro L. Scheeren, Márcio Só e Silva, Ricardo Lima de Castro, and Adeliano Cargnin.

Embrapa has about 25% of wheat seed market in Brazil. The wheat breeding program of Embrapa started in 1974 and, since that time, more than 100 new cultivars have been released. Since 2005, the number of recommended wheat cultivars in Brazil is over 100; only in 2009 was the number was lower (Table 8). The number of cultivars that use Embrapa germ plasm (NCWEG) in this period range according to new releases and retry of materials. The relationship between indicated cultivars and NCWEG was 65.4%, 65.4%, 64.9%, 62.3%, 64.9%, 65.0%, 71.3%, and 71.8%. In 2012, more than 71% of the wheat cultivars recommended to producers had at least one Embrapa's genotype in your genealogy.

Table 8. Wheat cultivars indicated by Brazilian breeding institutions for cultivation, from 2005 to 2012; number of cultivars indicated with Embrapa germ plasm (NCWEG) in the genealogy and percent of use (% USE). Passo Fundo, RS, Brazil, 2013.

	2005	2006	2007	2008	2009	2010	2011	2012	Mean
Indicated cultivars	104	104	111	106	94	100	101	110	104
NCWEG	68	68	72	66	61	65	72	79	69
% UGE	65.4	65.4	64.9	62.3	64.9	65.0	71.3	71.8	66.4

Pyramiding genes for tolerance to preharvest wheat sprouting by backcrossing.

Adeliano Cargnin, Flávio Martins Santana, Eduardo Caierão, Ricardo Lima de Castro, Pedro Luiz Scheeren, Marcos Fabris, and Marcos Kovaleski.

A study of gene pyramiding for tolerance to preharvest sprouting into new wheat lines began in July 2011, at Embrapa Trigo, through the backcross method. The study obtained and evaluated the first generation backcross (BC_1) originated from a cross between contrasting wheat germ plasm for tolerance to preharvest sprouting. The study was conducted at Embrapa Trigo in Passo Fundo, RS. The recurrent parental was the cultivar BRS Pardela. As parental donors, we used the cultivars Lagoa Vermelha and Syrimex. Crosses were made between the recurrent and the donors, and then the F_1 was backcrossed with the recurrent parent to produce the BC_1. Seeds of the BC_1 were evaluated for tolerance to preharvest sprouting by a germination test. For this test, the seeds were sown on RC1 germitest paper moistened and placed in a Mangelsdorf germination chamber at 20°C ± 2°C. On the third and fourth day after sowing (72 and 96 hours), counts were performed and germinated seeds removed. The remaining seeds were sown in plastic pots under greenhouse conditions in order to give rise the next generation of backcrossing. The methodology applied in this study was effective for discriminating BC_1 populations with regard to the level of tolerance for preharvest sprouting. By this method, 20% to 40% of the BC_1 seeds were eliminated (Fig. 1). Removing a greater number of seeds enhances the probability of making the next backcross with plants with good tolerance to preharvest sprouting. Thus, the cross 'Pardela BRS/Syrimex' was more efficient in selection; more than 40% of the seeds were eliminated. On the other hand, the cross 'BRS Pardela/Lagoa Vermelha' proved to have promising cross-tolerance to preharvest sprouting, because approximately 80% of the seeds did not germinate, even after 96 hours of germination treatment.

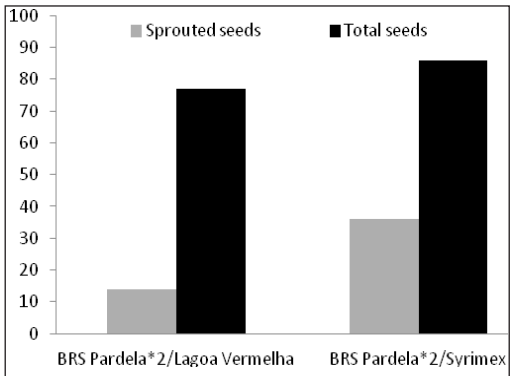

Fig. 1. Sprouted seeds and total seeds in two backcrossing population (BC_1) of wheat (Passo Fundo, Brazil, 2012).

Resistance gene pyramiding for Fusarium head blight (FHB) in wheat by backcrossing.

Adeliano Cargnin, Flávio Martins Santana, Eduardo Caierão, Ricardo Lima de Castro, Pedro Luiz Scheeren, Marcos Fabris, and Marcos Kovaleski.

A project pyramiding resistance genes for FHB on new wheat lines was initiated in July 2011 at Embrapa Trigo, using a backcrossing method. The objective was evaluate plants from the first generation of backcrossing (BC_1) that came from crossing germ plasm resistant vs. susceptible to FHB. This research was done at Embrapa Trigo, Passo Fundo, RS, Brazil. The recurrent parent was the commercial cultivar BRS Guabiju. The donor parental was the cultivar Sumai 3. Crosses between the recurrent and donor parents were made during the winter, 2011. The F_1 offspring was backcrossed with the recurrent parent in the summer of 2011, producing the first backcross generation (BC_1). Plants from the BC_1 were evaluated for reaction to FHB. Seeds of BC_1 were sown in plastic plots in the green-house. At the flowering time (Feekes and Large scale 10.5.1), a spore suspension of the *Gibberella zeae* isolate 6047 was sprayed over the spikes at a concentration of 100.000 spores/mL applied at 25°C ± 2°C and 80% humidity. Ten days after the inoculation. the plants were evaluated for incidence and severity of FHB. The methodology applied for this research was efficient fpr screening the BC_1 population for FHB resistance level. The genotypes were separated into six classes of FHB severity, from 0 to 100% (Fig. 2). We evaluated 165 spikes, of which 92 showed no FHB symptoms. Hence, the frequency of spikes resistant to FHB (0% of severity) in the BC_1 generation was 56%. The cross 'BRS Guabiju/Sumai 3' is a good source of genetic variability for resistance to FHB.

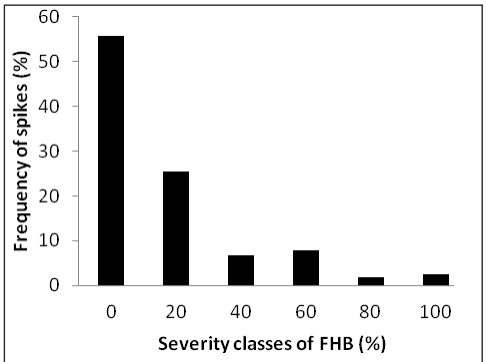

Fig. 2. Frequency of severity of Fusarium head blight in wheat spikes in a backcross (BC_1) population inoculated under greenhouse conditions (Passo Fundo, Brazil, 2012).

Response of wheat cultivars to inoculant use in southern Brazil.

Ricardo Lima de Castro, José Pereira da Silva Júnior, Eduardo Caierão, Adeliano Cargnin, and Luciano Consoli.

Inoculation of wheat seeds with *Azospirillum brasiliense* bacteria has resulted in an increase in wheat grain yield in southern Brazil. However, the effect of inoculation with this facultative endophytic bacterium, capable of fixing nitrogen from the atmosphere and providing part of the associated N required by the plant, has been dependent on the wheat cultivar. To identify the cultivars most responsive to the association with *A. brasiliense*, 32 wheat cultivars of southern Brazil were evaluated with and without seed inoculation. The experiment was carried out at Embrapa Wheat (lat 28°15'S, long 52°24'W, alt 684 masl), in Passo Fundo, state of the Rio Grande do Sul, Brazil, in a randomized block design with three replications. Each plot consisted of three 3-m rows with 0.17 m spacing between rows (1.8 m^2). The variables studied were aerial part dry mass, grain yield, harvest index, plant height, and number of spikes per area. Data were subjected to analysis of variance and the treatments with and without inoculation were compared by the t test ($P < 0.05$) for each wheat cultivar. Inoculation of wheat seeds with *A. brasiliense* bacteria resulted in an increase of grain yield in wheat cultivars Turquesa (+1,535 kg/ha or +43%), TBIO Itaipu (+971 kg/ha or +35%), CD 123 (+800 kg/ha or +40%), TBIO Alvorada (+775 kg/ha or +24%), CD 1550 (+675 kg/ha or +22%), BRS 331 (+607 kg/ha or +22%), BRS 374 (+570 kg/ha or +16%), Fundacep Raízes (+544 kg/ha or +20%), TBIO Seleto (+458 kg/ha or +14%), TEC Frontale (+367 kg/ha or +10%), Marfim (+361 kg/ha or +12%), Ametista (+356 kg/ha or +12%), CD 114 (+296 kg/ha or +10%), JF 90 (+272 kg/ha or +9%), TBIO Mestre (+267 kg/ha or +7%), TBIO Pioneiro (+172 kg/ha or +4%), and CD 122 (+84 kg/ha or +3%). Only for cultivars CD 123 and TBIO Alvorada was the difference in grain yield mean with and without inoculation statistically significant by the t test ($P < 0.05$). The most responsive cultivars to inoculation with *A. brasiliense* will be used in wheat breeding programs in southern Brazil.

ITEMS FROM GERMANY

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Whole genome association mapping of resistance to Fusarium head blight and Septoria tritici blotch in European winter wheat.

A total of 358 recent European winter wheat cultivars plus 14 spring wheat cutivars were genotyped (in collaboration with industrial partners) with 732 microsatellite markers resulting in 782 loci of which 620 were placed on the ITMI map. The resulting average marker distance of 7.2 cM allowed genome-wide, association mapping employing a linear mixed model.

A minor allele frequency threshold of 3% (equalling 11 cultivars) was set, and alleles with a lower frequency were excluded from the analysis. After filtering, 3,176 alleles remained and were employed for the association mapping approach. Although no clear population structure was discovered, a kinship matrix was used for stratification.

All cultivars were evaluated for resistance to Fusarium head blight (FHB) caused by *Fusarium graminearum* and *F. culmorum* in four separate environments (2009 and 2010 in Ahlum; 2009 in Cecilienkoog; 2010 in Halle-Bodenwerder; by B. Rodemann, Julius Kühn Institute, Braunschweig (JKI), Germany). The FHB scores based on 'FHB incidence (type-I resistance) / FHB severity (type-II resistance)' indicated a wide phenotypic variation of the cultivars with BLUEs (best linear unbiased estimation) ranging from 0.07 (most resistant) to 33.67 (most susceptible). A total of 794 significant (-log10(p)-value > 3.0) associations between SSR loci and environment-specific FHB scores or BLUE values were detected, which included 323 SSR alleles. For FHB incidence and FHB severity, a total of 861 and 877 individual marker-trait associations (MTA), respectively, were detected. Of these marker alleles, 210, 229, and 194 were significant for BLUE values for FHB incidence, FHB severity, and FHB score, respectively. In most cases, the same marker alleles detected significant associations for all three parameters, i.e., FHB incidence, severity, and FHB score.

Consistent associations detected in three or more environments were found on all chromosomes except chromosome 6B and with the highest number of MTA on chromosome 5B. The dependence of the number of favorable and unfavorable alleles within a cultivar to the respective FHB scores indicated an additive effect of favorable and unfavorable alleles, i.e., genotypes with more favorable or less unfavorable alleles tended to show greater resistance to FHB. Assessment of a marker specific for the dwarfing gene *Rht-D1* resulted in strong effects. The results provide a prerequisite for designing genome wide breeding strategies for FHB resistance.

The same cultivars were evaluated for resistance to *Septoria tritici* blotch caused by the fungal pathogen *Mycosphaerella graminicola* (by B. Rodemann, Julius Kühn Institute, Braunschweig, Germany). The MTA were based on field data in two consecutive years (2009 and 2010 in Cecilienkoog) and genotypic data of 732 microsatellite markers. BLUEs for resistance were calculated across the trials and ranged from 0.67 (most resistant) to 19.63 (most susceptible) with an average value of 4.93. A total of 115 MTAs relating to 68 molecular markers were discovered for the two trials, and BLUEs by using a mixed linear model corrected by a kinship matrix. Two candidate genes, Ppd-D1 for photoperiodism and the dwarfing gene *Rht-D1*, also were significantly associated with resistance to *Septoria tritici* blotch. Several MTA co-located with known resistance genes, i.e., *Stb1*, *Stb3*, *Stb4*, *Stb6*, and *Stb8*, whereas multiple additional MTA were discovered on several chromosomes, such as chromosomes 2A, 2D, 3A, 5B, 7A, and 7D. The results provide proof of concept for the method of genome wide association analysis and indicate the presence of further Stb resistance genes in the European winter wheat pool.

Seed longevity in near-isogenic lines.

The artificial ageing (AA) method was used to compare seed longevity of two Siberian spring cultivars Saratovskaya 29 (S29) and Novosibirskaya 67 (N67), near-isogenic lines (NILs) developed from S29 and N67, and several control cultivars. The AA test was performed at 42°C, 44°C, 46°C, and 48°C in three replicates (100 seeds/replicate). A noticeable effect of AA on germination of S29 and N67 was achieved only at 48°C. We compared germination of seeds of S29 and N67 and their NILs after AA test performed at 48°C. From the two S29-based NILs having purple pericarp color, both showed more than 20% higher germination rate in comparison with their red-grained recurrent parent S29.

From the two N67-based NILs having purple pericarp color, one showed more than 10% higher germination rate compared with their white-grained recurrent parent N67. The second line did not differ from N67. Microsatellite-based genotyping showed that, unlike other lines, this line is still carrying extended fragment of the donor parent on chromosome 2A, covering both locus for pericarp color and locus corresponding to the QTL for seed longevity described on chromosome 2A earlier. Thus, the donor's allele in the latter locus could negate the positive effect of purple flavonoid substances in the pericarp on seed longevity.

Chromosomal location of genes for tolerance of prolonged drought in bread wheat.

Drought is one of the most damaging environmental stresses affecting grain yield of bread wheat throughout almost the full geographic range of the crop's production. Bread wheat germ plasm includes a measure of variation in drought tolerance, reflected by its cultivation under a wide range of climatic and edaphic conditions. Among its wild relatives are species naturally adapted to droughted environments, and these offer some potential for extending the genetic variability of wheat via wide hybridization and introgression. Variation in tolerance of prolonged drought was identified among a set of single-chromosome, bread wheat substitution lines involving the replacement of each Chinese Spring (CS) chromosome in turn with its homologue from a synthetic hexaploid (*T. turgidum* subsp. *dicoccoides*/*Ae. tauschii*) wheat (SH). Water stress was applied under controlled conditions by limiting the supply of water to 30% from 100% aqueous soil. Our aim was to identify which chromosomes were most critical to the determination of the drought tolerance displayed by CS and SH. The reaction to the resulting long-term drought stress was quantified by three indices, based on grain yield components. The individual chromosome substitution lines varied with respect to their tolerance, particularly those involving either the A or the D genome (in particular, chromosomes 1A, 5A, 1D, 3D, 5D, and 6D). The least tolerant line was 'CS/SH 4B', followed by 'CS/SH 3A', and 'CS/SH 7D'. The best combination of productivity under well-watered conditions and tolerance to drought was exhibited by 'CS/SH 1D'.

Leaf dehydroascorbate reductase and catalase activity is associated with soil drought tolerance in bread wheat. The study was to identify chromosomes that are responsible for the antioxidant enzymes dehydroascorbate reductase (DHAR, EC 1.8.5.1) and catalase (CAT, EC 1.11.1.6) activities in the leaves of wheat plants and stability of yield components under water deficit. The single-chromosome CS substitution lines have separate chromosomes from the donor synthetic hexaploid, an artificial hexaploid combining the genomes of *T. turgidum* subsp. *dicoccoides* (AABB) and *Ae. tauschii* (DD).

The lines carrying a synthetic hexaploid homologous pair of chromosomes 1B, 1D, 2D, 3D, or 4D all expressed a low constitutive level of DHAR and the lines with chromosomes 3B, 1D, 2D, and 3D a low constitutive level of CAT. All were able to increase this level in response to stress caused by water deficit. When challenged by drought stress, these lines tended to be the most effective in retaining the water status of the leaves and preventing the grain yield components from being compromised. As a result of the investigation, we may conclude that DHAR and CAT activities in leaves of wheat at the tillering stage are associated with resistance to drought of the yield components. The discovered genetic variability for enzymes activity in leaves of wheat might be a useful selection criterion for drought tolerance.

Resistance to tan spot and Septoria leaf blotch in hexaploid wheat.

Tan spot and Septoria leaf blotch are very important foliar diseases in wheat and cause significant yield losses. QTL for resistance against both diseases were mapped in different populations. Segregation for resistance among a set of 49 recombinant inbred lines from the cross 'W7984/Opata 85' was used to identify the basis for resistance to tan spot against

two fungal isolates. A QTL stable across several environments was located on the short arm of chromosome 6A, linked to the microsatellite locus *Xksuh4c* in seedlings.

To identify the location of the resistance to Septoria leaf blotch, a population of 87 single-chromosome recombinant doubled-haploid lines bred from the cross between the landrace Chinese Spring and a Chinese Spring-based line carrying chromosome 7D from *T. aestivum* subsp. *spelta* was used. Two regions of the chromosome 7D were associated with isolate-specific QTL expressed one at the seedling and another at the adult-plant stage. The seedling resistance locus *QStb.ipk-7D1* was found in the centromeric region of chromosome 7D, which corresponds to the location of the major resistance genes *Stb4* originating from bread wheat cultivar Tadinia, and *Stb5* originating from *Ae. tauschii*. The adult-plant resistance locus *QStb.ipk-7D2* was found on the short arm of chromosome 7D in a similar position to the locus *Lr34*/*Yr18* known to be effective against multiple pathogens. Composite interval mapping confirmed *QStb.ipk-7D1* and *QStb.ipk-7D2* to be two distinct loci.

Physiological response of wheat-rye hybrids with dwarf 'grass-clump' phenotype to GA3 application.

A gibberellin treatment has been used to try to induce elongation of stems and seed production in the grass-clump wheat hybrids. However, the effect of gibberellic acid on shoot elongation of the wheat–rye hybrid dwarfs from crosses between Chinese Spring wheat (CS) and inbred rye lines V1 and V10, which carry the allele *Hdw-R1b* was never studied before. For the comparative analysis wheat–rye hybrids (CS/V1, CS/V10, and CS/L6), wheat hybrids (Gabo/Sun, Sun/Gabo, CS/Gabo, and CS/Sun), and their parental forms were tested in this experiment.

The effect of gibberellic acid on shoot elongation of the dwarf wheat–rye hybrids, wheat hybrids, and parental forms was different. All parental forms respond with a highly significant increase of the shoot length to GA-application (about 135–190%). Hybrid plants from crosses 'CS/Gabo' and 'CS/Sun' did repond in a similar way, whereas the hybrid plant with dwarf phenotype both in interspecific (CS/V1 and CS/V10) and intraspecific (Gabo/Sun) crosses show no response to GA3 application. We concluded that hybrid dwarfness is not directly connect with gibberrellic acid pathway and has another nature.

Association mapping of aluminium tolerance loci in bread wheat.

Aluminium (Al) toxicity in acid soils remains a constant abiotic stress that affects wheat production in many parts of the world. Al^{+3} ions in small amounts can inhibit root growth thus negatively affecting the water and nutrient uptake by plants. A large body of research has documented the output from bi-parental QTL analyses, which mainly concludes a major effect QTL on chromosome 4DL and few minor loci. Here we report the results from a genome-wide association mapping.

The test material consisted of 96 winter wheat accessions originated from 21 countries and was genotyped with 874 DArT markers. Al tolerance was evaluated by a hematoxylin staining assay. The analysis identified two highly significant loci ($P<0.001$), one on chromosome 1DL and the other on 3BL. The locus on 1DL was assigned to a chromosomal bin, where we identified a relevant Al-tolerance gene candidate *Wali5* (wheat aluminium induced protein) among others. The 3BL locus was assigned to a chromosomal bin, where the possible candidates encoded reactive oxygen species, zinc finger proteins, and metallothioneines, which were reportedly involved in response to Al stress. When the significance threshold was reduced to $P<0.01$, several loci were detected on chromosomes 1A, 1D, 6A, and 7B. Some of the results are in parallel with prior reports and are strong candidates for future research to develop Al-tolerant cultivars suited to grow on highly acidic and Al-toxic soils worldwide.

Identification of QTL determining osmotic stress tolerance and seed parameters in durum wheat.

A mapping population was created from a cross between a drought-tolerant breeding line, Omrabi5, and a salt-tolerant line, Belikh2. A set of 114 recombinant inbred lines (RILs) was examined for their ability to deal with osmotic stress during germination and seedling stage. Seeds of the RILs were subjected to 12% polyethylene glycol (PEG) and compared for vigor and root, shoot, and coleoptile length with the untreated controls. Results were directed to a QTL analysis using 265 microsatellite markers mapped on 14 linkage groups of A and B genomes and spanning 2,864 cM.

Composite interval mapping revealed nine major QTL on chromosomes 1B, 3B, 4B, 6A, and 7B, including five QTL for coleoptile lengths explaining a phenotypic variation up to 28.6%.

Additionally tested seed parameters, such as 1,000-kernel weight, seed area, seed width, and seed length resulted in eight major QTL mainly on chromosome 7B, reaching an explained phenotypic variation up to 57.7%.

Preharvest sprouting and dormancy.

Three wheat populations (ITMI, D-genome introgression lines, and association panel) were studied in order to find responsible loci for dormancy and preharvest sprouting. In addition, two barley populations (OWB and Steptoe/Morex) were investigated. A classical quantitative trait locus analysis was combined with an association mapping approach. Many quantitative trait loci and marker trait associations could be detected on all seven chromosome groups of wheat and on the chromosomes 2H, 3H, 5H, 6H, and 7H of barley. Especially, the known regions on chromosomes 3A and 4A for wheat and 5H for barley were confirmed. Via a candidate homologue search and expressed sequence tag annotation, putative functions could be found. The *viviparous1* gene is located on chromosome 3A, which is associated to preharvest sprouting and dormancy. On chromosome 4A, a protein is detected that belongs to the aquaporin family. Aquaporins are responsible for water flow through the cell membrane. In barley, an association with the aleurain gene on chromosome 5H was found. The expression of aleurain is regulated by abscisic acid and gibberelic acid. From both hormones, an influence on dormancy and preharvest sprouting is known. We concluded that dormancy and preharvest sprouting are very complex traits regulated by multigenes and/or quantitative trait loci.

Susceptibility to wheat midge infestation.

A spring wheat panel consisting of 117 wheat accessions originated from the Gatersleben Genebank collection was cultivated in the field. The population was used for a genome-wide, association mapping analysis for resistance against oange and yellow wheat blossom midges. Midges were surveyed by white water traps. Highly significant marker trait associations (MTAs) were identified on 18 out of 21 chromosomes. For orange wheat midge adults and larvae, 43 and 25 MTA were detected, respectively. Alternatively, 22 and 19 MTA for yellow wheat midge adults and larvae, respectively, were identified. Assessment of ears stored in a freezer for larvae of wheat midges is done at present.

Drought tolerance at early stages in hexaploid wheat.

Two panels consisting of 96 winter and 117 spring wheats were subjected to osmotic stress induced by polyethylene glycol. Root, coleoptile, and shoot lengths and root and shoot dry weights were measured under stress and control conditions. Both panels will be used for a genome-wide, association mapping analysis in order to detect loci responsible for osmotic stress. In addition, the panels will be analyzed for drought tolerance at the adult-plant stage.

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ITEMS FROM HUNGARY

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Wheat season. After the extremely dry 2011 (263 mm), the drought continued in 2012. The total yearly precipitation in 2012 reached only 337 mm, compared to the average 550 mm. Due to the dry conditions, no disease epidemics occurred. The national wheat average reached only 3.7 t/ha, which is lower than the last 20-year average. Wheat still performed relatively much better than corn, which had only half the yield compared to the potential. The quality of the harvested wheat was good.

Breeding.

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New releases. Four Martonvásár winter wheat cultivars were registered in Hungary in 2012.

Mv Pengő (Mv 10-09) is a very high-yielding, early maturing, hard red winter wheat. The origin of the cultivar is 'Marsall/CF621'. Mv Pengő is supposed to replace Mv Marshall, the highest yielding Martonvásár wheat present on the market at the moment. The cultivar has good winter hardiness and good lodging resistance thanks to the presence of Rht1. Mv Pengő has better bread-making quality than that of Mv Marsall; its wet gluten content is 30–32%, Farinograph quality B1(A2), Alveograpg W value 220–300, with a low P/L ratio. Mv Pengő is moderately resistant to powdery mildew and leaf rust.

Mv Nádor (Mv15-09) is a very high-yielding, early maturing, dwarf cultivar designed for farmers applying high doses of fertilizer and seeking high yield. Under high-input conditions, Mv Nádor is capable of achieving record yields. Selected from the cross 'DI9812/Vekni', Mv Nádor inherited a high wet gluten content (34–36%) from Mv Vekni, with a dough quality characterized as B1 Farinograph category. Mv Nádor carries the T1B·1R translocation, and possess medium Alveograph and Extensograph quality. Mv Nádor has reliable winter hardiness, excellent lodging resistance, and is moderately resistant to leaf diseases.

Mv Kokárda (Mv09-09) is a high-yielding, soft red winter wheat with excellent agronomical properties from the cross 'Fleming/Amanda'. The cultivar has been selected for industrial utilization. Mv Kokárda has a low (23–26%) wet gluten content, flour water uptake lower than 50%, and very low Farinograph stability. Mv Kokárda has excellent resistance to lodging, leaf rust, and powdery mildew. Because of good adaptability, Mv Kokárda can be successfully grown under different yield levels.

Mv Pántlika (Mv24-09) is a medium-early, quality hard red winter wheat selected from the cross 'MV10-2000/Marsall', with a maturity time is similar to that of Mv Suba. In spite of its excellent quality, the productivity of Mv Pántlika is comparable to that of medium-quality, high-yielding cultivars. The cultivar has a high gluten content (34–37%), very good rheological quality, a A1–A2 Farinograph category, and 320–390 Alveograph W value.

Breeding for quality traits. ***β-glucan content of wheat.*** The effects of heat (H), drought (D), and the combination of the two stresses (H+D) on the dietary fiber content and composition (arabinoxylanand β-glucan) of three winter wheat cultivars (Plainsman V, Mv Magma, and Fatima 2) were determined. The stress treatments were applied on the 12th day after heading and continued for 15 days. We used the 1,000-kernel weight and protein content, β-glucan and arabinoxylan (AX), and the structure of AX β-glucan to determine the bt enzymatic fingerprint. Principal component analysis showed that heat and drought stress decreased the 1,000-kernel weight and the β-glucan contents of the seeds relative to the untreated control samples, whereas the protein and AX contents of the same samples increased when calculated on a dry weight basis. The highest amounts of AX and protein were in the H+D-stressed samples, with heat stress also increasing the water extractability of the AX. All the stresses decreased the β-glucan content of the seeds as well as the quantity of the DP3 and DP4 unit in all cultivars. However, whereas the content of AX content generally increased under all stresses, drought stress had negative effect on the AX content of the drought-tolerant Plainsman V. Fatima 2 behaved similar to Plainsman V with regard to drought tolerance, but was very sensitive to heat stress, whereas Mv Magma was the most resistant to heat stress.

Breeding for high arabinoxylan content. Cereals contain some promoting bioactive components, but their advantages have not or only partly been utilized. Our goal is to develop different cereal-based, functional products for the bakery, pasta-, flour, and the ready-to-eat food market keeping in mind the requirements of regional Hungarian tradition and easy utilization. The first step is to develop an excellent raw/plant material for the processing industry. During the 'Healthgrain' project, the exotic, Chinese wheat cultivar Yumai-34 was identified to contain extremely high quantities of water-extractable arabinoxylans (WE–AX) in the flour compared to wheat cultivars now in commercial worldwide. This property was utilized to start a breeding program to produce wheat cultivars with good adaptability and a high content of dietary fiber. We studied whether arabinoxylans could effectively be used for breeding purposes, and the improvement of the nutritional properties could have an influence on the yielding potential and processing quality of wheat. The F_7–F_8 generations of Yumai-34 with Mv-Emese, Mv-Mambo, Ukrainka, Lupus, or Courtot had already been developed. The greatest number of crossing lines was produced with Ukrainka, whereas the highest WE–AX content, similar to that

of the control (~1% d.m.), was analyzed in lines from the 'Lupus/Yumai-34' cross. The increase in WE–AX content had a negative effect on the yield (r 1% = –0.427), heading date (r 1% = –0.510), and the Zeleny sedimentation (r 5% = –0.299) of the breeding lines in all the three years of this study. These results will help to develop the selection strategy of high dietary fiber lines by focusing on the early maturing breeding lines. Recently, 36 lines were planted in three replications to check the homogenity and stability of the selected lines, especially with regard to arabinoxylan content.

Breeding for starch properties. The ratio of amylose to amylopectin in starch is decisive for determining the physicochemical properties of the starch, susceptibility to enzymatic hydrolysis, gelling, and pasting behavior, which could be of technological quality importance. Both low- and high-amylose wheat are useful for the different effects they have on flour or semolina functionalities. High-amylose starches are used widely as thickeners and strong gelling agents, but they also have a positive effect on the dietary fiber content of the flour by increasing the level of starch resistant to human enzymatic digestion after consumption. The resistant starch plays significant role inside the intestine, protecting against diseases such as colon cancer, type-II diabetes, and obesity.

In order to produce healthier wheat genotypes with high amylose content (40%) and good agronomical properties, triple mutant lines (*Sgp-A1*, *Sgp-B1*, and *Sgp-D1*) have been created (Lafiandra et al.). These lines were multiplied and crossed with five current commercial varieties of bread wheat (Solstice, Lona, Koreli, Ukrainka, and Yumai-34) to introgress the three mutations. Three backcrosses were carried out, during which marker-assisted selection was made in each generation in order to select the possible triple mutant lines for backcrossing. Finally, in the BC_3 generation, 0.82% of the 380 studied lines were found to be triple mutants, 13% mutant and/or heterozygote for a given allele, and 30.79% of the lines were possible double mutants. Forty-seven possible triple mutant lines were analyzed in details for amylose content, and 17 genotypes were found to have nearly 40% of amylose content (37–44%) comparable to the mutant control. From these high-amylose lines, three had already been mutants for all the three alleles. Seeds of these lines will be multiplied and studied for processing quality by measuring starch viscosity properties.

Pannon wheat R&D system. The Pannon wheat R&D system is a complex, regional program in which internationally compatible quality criteria were developed in order to improve the exportability of the Hungarian wheat cultivars by taking into consideration the expectations of the processing industry and the consumers. As a next step, drought-resistant, hard wheat cultivars with high protein and gluten content were developed that are sufficient to the quality criteria. To reach the appropriate quality, a plant management and consulting system also was developed for supporting the work of the farmers, and a fast screening and transfer system was developed to help millers and the processing industry to identify the appropriate seed resources.

Based on these results, critical points setting back the realization of the whole system were identified. These are the questions of extra production costs, the quality stability of the cultivars, the different scaling of the real seed production, or the realization of raw material quality in the processing industry.

In overcoming these problems, the development of a new, reconsidered, national strategy was begun in 2009. In the frame of this project, new breeding lines with stable quality were developed with special attention relating to the kernel type and the rheology. Cultivars and breeding lines were tested in large-scale experiments, agrotechniques optimized, and the stability of the quality monitored. Confirmation of the benefits of the Pannon wheat quality and its realization in the processing technologies has already begun.

Disease resistance studies. ***Stem rust resistance.*** Among the genotypes carrying designated *Sr* genes, lines with *Sr28*, *Sr30*, *Sr34*, or *Sr36* were highly resistant to stem rust, and additionally some more major resistance genes (*Sr6*, *Sr9e*, *Sr17*, and *Sr31*) provided effective resistance against the spread of the pathogen.

Powdery mildew virulence survey. Studies on the composition of the wheat powdery mildew population showed that race 77 was present even in higher ratio (81.5%), than in 2011, followed by the race 76 (11.2%). Race 51, which infected all the differentials of Nover's set, appeared in even lower frequency than in the previous year (2.1%). The virulence complexity of the pathogen population was 5.16, which represented a further decline compared to previous years.

Fusarium head blight resistance. The molecular marker linkage map, based on a population with Ning 8331 (resistant parent) and Martonvásári 17 (moderately susceptible parent) origin, was constructed. Two major QTL linked with FHB resistance were identified in Ning 8331. One of them, with a putative effect on type-II FHB resistance, is located on the 2DL chromosome arm. The effect of 3BS QTL (*Fhb1*) was proven both in sprayed and in single floret inoculated experiments.

Abiotic stress resistance studies. Investigations on the osmotic stress tolerance of the dihaploid population in the seedling stage showed variation in the sensitivity of DH lines to the 15% PEG-6000 solution. The mean value of shoot length and shoot and root fresh and dry weight of the population decreased considerably in response to PEG treatment compared to control values.

The DArT analysis of the RIL population originating from the cross between the heat stress tolerant Mv Magma and the sensitive Plainsman V wheat cultivars has been completed resulting in 967 DArT markers with known chromosomal locations. Significant differences in the marker coverage of the three wheat genomes were noted; more than 50% of the markers are located on the B genome, 29% on the A genome, and only 20% on the D genome. The marker coverage of the homologous chromosome groups was the highest for groups 7 and 3 and the lowest for groups 4 and 5. Chromosome 1B had the highest number of markers (153), whereas chromosome 6D had only two DArT markers.

Investigations on the water use efficiency of the small-grain cereals revealed that water uptake was most intense during the heading. A connection also was found between the length of the vegetation period and the developmental stage in which plants were most sensitive to water shortages. Water withdrawal during the stage of the first node appearance resulted in a significant yield loss of the cultivars, which had a relatively rapid rate of development. For genotypes with a longer vegetation period, however, the highest sensitivity to water shortage was recorded at the grain-filling stage. In a greenhouse experiment, the water use efficiency (grain yield/total water use) of the winter wheat genotypes was found to be between 0.7 and 1.6 at the optimum water supply level. Drought stress treatment resulted in a decline of the water use efficiency in different phenological stages. The WUE values were between 0.56 and 1.39 at the stage of the first node appearance, 0.28–1.28 at heading, and 0.16–1.18 at the grain-filling period.

Growth of wheat plants at elevated CO_2 level (EC, 750 µmol/mol) was found to result in a more relaxed state of the antioxidant enzyme system in the plants of all three cultivars. The very low, uniform activity values of glutathione reductase, glutathione-S-transferase (GST), ascorbate peroxidase (APX), and guaiacol peroxidase were caused by activity decreases in cultivars with higher ambient values. A parallel increase in catalase (CAT) was recorded in two cultivars. Despite the fact, that the sensitive cultivar had better tolerance to drought at elevated CO_2 levels, drought triggered a more pronounced, general response in the antioxidant enzyme system at EC, leading to very high values of APX, CAT, and GST that were very similar in all the cultivars. These high values were due to a higher level of oxidative stress under drought at EC; because the decline in net photosynthesis was faster compared to ambient values, while there was no change in the rate of electron transport.

Molecular breeding. Using diagnostic molecular markers, the allele compositions of the *Vrn-A1*, *Vrn-B1*, *Vrn-D1*, *Ppd-B1*, and *Ppd-D1* genes were determined in a wheat collection consisting of 683 genotypes. On the basis of these five genes, the cultivar collection (521 European, 62 Asian, 6 African, 90 American, and 4 Australian cultivars and lines) could be divided into 24 combination groups. The distribution frequencies of the allele groups differed not only from one continent to the other, but also between various geographic regions of Europe and between the countries of Central Europe. The dominant (spring) *VRN-A1*, *VRN-B1*, and *VRN-D1* alleles were present at a frequency of 6–7% in the total set of the genotypes. The *VRN-A1* and *VRN-B1* alleles were more frequent in cultivars from Australia, Africa, and America, whereas the *VRN-D1* allele was more common in Asian cultivars. The semi-dominant *Ppd-D1*a photoperiod-insensitive allele was present in 57% of the cultivars and most frequently observed in Asian, Australian, and European cultivars. Within Europe, the photoperiod-insensitive allele was dominant in the eastern, southern, and southeastern regions (93%, 85%, and 93%, respectively), and the photoperiod-sensitive allele was more frequent (63%) in western Europe. The two alleles of *Ppd-D1* gene were present in almost equal proportions (52% vs. 48%) in central Europe. The *Ppd-B1* photoperiod-insensitive allele was present in 22% (151) of the genotypes, all representing breeding materials from the Asian, American, and European continents; 38 genotypes from Asia (61%), 23 from America (26%), and 88 from Europe (17%). The vast majority of the European genotypes bearing this allele type originated from central and southeastern Europe.

The molecular marker linkage map of the 'MvTD10-98/PWD1216' population was constructed using AFLP (six primer combinations) and DArT (~2,000 probes) markers. The map contains 462 markers arranged in 21 linkage groups and extends over 1,784 cM. The map is based on 347 DArT markers, which assigned the 21 linkage groups to 14 chromosomes. Four different QTL linked with yellow index (Minolta b*) were identified on chromosome 1B, 3B, 5B, and 7A. The strongest QTL, located on the distal part of the short arm of chromosome 7A, explained 17.3% of the phenotypic variation, and the remaining three QTL explained 7.3–11.9% of the variation of the yellow index. The significant effect of the QTL located on the chromosome 3B and 7A could be detected in five consecutive years (2007–11). The alleles with positive effect were in all cases originated from the PWD1216 parent.

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Identification and phenotypic description of new wheat – six-rowed, winter barley disomic additions. To increase the allelic variation in wheat–barley introgression lines, new wheat–barley disomic addition lines were developed containing the 2H, 3H, 4H, 6H, and 7H chromosomes of the six-rowed Ukrainian winter barley Manas. This cultivar is agronomically much better adapted to the central European environment than the two-rowed spring barley Betzes previously used. A single 'Asakaze/Manas' wheat × barley hybrid plant was multiplied in vitro and one backcross plant was obtained after pollinating 354 regenerated hybrids with wheat. The addition lines were selected from the self-fertilized seeds of the 16 BC2 plants using genomic in situ hybridization. The addition lines were identified by fluorescence in situ hybridization (FISH) using repetitive DNA probes (HvT01, GAA, pTa71, and Afa family), followed by confirmation with barley SSR markers. The addition lines were grown in the phytotron and in the field, and morphological parameters (plant height, fertility, tillering, and spike characteristics) were measured. The production of the disomic additions will make it possible to incorporate the DNA of six-rowed winter barley into the wheat genome. Addition lines are useful for genetic studies on the traits of six-rowed winter barley and for producing new barley dissection lines.

High-resolution, molecular cytogenetic techniques in plants: pachytene- and fiber-FISH. FISH is the most versatile and accurate molecular cytogenetic technique for determining euchromatic-heterochromatic boundaries and the locations of repetitive and single-copy DNA sequences and of chromosome-specific BAC clones on chromosomes. The combination of cytogenetic and genetic methods yields a high-resolution physical map. FISH allows direct mapping of specific DNA sequences inside the cell (interphase nuclei), along meiotic pachytene chromosomes and isolated chromatin (DNA fibers). The increased sensitivity of the technique, and its ability to detect gene locations provide a powerful research tool for genetic and prebreeding studies. FISH-based, physical mapping plays an important role and is increasingly

used for studies at the cytological level on the chromatin organization that controls gene expression and regulation. The mini-review describes some of the benefits of alternative FISH-based techniques and their application for studying plant chromosomes and genomes.

Karyotypic analysis of *Triticum monococcum* using standard repetitive DNA probes and simple-sequence repeats. *Triticum monococcum* represents an important source of useful genes and alleles that it would be desirable to use in wheat breeding programs. The well-defined landmarks on the A^m chromosomes could accelerate the targeted introgression of *T. monococcum* chromatin into the wheat genome. FISH, using the repetitive DNA probes pSc119.2, the Afa family, and pTa71, showed that the pSc119.2 probe was not suitable for the identification of A^m chromosomes. In contrast, the whole set of A^m chromosomes (especially chromosomes 1, 4, 5, and 7) could be discriminated based on the hybridization pattern of pTa71 and the Afa family. In situ hybridization with microsatellite motifs (GAA, CAG, AAC, and AGG) proved that SSRs represent additional landmarks for the identification of A^m chromosomes. The most promising SSR probes were the GAA and CAG motifs, which clearly discriminated the $6A^m$ chromosome and, when used in combination with the Afa family and pTa71 probes, allowed the whole set of A^m chromosomes to be reliably identified. In conclusion, FISH using the repetitive DNA probes of the Afa family and pTa71, combined with SSR probes, makes it possible to identify the A^m chromosomes of *T. monococcum* and discriminate them from A^u chromosomes in the polyploid wheat background.

Detection of various U and M chromosomes in wheat–*Aegilops biuncialis* hybrids and derivatives using FISH and molecular markers. The aim of the study was to select wheat–*Ae. biuncialis* addition lines with *Ae. biuncialis* chromosomes differing from those that were introgressed into the wheat–*Ae. biuncialis* addition lines produced earlier in Martonvásár, Hungary. During the experiments, new wheat–*Ae. biuncialis* addition lines with chromosomes $2U^b$, $3U^b$, $5U^b$, $6U^b$; $7U^b$, $5M^b$, $6M_b$, and $7M^b$ were selected. The $2U^b$ disomic addition line is relatively stable; 91% of the progenies contain this chromosome pair. The $6M^b$ disomic addition line proved to be dwarf and sterile, but still exists as a monosomic addition line. Progenies analyzed from the $6U^b$ monosomic addition line did not carry the $6U^b$ chromosome. One plant containing the $5U^b$, $3U^b$, and $7U^b$ chromosomes and one plant with chromosomes $5M^b$, $6M^b$, and $7M^b$ showed very low fertility. Each of the plants produced a single seed, but seeds of the parent plants are still available. Line 49/00 carried a submetacentric *Ae. biuncialis* chromosome pair and the chromosome number 44 has been constant for several generations. After FISH, no hybridization site was observed on the *Ae. biuncialis* chromosome pair using the pSc119.2 and Afa family repetitive DNA probes, so it was not possible to identify the *Ae. biuncialis* chromosome pair. However, using wheat SSR markers and the (GAA)n microsatellite DNA probe allowed it to be characterized more accurately. These new lines facilitate gene transfer from *Ae. biuncialis* into cultivated wheat and the selection of U- and M-genome-specific wheat SSR markers.

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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.

Pedigree	Biological yield			Grain yield		
	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.

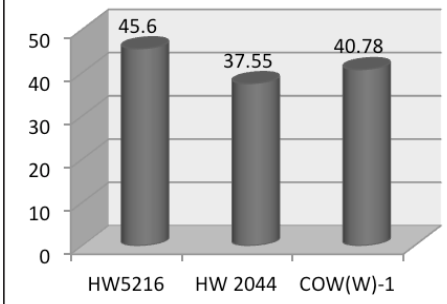

Fig. 1. Comparative mean grain yield of *T. aestivum* cultivar HW 5216 and checks over four years of testing at multiple locations.

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)
Rome, Italy.

AGENZIA PER LA SPERIMENTAZIONE TECNOLOGICA E LA RICERCA AGROAMBIENTALE (ASTRA)
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.

CSBMV pressure was relatively high, as testified by the high mean symptom scores (≥ 3.0) recorded for eight of the 31 cultivars assayed (Table 1). Low mean symptom scores (≤1.0) accompanied by low ELISA values were recorded for the cultivars Dylan, Ramirez, Biensur, Tirex, Serafo Nick, and Marco Aurelio. The latter two had not been tested for CSBMV resistance before. Five of the other new entries, i.e., Colombo, Core, Cesare, Trapezio, and Massimo Meridio, showed relatively mild symptom scores (1.0–2.0) and relatively low ELISA values, and three (Miradoux, Mimmo, and Odisseo) proved susceptible.

Table 1. Response to ereal soil-borne mosaic virus of 31 durum wheat cultivars grown near Bologna, Italy, in 2011–12. Items with the same letter(s) are statistically similar. Symptom severity was rated on a 0–4 scale and are the mean of four dates.

Cultivar	Mean symptom severity score	ELISA value	Grain yield (t/ha)	Plant height (cm)	Kernel weight (g)	Test weight (kg/hl)	Days-to-heading (from April 1)
Anco Marzio	2.46 cd	0.394 df	5.21 ej	91.3 ad	36.7 dg	72.4 ae	33.7 hk
Achille	3.65 a	1.634 ac	3.51 jl	66.0 i	31.1 hk	71.6 af	37.0 bc
Biensur	0.90 hk	0.012 f	7.64 ab	80.7 fg	34.0 ej	73.2 ad	36.0 cf
Cesare	1.61 eg	0.573 cf	3.85 hl	94.0 ac	38.7 ce	67.3 fh	35.0 dh
Claudio	2.95 ac	1.783 a	3.75 il	76.7 gh	34.1 ej	72.2 af	36.7 bd
Colombo	1.24 fj	0.008 f	6.13 bg	80.3 fg	37.9 dg	73.7 ad	37.0 bc
Core	1.53 eh	0.006 f	7.47 ac	93.0 ac	39.6 bd	74.9 ab	31.3 ln
Duilio	1.63 eg	0.356 df	6.10 bg	94.3 ac	38.3 cf	72.1 af	31.3 ln
Dylan	0.55 k	0.008 f	5.48 di	92.3 ac	30.7 ik	66.0 gh	33.0 il
Iride	1.93 df	0.002 f	5.27 ej	83.0 dg	33.2 gj	67.7 eh	33.0 il
Ismur	3.40 ab	1.799 a	4.58 fk	64.0 i	30.1 jk	72.3 af	36.0 cf
Kanakis	2.81 bc	1.202 ad	5.23 ej	91.0 ad	37.3 dg	72.9 ad	35.7 cg
Karur	2.83 bc	1.193 ad	5.47 di	82.7 eg	36.8 dg	70.8 bg	38.0 b
Levante	1.54 eh	0.010 f	8.48 a	94.7 ac	35.5 dh	76.1 a	33.0 il
Liberdur	3.32 ab	0.678 bf	4.56 fk	66.3 i	35.7 dh	71.9 af	36.3 be
Marco Aurelio	0.98 gk	0.322 df	7.62 ab	87.7 cf	44.4 a	73.5 ad	31.0 mn
Massimo Meridio	1.67 eg	0.003 f	6.01 bg	91.3 ad	43.7 ab	71.9 af	33.0 il
Mimmo	3.14 ac	1.158 ae	3.18 kl	80.7 fg	38.3 cf	64.8 h	34.0 gj
Miradoux	2.98 ac	0.593 cf	4.03 hl	75.7 gh	42.7 ac	70.9 bg	39.7 a
Normanno	1.41 ei	0.640 bf	4.91 fk	90.3 ae	33.2 gj	68.8 dh	31.3 ln
Odisseo	3.28 ab	0.466 df	5.50 di	80.7 fg	38.0 dg	73.3 ad	33.3 hk
Ramirez	0.78 ik	0.007 f	6.27 bf	92.3 ac	33.2 gj	75.4 ab	35.7 cg
Saragolla	1.42 ei	0.042 ef	5.72 ch	88.3 bf	34.5 ej	67.5 eh	32.3 jm
Sculptur	3.08 ac	1.195 ad	5.47 di	71.0 hi	35.0 di	72.3 af	34.0 gj
Serafo Nick	0.68 jk	0.017 f	7.22 ad	96.7 ab	37.3 dg	74.4 ac	33.0 il
Simeto	3.05 ac	1.263 ad	4.07 hl	71.7 hi	35.5 dh	67.4 eh	33.3 hk
Svevo	1.98 de	0.007 f	6.42 bf	97.3 a	38.3 cf	72.8 ad	30.3 n
Tirex	0.92 hk	0.010 f	6.93 ae	95.3 ac	33.5 fj	75.2 ab	32.0 kn
Trapezio	1.63 eg	0.004 f	8.39 a	93.3 ac	38.0 dg	74.5 ab	34.3 fi
Yelodur	1.86 df	0.422 df	4.32 gl	75.7 gh	36.3 dg	68.7 dh	36.7 bd
Grazia	3.06 ac	1.724 ab	2.57 l	72.0 hi	27.7 k	69.3 ch	34.7 ei

Mean ELISA value and mean symptom severity score were correlated significantly (0.809**), and both resistance parameters were significantly correlated (P 001) with grain yield, plant height, and heading date, but not with kernel and test weight (Table 2, p. 36). A regression analysis estimated quite accurately the agronomic CSBMV effects corresponding to different symptom severity scores; in the 2011–12 season, for instance, a mean symptom score of 3.5 was

associated with a grain yield loss of about 49%, a plant height reduction of 27%, and a heading delay of about 4 days (Table 3). As in previous experiments, results indicated that even mild symptoms cause appreciable grain yield losses.

Table 2. Simple correlation coefficients between mean symptom severity, mean ELISA value, and various agronomic characters for 31 durum wheat cultivars grown in a field with cereal soil-borne mosaic virus near Cadriano (Bologna), Italy, during 2011–12. Items with a ** are significantly correlated; all others are nonsignificant.

	ELISA value	Grain yield	Plant height	Heading date	Kernel weight	Test weight
Symptom severity	0.809**	–0.673**	–0.740**	0.437**	–0.139	–0.208
ELISA value	—	–0.732**	–0.710**	0.433**	–0.376*	–0.272

Table 3. Estimated effects of cereal soil-borne mosaic virus on grain yield, plant height, and heading date for different symptom severity scores (Cadriano (Bologna), Italy, 2011–12).

	Grain yield loss		Plant height reduction		
Disease score	t/ha	%	cm	%	Heading delay (days)
0.5	0.54	6.9	3.9	3.9	0.5
1.5	1.62	20.8	11.8	11.8	1.6
2.5	2.69	34.7	19.7	19.6	2.6
3.5	3.77	48.6	27.6	27.4	3.7

Agro-biological effects of cereal soil-borne mosaic virus over 10 seasons.

V. Vallega (CRA-QCE, Rome), C. Rubies-Autonell (DSA, Bologna), A. Sarti (ASTRA, Faenza) and R. Canestrale (CRPV, Imola).

Different sets of durum wheat cultivars were evaluated for resistance to cereal soil-borne mosaic virus (CSBMV) at two fields with the virus situated near Minerbio and Cadriano (Bologna), in 10 seasons between 1996 and 2010. Five of the 15 trials programmed for this period could not be carried out due to the lack of adequate funds and/or sufficiently uniform fields. The cultivars were evaluated for CSBMV resistance on the basis of symptom severity, DAS-ELISA readings, and agronomic performance, i.e., grain yield, plant height, kernel weight, test weight, and heading date. Each trial was comprised of 30–33 cultivars, grown in 10-m^2, solid-seeded plots distributed in the field according to a randomized block design with three replicates. A total of 124 durum wheat cultivars were assayed. In each season, symptom severity was evaluated on three or more dates using a 0–4 scale where 0–1.0 = no or slight symptoms, 1.1–2.0 = mild mottling and stunting, 2.1–3.0 = mottling and stunting, and 3.1–4.0 = severe mottling and stunting with virus-killed plants. Symptom severity score and grain yield were highly and significantly correlated in all seasons, thus offering the opportunity to estimate the effect of various levels of CSBMV symptom severity on grain yield under diverse conditions using simple linear regressions. On average, symptom severity scores of 3.5, 2.5, 1.5, and 0.5 were associated with grain yield losses of 53, 38, 23, and 8%, respectively (Table 4). Symptom scores also were significantly correlated with the other four agro-biological characters investigated, but not in all seasons. By and large, symptom scores of 3.5 were associated with plant height, kernel weight, and test weight reductions of about 25, 20, and 10%, respectively, and with a heading delay of about 3 days.

Table 4. Estimated effects of cereal soil-borne mosaic virus on grain yield for different symptom severity scores in ten seasons (Minerbio 1996–97 and Cadriano 2001–10).

	Symptom severity score			
Season	0.5	1.5	2.5	3.5
1995–96	7.0	21.1	35.2	49.3
1996–97	10.4	31.2	52.0	72.7
2000–01	8.4	25.1	41.9	58.7
2001–02	6.7	20.1	33.5	46.9
2002–03	7.6	22.9	38.1	53.4
2003–04	7.2	21.5	35.9	50.2
2004–05	5.9	17.8	29.6	41.4
2006–07	9.6	28.7	47.8	66.9
2008–09	5.2	15.7	26.1	36.5
2009–10	7.7	23.2	38.6	54.0
Mean	7.6	22.7	37.9	53.0
Minimum	5.2	15.7	26.1	36.5
Maximum	10.4	31.2	52.0	72.7

Response to CSBMV of 133 durum wheat cultivars assayed from 1996 to 2011.

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A total of 133 durum wheat cultivars were grown in different trials over eleven seasons (from 1996 to 2011) at two fields near Bologna with cereal soil-borne mosaic virus (CSBMV). The cultivars were evaluated for resistance to CSBMV on the basis of grain yield (except in 2010–11), symptom severity, and DAS-ELISA readings. Each trial was comprised of

Table 5. Response to cereal soil-borne mosaic virus of 133 durum wheat cultivars assayed in trials near Bologna (Minerbio 1996–97 and Cadriano 2001–11), Italy, and the number of years in which each cultivar was tested.

Resistant				Moderatately resistant			
Cultivar	**Years**	**Cultivar**	**Years**	**Cultivar**	**Years**	**Cultivar**	**Years**
Alemanno	2	Lloyd	3	Ariosto	1	Latinur	4
Ares	4	Louxor	1	Arnacoris	3	Neolatino	4
Asdrubal	1	Meridiano	8	Artemide	1	Normanno	6
Baio	1	Nefer	1	Avispa	3	Orfeo	1
Biensur	3	Neodur	8	Brindur	1	Peleo	**1**
Campodoro	1	Parsifal	2	Canyon	1	Portofino	2
Ceedur	1	Pharaon	2	Catervo	1	Pr22d89	3
Colorado	5	Pietrafitta	2	Chiara	1	Preco	1
Dario	1	Provenzal	5	Cosmodur	2	Rusticano	1
Dylan	7	Ramirez	1	Duilio	11	San Carlo	5
Giusto	1	Saragolla	4	Fiore	2	Sfinge	1
Hathor	1	Solex	7	Flavio	2	Svevo	3
Kanakis	1	Tiziana	3	Gianni	5	Tirex	3
Levante	6	Valerio	1	Grecale	2	Torrese	1
Ignazio	1	Valsalso	1	Imhotep	3	Virgilio	1
				Iride	10	Vitomax	3
				Isildur	2	Vitron	2
				K26	1	Yelodur	1
Moderatately susceptible				**Susceptible**			
Cultivar	**Years**	**Cultivar**	**Years**	**Cultivar**	**Years**	**Cultivar**	**Years**
Appio	2	Norba	1	Achille	4	Karur	3
Aureo	1	Ofanto	2	Agridur	1	Liberdur	3
Claudio	9	Perseo	1	Anco Marzio	5	Marco	2
Colosseo	4	Plinio	1	Balsamo	2	Orobel	7
Creso	10	Portobello	1	Bronte	1	Peres	1
Dorato	1	Principe	1	Cannavaro	1	Platani	2
Duetto	2	Quadrato	4	Cannizzo	3	Portorico	5
Ermecolle	1	Sculptur	1	Capri	1	Pr22d40	1
Exeldur	2	Sorrento	1	Carioca	1	Prometeo	2
Gardena	2	Torrebianca	5	Casanova	2	Severo	2
Giotto	3	Tresor	2	Ciccio	5	Simeto	11
Giove	1	Vendetta	2	Ciclope	1	Sorriso	1
Italo	2	Verdi	3	Cirillo	3	Trionfo	2
Ixos	3	Virgilio	2	Concadoro	1	Tripudio	2
Minosse	2	Zenit	2	Derrick	2	Vesuvio	3
				Giemme	2	Vetrodur	3
				Granizo	1	Vettore	2
				Grazia	8	Vinci	1
				Ismur	1		

30–33 cultivars. The data collected for each cultivar and for each of the three parameters in each season from 1995 to 2005 were indexed as a percent of the highest value observed among all the cultivars assayed in that season and then averaged to minimize the confounding effects of differences in disease pressure between years. For various reasons, including the lack of agronomic data for the 2010–11 season, the cultivars assayed in the subsequent trials (2007 to 2011) could not be classified according to the same criteria. On the other hand, because the new entries were grown along with cultivars already assayed for CSBMV resistance in other seasons, there was ample opportunity to adequately classify their response to CSBMV by the use of numerous direct comparisons and, thus, produce a synoptic table comprising all the 133 cultivars assayed (Table 5, p. 37). The CSBMV responses presented in the table are obviously most dependable for cultivars assayed in numerous trials (up to 10 or 11 in the case of Duilio, Iride, Creso, and Simeto), whereas those based on the results of one trial only are merely indicative. We note that although nearly half of the 133 cultivars listed carry a major gene for CSBMV resistance located on the short arm of chromosome 2B, none were immune to CSBMV infection and only 16 consistently showed high levels of resistance.

ITEMS FROM MEXICO

NATIONAL INSTITUTE FOR FORESTRY, AGRICULTURE, AND LIVESTOCK RESEARCH (INIFAP–CIRNO)
Campo Experimental Valle del Yaqui, Apdo. Postal 155, km 12 Norman E. Borlaug, entre 800 y 900, Valle del Yaqui, Cd. Obregón, Sonora, México CP 85000.

CEVY Oro C2008: Yield and quality performance in experimental and semicommercial plots and status of area grown with this durum wheat cultivar in southern Sonora, Mexico.

Guillermo Fuentes-Dávila, Pedro Figueroa-López, Gabriela Chávez-Villalba, José Luis Félix-Fuentes, Miguel Alfonso Camacho-Casas, and Alberto Borbón-Gracia.

Abstract. Commercial durum wheat cultivar CEVY Oro C2008, developed for the wheat-producing areas of northwest Mexico, is spring type and resistant to leaf rust, with an experimental average grain yield of 5.6 t/ha, test weight of 83.0 kg/hl, 13.5% protein, and 28.1 points of the Minolta b value. CEVY Oro C2008 averaged 7.1, 7.76, and 7.2 t/ha in 2008–09, 2009–10, and 2010–11, respectively, with a maximum yield potential of 8.7 t/ha, in commercial fields of cooperating wheat producers from southern Sonora, and an average of 29.08 points of the Minolta b value. After its release, this cultivar was cultivated in 3,233 ha in southern Sonora during 2009–10, 6,161 ha during 2010–11, 931 ha during 2011–12, and in 2012–13 it was cultivated only in 185 ha. Cultivar CIRNO C2008, which has a high yield potential, was cultivated in 3,256, 87,105, 154,915, and 196,295 ha during those four seasons. Despite research efforts to diversify the sources of resistance to leaf rust and to improve quality, wheat production and comercialization are still based on grain yield, despite the risks that monocultivar culture represents in a region where leaf rust is endemic with the appearance of new races of the causal agent.

Introduction. Worldwide production of durum wheat in 2009 was 686.6 x 10^6 tons (FAO 2011a). China was the main producer, with 115.1 t, followed by India with 80.6 t. Mexico produced 4.1 x 10^6 tons from which 1.1 x 10^6 were exported (FAO 2011b). Of the area grown with wheat in Mexico, 63.9% (457,541 ha) corresponded to the states of Sonora, North Baja California, and Sinaloa, during the fall–winter 2008–09 crop season, with an estimated value of US$646 x 10^6 (SIAP 2011) (Exchange rate 1:12, May 2013). During the 2010–11 crop season, 288,766 ha were grown with wheat in southern Sonora, 70% corresponded to durum wheat predominating cultivars CIRNO C2008 and Átil C2000, and bread wheat cultivar Tacupeto F2001 (OEIDRUS 2011). In northwest Mexico, spring wheat is grown during the fall–winter season under irrigation. Durum wheat is mostly cultivated in the state of Sonora; since the 2001–02 crop season, this type of wheat has occuppied more than 70% of the area grown with wheat (Camacho et al. 2004). The preference for durum wheat by farmers in northwest Mexico was greatly influenced by the implementation of federal quarantine No. 16 against Karnal bunt of wheat (SARH 1987), because durum wheat is more tolerant to this disease than bread wheat. Durum wheat also has greater yield potential, better value in the international market, and, at that time, the durum

wheat cultivar grown Altar C84 was resistant to leaf rust. This disease is endemic in the region and develops rapidly on susceptible cultivars giving rise to new and more virulent races (Figueroa et al. 2010). The durum wheat cultivar mostly grown in the region up to crop season 2003–04 was Altar C84, despite the loss of its resistance to the new race BBG-BN (Singh et al. 2004), which affected production during the 2000–01 and 2001–02 crop seasons (Figueroa-López et al. 2002). Durum wheat cultivar Júpare C2001 (Camacho-Casas et al. 2004), resistant to the leaf rust races that affected durum wheat cultivars released until 2000, dominated the area grown with wheat in southern Sonora from 2003–04 to 2008–09, reaching 119,327 ha (42.3%) during that last season (Fig. 1). However, leaf rust overcame the resistance of this cultivar, therefore efforts are not only focused on diversifying the source of resistance to this disease, but also to increase grain pigment, an important characteristic of quality for the export market. Our objective is to present the current status of durum wheat cultivar CEVY Oro C2008, released for commercial use in northwest Mexico.

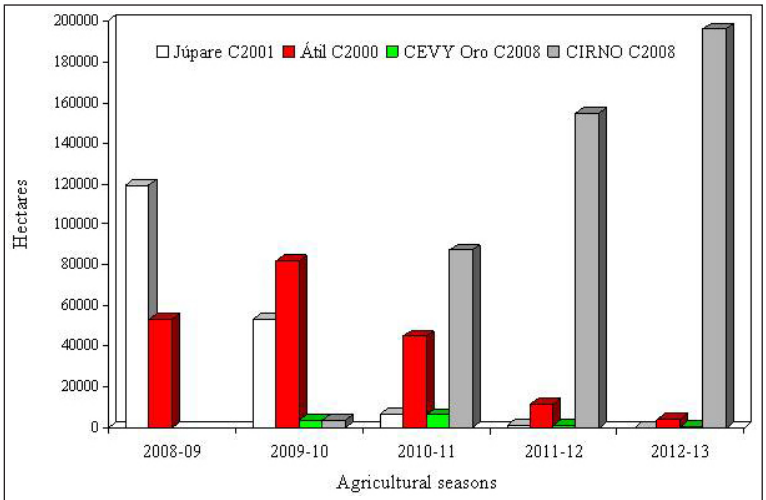

Fig. 1. Area (ha) sown with several durum wheat cultivars in southern Sonora, Mexico, during the 2008–09 to 2012–13 agricultural seasons.

Origin of CEVY Oro C008. The collaborative project between the Norman E. Borlaug Experimental Station, which belong to the Mexican National Institute For Forestry, Agriculture, and Livestock Research (INIFAP), and the International Maize and Wheat Improvement Center (CIMMYT) has the objectives of improving the cost/benefit and competitiveness of wheat by releasing cultivars with better yield, quality, resistance to disease, and efficient water use (Figueroa-López et al. 2011a). As a result of this project, the line 'SCRIP_1//DIPPER_2/ BUSHEN_3/4/ARMENT//SRN_3/NIGRIS_4/3/ CANELO_9.1' was released under the name CEVY Oro C2008 (Fuentes-Dávila et al. 2010), which is a durum wheat cultivar originated from hybridizations made in the Durum Wheat Breeding Program of CIMMYT. The cross and selection history is CDSS02Y00381S-0Y-0M-19Y-0M. CEVY Oro C2008 has the registration TRI-111-240209 in the National Catalogue of Plant Cultivars of the National Seed Inspection and Certification Service.

General characteristics of CEVY Oro C2008. Cultivar CEVY Oro C2008 is a spring wheat with a biological cycle that averages 121 days to physiological maturity under three complementary irrigations and depending on the sowing date (Fuentes-Dávila et al., 2010). Heading takes from 74 to 92 days with an average of 81. CEVY Oro C2008 is tall with a average height of 93 cm, a maximum of 105, and minimum of 85. Plant growth habit is erect and shows null or low frequency of recurved flag leaves. Grain shape is semi-elongated, 7.26 mm long, 3.0 mm wide, with an average weight of 52 mg. Grain coloration when treated with phenol is nil or very light. CEVY Oro C2008 is resistant to the leaf rust races prevailing in the wheat-producing areas of northwest Mexico, therefore, farmers will not have to depend on application of fungicides for control. Leaf rust in the most important disease in Mexico economically, historically causing 30–60% loss according to the cultivar and weather conditions (Villaseñor et al. 2003). CEVY Oro C2008 also is resistant to Karnal bunt.

Yield and quality, experimental evaluation. CEVY Oro C008 was evaluated at the Norman E. Borlaug Experimental Station (CENEB) in the Yaqui Valley, Sonora, during the fall–winter 2006–07, 2007–08, and 2008–09 crop seasons with three complementary irrigations; average experimental yield was 5.97, 4.79, and 5.96 t/ha, respectively. The maximum yield obtained was 6.53 t/ha with a 15 November sowing date with three complementary irrigations. Based on the statistical analysis, the best dates for sowing this cultivar are between 15 November and 1 December 1. No yield differences were noted between the first two sowing dates, but late ones do (15 December and 1 January).

Several physico-chemical parameters affect the industrial quality of durum wheat cultivars, however, protein content and quality and the pigment present in the endosperm of the grain affect noticeably the parameters considered for evaluation of semolina used for pasta making. Although protein content and quality are affected by crop management, mainly by nitrogen fertilization, cultivar and its yield potential are known to be associated with protein present in the grain. Evaluations carried from season 2006–07 to 2008–09 have shown that CEVY Oro C2008 is consistently superior

to cultivar check Júpare C2001 in content of yellow pigment. CEVY Oro C2008 produces a grain with an average specific weight of 83 kg/hl and 13.5% protein at 12% moisture content. The intensity of yellow pigment in the endosperm of CEVY Oro C2008 grain has an average of 28.1 points in the Minolta b scale and is the reason for the designation 'Oro' (golden), which confers a high value in the international market.

Yield and quality, semi-commercial validation. Wheat, as with other crops, fluctuats in yield between years and between locations depending, among other factors, on water availability and nutrients. In validation plots with two cooperating farmers in the Yaqui Valley in blocks 609 and 2518, during the fall–winter 2008–09 crop season, the highest yield obtained from CEVY Oro C2008 was 7.5 t/ha, with an average of 7.1 t/ha. During the 2009–10 crop season, in blocks 1718 and 2520 and in the Mayo Valley experimental site, the highest yield potential obtained was 8.5 t/ha with an average of 7.76 t/ha. During 2010–11, in blocks 809, 1718, and 2518, and in Bacorehuis, Sinaloa, the highest yield obtained was 8.7 t/ha with an average of 7.06 t/ha.

From the farmer`s point of view, grain yield has been the main parameter for choosing a cultivar; however, in the case of durum wheat, pigment is a very important factor to consider for export. In validation plots with two cooperating farmers in the Yaqui Valley, in block 1718 and 2520, during the fall-winter 2009–10 crop season, the average of the b Minolta value was 29.4, while in 2010–11 it was 28.75 in blocks 1718 and 2518.

Table 1. Area (ha) grown with wheat during the 2009–10 and 2012–13 agricultural seasons in southern Sonora, Mexico (% indicates the percent of the total area growing wheat).

2009–10			2012–13		
Cultivar	**Area (ha)**	**%**	**Cultivar**	**Area (ha)**	**%**
Durum wheat					
Átil C2000	81,777	33.07	CIRNO C2008	196,295	77.90
Júpare C2001	53,164	21.50	Movas C2009	8,576	3.40
Samayoa C2004	23,318	9.43	Huatabampo Oro C2009	6,222	2.47
Sáwali Oro C2008	4,761	1.93	RSM Imperial C2008	4,538	1.80
CIRNO C2008	3,256	1.32	Átil C2000	4,080	1.62
CEVY Oro C2008	3,233	1.31	Patronato Oro C2008	3,183	1.26
Platinum	2,655	1.07	Sáwali Oro C2008	1,181	0.47
Patronato Oro C2008	2,325	0.94	RSM Chapultepec C2008	848	0.34
Aconchi C89	1,019	0.41	CEVY Oro C2008	185	0.07
RSM Imperial C2008	980	0.40			
Banámichi C2004	826	0.33			
RSM Chapultepec C2008	499	0.20			
Rafi C97	351	0.14			
Río Colorado	296	0.12			
Nácori C97	241	0.10			
Altar C84	105	0.04			
TOTAL	178,806		TOTAL	225,108	
Bread wheat					
Tacupeto F2001	40,552	16.40	Tacupeto F2001	8,824	3.50
Kronstad F2004	25,021	10.12	Roelfs F2007	5,223	2.07
Abelino F2004	736	0.30	Kronstad F2004	4,166	1.65
RSM-Norman F2008	659	0.27	RSM-Norman F2008	2,312	0.92
Rayón F89	636	0.26	Navojoa M2007	2,301	0.91
Tarachi F2000	384	0.16	Villa Juárez F2009	1,942	0.77
Roelfs F2007	248	0.10	Onavas F2009	1,724	0.68
Navojoa M2007	235	0.10	Abelino F2004	160	0.06
Monarca F2007	4	0.00	Ocoroni F86	149	0.06
			Tepahui F2009	92	0.04
			Japaraqui F2009	20	0.01
TOTAL	68,475		TOTAL	26,913	

Commercial cultivation of CEVY Oro C2008. After the release for commercial cultivation, durum wheat cultivar CEVY Oro C2008 occuppied 3,233 ha in southern Sonora (Table 1, p. 40) during the crop season 2009-10 (Fuentes-Dávila et al. 2011) (Fig. 1, p. 39) and 6,161 ha during 2010–11 (Valenzuela-Herrera et al. 2012). The most popular cultivars during 2008–09, Júpare C2001 and Átil C2000, were sown in 119,327 and 53,106 ha, respectively. The area planted to Júpare C2001 was reduced 53,164 ha in 2009–10 and Átil C2000 increased to 81,777 ha. The susceptibility of Júpare C2001 to leaf rust since 2008 and its reduction in preference by farmers, was more obvious during the 2010–11 season when only 6,638 ha were sown. Átil C2000, which has shown higher grain yield than Júpare, also had a reduced area to 44,826 ha. During the 2011–12 crop season, Átil was sown in 11,343 ha and Júpare in 913 ha, while in 2012–13, Átil was sown in 4,080 ha and Júpare was not grown in southern Sonora (Table 1, p. 40). The availability of the new durum wheat cultivars under the designation 'Oro' (golden), due to the high pigment content, and generated by the collaborative breeding project between INIFAP and CIMMYT, in response to the requests by the farmers of the region, seemed to have a rise in area sown with this type of cultivar in 2010–11; CEVY Oro C2008 occupied 6,161 ha, however, in 2011-12 it was sown in 931 ha and in 2012–13 in only 185 ha. On the other hand, durum wheat cultivar CIRNO C2008, publized as the improved Átil due to leaf rust resistance, has occupied 3,256 ha during 2009–10, 87,105 in 2010–11, 154,915 ha in 2011–12, and 196,295 in the 2012–13 crop seasons since its release. Despite research efforts in durum wheat to diversify the sources of resistance to leaf rust and to improve quality for export, efforts that have generated cultivars with both attributes since 2008, e.g., CEVY Oro C2008, Patronato Oro C2008, Sáwali Oro C2008, and, more recently, Huatabampo Oro C2009, the production and commercialization of durum wheat in this region of Mexico, continues to be based on grain yield. Due to the risks that monocultivar culture represents (Figueroa-López et al. 2011b), and based on scientific knowledge and the experiences with the durum wheat cultivars Altar C84 (Figueroa-López et al. 2002) and Júpare C2001 (Figueroa-López 2009) in northwest Mexico, we expect that the commercial longevity of durum wheat cultivar CIRNO C2008 will be more limited than expected. The appearance of new races of leaf rust in the next few crop seasons have a high possiblity to overcome its resistance, with consequent negative effects on economics and the environment, for the application of fungicides that will be neeed in the region and the lack of sufficient seed of other cultivars with resistance that could replace CIRNO C2008.

Conclusions. Durum wheat cultivar CEVY Oro C2008 released in 2008 for commercial cultivation, was sown in an area of 3,233 ha in southern Sonora during the crop season 2009–10, 6,161 ha during 2010–11, 931 ha during 2011–12, and only 185 ha during 2012–13. New or different schemes for durum wheat commercialization are needed, so that quality can be better remunerated and, therefore, genetic diversity for resistance to leaf rust is maintained in the region, which in turn will increase the commercial longevity of the cultivars generated.

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Characteristics and description of phenotypic components of Ónavas F2009 a new bread wheat cultivar for southern Sonora, Mexico.

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Abstract. Commercial cultivar Ónavas F2009 was developed at the Norman E. Borlaug Experimental Station through a collaborative project between INIFAP and CIMMYT, for the wheat-producing areas of the states of Sinaloa, Sonora, South Baja California, and Baja California in Mexico. The pedigree and selection history is 'KAMBARA1*2/BRAMBLING' (selection history: CGSS01B00069T-099Y-099M-099M-099Y-099M-20Y-0B). Ónavas F2009 has the registration TRI-121-100910 in the Catalog of Cultivars Feasible for Registration. This cultivar is spring type and resistant to leaf rust, with an experimental average grain yield of 6.012 t/ha. Ónavas F2009 has an average of 81 days to heading and 121 to physiological maturity, average height of 97 cm, plant growth habit is intermediate, and shows a high frequency of recurved flag leaves. Ear shape, in profile view, is parallel sided, density is lax, and the length excluding awns is very long. Ear glaucosity is medium and, at maturity, becomes white. The shoulder width of the lower glume is absent or very narrow, shoulder shape is sloping, the beak length is short and slightly curved. Grain shape is semi-elongated, and grain coloration, when treated with phenol, is light.

Introduction. Bread wheat production in Mexico was 3.9 x 10^6 tons in 2010, which was not enough to fulfill the national demand, so 3.3 x 10^6 tons had to be imported to suffice consumption needs (OEIDRUS 2011). In order to assure minimum strategic reserves with the objective of reducing the risk of depending on fluctuating international market prices, the regional industry reacted by implementing 'agriculture by contract' with wheat producers several years ago (Melis-Cota 2008).

Despite the problems caused by Karnal bunt at the beginning of the 1980s (SARH 1987), 220,409 ha were sown with bread wheat in 1990–91, which represented 89% of the area grown with wheat in the state of Sonora. However, since the 1994–95 season, durum wheat has reached more area than bread wheat in the state. The area grown with wheat in 2010–11 in Sonora was 292,247 ha (Table 2); 87% (254,531 ha) corresponded to the southern region (OEIDRUS 2011). The predominate durum wheat cultivars were CIRNO C2008 and Átil C2000 with more than 40% of the total area. Bread wheat cultivars occupied 30% of the area.

Leaf rust and Karnal bunt are the most important diseases in southern Sonora (Figueroa-López et al. 2011). However, since year 2000, incidence of stripe or yellow rust has increased in the region with the appearance of new races of the fungus. The cultivar Tacupeto F2001 was the most grown bread wheat cultivar in 2009–10 and 2010–11 in southern Sonora (Table 2). Despite its quality attributes for the milling industry, Tacupeto F2001 has shown susceptibility to stripe rust (up to 80% severity) and moderate susceptibility to leaf rust (up to 40% severity), levels that make chemical application necessary, increasing production costs. Therefore, the collaborative project between the Mexican National Institute For Forestry, Agriculture, and Livestock Research (INIFAP) and the International Maize and Wheat Improvement Center (CIMMYT) with support by the farmer´s union (PIEAES) of the Yaqui Valley, has the objective of improving the cost/benefit and competiveness of bread wheat by producing advanced lines with better quality, yield, resistance to disease, and better water use efficiency, that could be released as commercial cultivars (Figueroa-López et al. 2011).

Table 2. Area (ha) grown with wheat during the 2010–11 and 2009–10 agricultural seasons in southern Sonora, Mexico (% indicates the percent of the total area growing wheat).

2010–11			2009–10		
Cultivar	**Area (ha)**	**%**	**Cultivar**	**Area (ha)**	**%**
Durum wheat					
CIRNO C2008	88,235	30.5	Átil C2000	81,777	33.07
Átil C2000	50,236	17.3	Júpare C2001	53,164	21.50
Sáwali Oro C2008	14,353	4.9	Samayoa C2004	23,318	9.43
Patronato Oro C2008	11,753	4.0	Sáwali Oro C2008	4,761	1.93
Júpare C2001	10,069	3.4	CIRNO C2008	3,256	1.32
RSM Imperial C2008	7,149	2.4	CEVY Oro C2008	3,233	1.31
CEVY Oro C2008	6,197	2.1	Platinum	2,655	1.07
Río Colorado	5,111	1.7	Patronato Oro C2008	2,325	0.94
Samayoa C2004	4,905	1.6	Aconchi C89	1,019	0.41
Rafi C97	1,806	0.6	RSM Imperial C2008	980	0.40
RSM Chapultepec C2008	1,650	0.5	Banámichi C2004	826	0.33
Others	1,210	0.4	RSM Chapultepec C2008	499	0.20
Platinum	1,173	0.4	Rafi C97	351	0.14
Aconchi C89	752	0.2	Río Colorado	296	0.12
			Nácori C97	241	0.10
			Altar C84	105	0.04
TOTAL	204,599	70	TOTAL	178,806	
Bread wheat					
Tacupeto F2001	36,819	12.6	Tacupeto F2001	40,552	16.40
Kronstad F2004	18,681	6.4	Kronstad F2004	25,021	10.12
Roelfs F2007	10,358	3.6	Abelino F2004	736	0.30
Navojoa M2007	8,046	2.8	RSM-Norman F2008	659	0.27
RSM Norman F2008	4,499	1.5	Rayón F89	636	0.26
Cachanilla F2000	3,493	1.2	Tarachi F2000	384	0.16
Rayón F89	2,576	0.9	Roelfs F2007	248	0.10
Abelino F2004	1,355	0.5	Navojoa M2007	235	0.10
Palmerín F2004	964	0.3	Monarca F2007	4	0.00
Others	538	0.1			
Oasis F86	319	0.1			
TOTAL	87,648	30	TOTAL	68,475	

Pedigree, history selection and description of Ónavas F2009. After evaluating grain yield since the 2007–08 agricultural season at the Norman E. Borlaug Experimental Station (CENEB), we proposed to release the experimental bread wheat line 'KAMBARA1*2/BRAMBLING' as the cultivar Ónavas F2009 (Figueroa-López et al. 2012). Ónavas F2009 is a spring type bread wheat cultivar, which originated from hybridizations made in the Bread Wheat Breeding Program of CIMMYT. The cross number and history selection is CGSS01B00069T-099Y-099M-099M-099Y-099M-20Y-0B. Shuttle breeding was carried out between the experimental stations of El Batán, state of Mexico (B) (19°30'N and 2,249 msnm); San Antonio Atizapán, state of Mexico (M) (19°17'N and 2,640 msnm); and the Yaqui Valley (Y) (27°20'N and 40 msnm) in Sonora (Table 3).

Table 3. Selection history and localities where cultivar Onavas F2009 was evaluated (For season: F–W = fall–winter and S–S = spring–summer; for irrigation type: RR = regular rainfed and NI = normal irrigation; and planting dates were 15 and 30 November, 15 December, and 1 January).

Activity	Locality	Season	Irrigation
Triple genetic cross	El Batan, Mexico	S–S/2001	RR
F_1 generation	Cd. Obregon, Sonora	F–W/2002–03	NI
F_2 generation	Atizapan, Mexico	S–S/2003	RR
F_3 generation	Atizapan, Mexico	S–S/2004	RR
F_4 generation	Cd. Obregon, Sonora	F–W/2004–05	NI
F_5 generation	Atizapan, Mexico	S–S/2005	RR
F_6 generation	Cd. Obregon, Sonora	F–W/2005–06	NI
F_7 generation	El Batan, Mexico	S–S/2006	RR
Yield trials by CIMMYT		F–W/2007–08	NI
Yield trials by INIFAP AT different planting dates	Cd. Obregon, Sonora	F–W/2008–09	NI

The most important phenotypic characteristics of Ónavas F2009, according to the International Union for the Protection of New Varieties of Plants (UPOV, 1994), are given (Table 4). Cultivar Ónavas F2009 has an average of 81 days to heading with a range of 72 to 88. With a biological cycle with an average of 121 days for physiological maturity, the cycle may be shortened due to the lack of cold hours if planting is late (Wardlaw and Moncur 1995), and may average 107 days when sowing is done at the end of December. Ónavas F2009 has an average height of 97 cm (Fig. 2, p. 45), a maximum of 114 and minimum of 85. Plant growth habit is intermediate and shows a high frequency of recurved flag leaves.

Table 4. Characteristics and description of the phenotypic components of cultivar Onavas F2009.

Structure	Characteristic	Description
Coleoptile	Anthocyanin coloration	Absent or very weak
First leaf	Anthocyanin coloration	Absent or very weak
Plant	Growth habit	Intermediate
	Frequency of recurved flag leaves	Very high
	Length (stem, spike, and awns)	Long
	Seasonal type	Spring
Spike	Time of emergence	Medium
	Glaucosity	Medium
	Length (excluding awns)	Long
	Hairiness of margin of first rachis segment	Weak
	Color (at maturity)	White
	Shape in profile view	Parallel sided
	Density	Lax
Flag leaf	Glaucosity of blade	Strong
Straw	Pith in cross section (halfway between base of ear and stem node below)	Thin
Culm	Glaucosity of neck	Medium
Lower glume	Shape of shoulder	Sloping
	Shoulder width	Absent or very narrow
	Length of beak	Short
	Shape of beak	Slightly curved
	Hairiness on external surface	Weak
Awns	Presence	Present
Awns at the tip of ear	Length	Short
Grain	Coloration with phenol	Light

Spike shape in profile view is parallel sided, density is lax, and the length excluding awns is very long. Ear glaucosity is medium, becoming white at maturity. The shoulder width of the lower glume is absent or very narrow, (spikelet in mid-third of spike), shoulder shape is sloping, beak length is short, and slightly curved. Grain shape is semi-elongated (Fig. 3), and grain coloration when treated with phenol is light.

Fig 2. Bread wheat cultivar Ónavas F2009 has an average height of 97 cm. Plants present an intermediate growth habit and a very high frequency of recurved flag leaves.

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Fig. 3. Grain shape of the bread wheat cultivar Ónavas F2009 is semi-elongated. Grain color after treatement with phenol is light.

Effect of the level of wheat seed infection with Karnal bunt on germination and tiller production.

Guillermo Fuentes-Dávila, Pedro Figueroa-López, Juan Manuel Cortés-Jiménez, Pedro Félix-Valencia, Miguel Alfonso Camacho-Casas, Gabriela Chávez-Villalba, José Luis Félix-Fuentes, and Alma Angélica Ortiz-Ávalos.

Abstract. The germination and tiller production of random selected seed showing different levels of infection with Karnal bunt were evaluated during the 2008–09 crop season at the Norman E. Borlaug Experimental Station in the Yaqui Valley, Sonora, Mexico. Sowing was on 8 and 17 December, 2008. Seed germination was 100%, 100%, and 95% for seed with 1, 2, and 3 levels of infection, respectively, sown on 8 December, while for sowing date 17 December, infection levels were 85%, 90%, and 85%. The range of the number of tillers produced by seed with infection levels 1, 2, and 3, sown on 8 and 17 December in the left and right rows of the bed was 7-37, 10-28, 7-31, 8-33, 5-33, and 7-42, and 16-39, 16-46, 8-33, 8-38, 19-43, 28-42, respectively. The average number of tillers produced was 17.7, 17.8, and 20.9 for seed with levels of infection 1, 2, and 3, respectively, sown on 8 December and 25.5, 20.8, and 27.2 for the 17 December sowing date. The average number of tillers produced was higher in seed with the highest level of infection for both sowing dates, and higher in seed sown on 17 December than on 8 December.

Introduction. Karnal bunt of wheat caused by Tilletia indica occurs on bread wheat (Mitra 1931), durum wheat, and triticale (Agarwal et al. 1977). The fungus affects partially the grains in a plant (Bedi et al. 1949). In general, infected grains are partially affected and in some occasions totally destroyed. Although the fungus may penetrate the embryo, it does not necessarily cause damage (Chona et al. 1961; Mitra 1935). Partially infected grains may give rise to healthy plants, although the percentage of germination decreases depending the level of infection (Bansal et al. 1984; Rai and Singh 1978; Singh 1980). Severely affected seed loses viability or shows abnormal germination (Rai and Singh 1978). Our objective was to determine the percentage of germination and tiller production under field conditions from wheat seed with different levels of infection caused by Karnal bunt.

Materials and methods. Germination and tiller production of random selected seed showing different levels of infection with Karnal bunt were evaluated during the 2008–09 crop season at the Norman E. Borlaug Experimental Station in the Yaqui Valley, Sonora, Mexico. The seed was obtained from the various nurseries evaluated for their reaction to Karnal bunt inoculation during the 2007–08 crop season. Sowing was on 8 and 17 December, 2008, on beds with two rows using 10 seeds in a 1-m row. For the first sowing date, 20 seeds with a 1 level of infection (point of infection at the base of the seed) were used, 20 with a 2 level of infection (~30% of the seed affected with karnal bunt), and 20 with a 3 level of infection (~50% of the seed affected) (Fig. 4), for a total of six rows. The same number of seeds and similar levels of infection were used for the second sowing date. Counts on seed germination and number of tillers produced by each seed were made several times during the season. The agronomic management followed the technical recommendations by INIFAP (Figueroa-López et al. 2011).

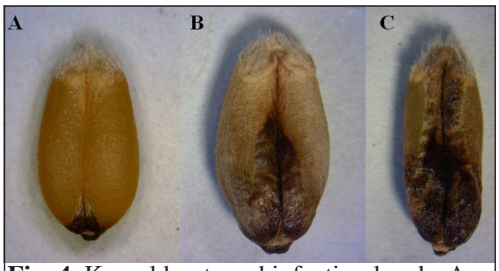

Fig. 4. Karnal bunt seed infection levels. A, level 1 infection (point of infection at base of seed); B, infection level 2 (~20 of the seed infected); and C, infection level 3 (~50% of the seed infected).

Results and discussion. Seed germination was 100%, 100%, and 95% for seed with 1, 2, and 3 levels of infection, respectively, sown on 8 December (Fig. 5A). For the 17 December sowing date, 85%, 90%, and 85% of the seed was infected at the 1, 2, and 3 levels, respectively (Fig. 5B). Rai and Singh (1978) reported a decrease in the percent seed germination with similar levels of infection as those used in our study, from 84% to 57% and to 49% in the cultivar Arjun from the harvest of 1975, and from 84% to 72% and to 58% from the harvest in 1976. Singh (1980) also reported a decrease from 74% to 72% and to 59% with cultivar CC-464, and Bansal et al. (1984) reported a decrease from 84% to 83% and to 79% with cultivar WL711. The percent seed germination observed in our study was much higher than those reported by these researchers, especially the germination of seed with an infection level 3; the average germination of seed sown on 8 December was 98.33%, whereas germination of seed sown on 17 December was 86.66%. The factor that could account for the difference in the results obtained in our study from the previously mentioned studies is that we did not use seed from the same cultivar with different levels of infection, but rather randomly selected seed from different nurseries and previous inoculation tests.

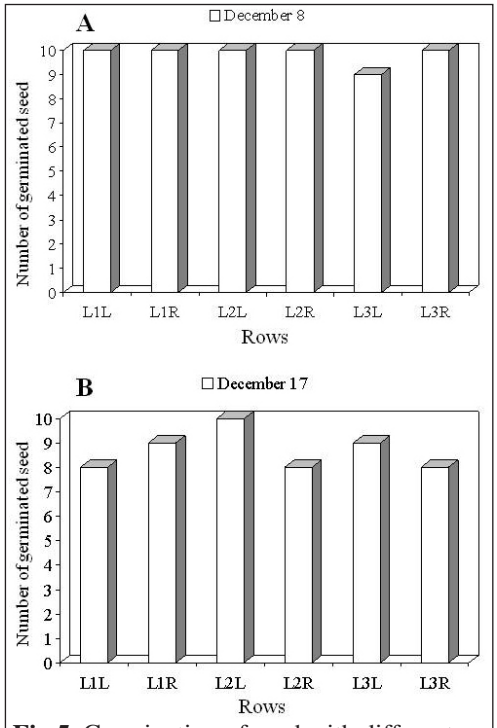

Fig 5. Germination of seed with different levels of Karnal bunt infection sown on 8 (A) and 17 (B) December, 2008 (for rows, L1L indicates an infection level of 1, left row in bed, L1R indicates a level 1 infection in the right row of the bed, etc.).

The range of the number of tillers produced by seed with infection levels 1, 2, and 3, sown on December 8 and 17 in the left and right rows of the bed was as follows: 7-37, 10-28, 7-31, 8-33, 5-33, and 7-42 (Fig. 6, p. 47), and 16-39, 16-46, 8-33, 8-38, 19-43, and 28-42 (Fig. 7, p. 47), respectively. The average number of tillers produced was 17.7, 17.8, and 20.9 for seed with levels of infection 1, 2, and 3, respectively, sown on 8 December and 25.5, 20.8, and 27.2 for the 17 December sowing date. Rai and Singh (1978) reported that infected seeds that retained their viability produced a higher proportion of abnormal seedlings compared to the healthy control, and viable seed producing abnormal seedlings in 1975 was 13.09% (level 1), 17.54% (level 2), and 22.44% (level 3) and

in 1976 was 7.14% (level 1), 20.83% (level 2), and 24.13% (level 3). Singh (1980) reported that the loss in seedling length over the healthy control was 6.74% (level 1), 12.06% (level 2), and 26.21% (level 3). Bansal et al. (1984) reported that seed with trace infection or moderately low infection produced normal seedlings, but severely affected seed showed very poor vigor and the majority of the seed belonging to grade 5 (seed with practically the whole endosperm destroyed), did not germinate, and in the rest of the grades, seedlings were pale and weak. The results of our study did not correlate with those of Rai and Sing, Singh, and Bansal et al., but rather a random response was observed. However, we did not evaluate seedling development, and the seed used was not obtained from a single cultivar, but rather from randomly selected seed from various nurseries. Another piece of interesting data from our study is that the average number of tillers produced was higher in seed with the highest level of infection for both sowing dates, and higher in seed sown on 17 December than on 8 December.

Conclusions. Seeds, with points of Karnal bunt infection at the base of the seed and ~30% and ~50% of the seed affected, were able to germinate and produce tillers. The range of germination was 85–100%, and the range of the number of tillers produced was 5–46.

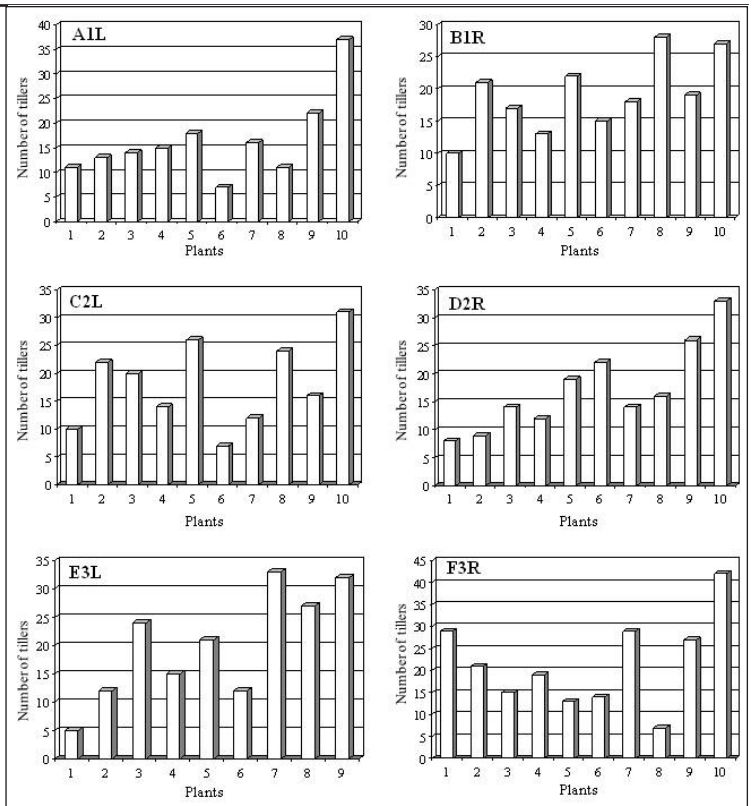

Fig. 6. Number of tillers produced in wheat plants originating from seed with Karnal bunt infection levels 1, 2, and 3 on the left (L) and right (R) of 1-m rows sown on 8 December, 2008.

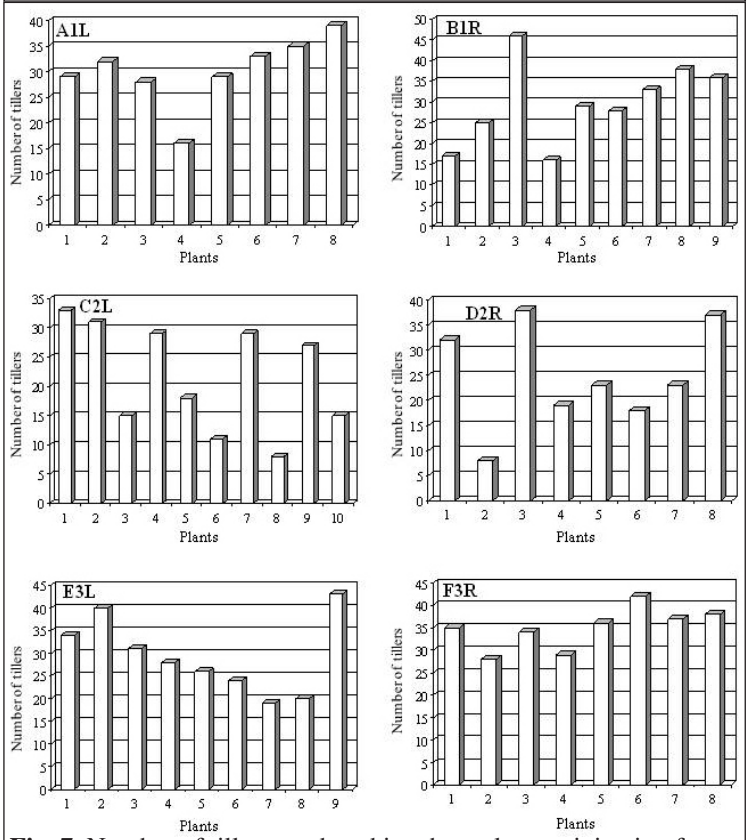

Fig. 7. Number of tillers produced in wheat plants originating from seed with Karnal bunt infection levels 1, 2, and 3 on the left (L) and right (R) of 1-m rows sown on 17 December, 2008.

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Reaction of selected cultivars and lines of durum and bread wheat to black point.

Guillermo Fuentes-Dávila, Pedro Figueroa-López, Juan Manuel Cortés-Jiménez, Pedro Félix-Valencia, Miguel Alfonso Camacho-Casas, José Luis Félix-Fuentes, Gabriela Chávez-Villalba, and Alma Angélica Ortiz-Ávalos.

Abstract. Twenty advanced lines of durum wheat and commercial cultivars Samayoa C84, CEVY Oro C2008, CIRNO C2008, Patronato Oro C2008, Sáwali Oro C2008, Imperial C2008, and Chapultepec C2008, 21 advanced lines of bread wheat, and the commercial cultivars Tacupeto F2001, Kronstad F2004, Navojoa M2007, Roelfs F2007, and Norman F2008, were evaluated for their reaction to black point under natural conditions during the 2009–10 crop season at three sowing dates. The range of infected grain for the first sowing date for durum wheat lines and cultivars was 0 to 50.66%, 0 to 17.75% for the second date, and 0 to 5.51% for the third. Only the line 'SOMAT_4/INTER_8//VERDI/10/PLATA_10/6/MQUE/4/USDA573//QFN/AA_7/3/ ALBA-D/5/AVO/HUI/7/PLATA_13/8/RAFI97/9/ MALMUK_1/ SERRATOR_1' did not present infected grain in any of the three dates. The range of infected grain for the first sowing date for bread wheat lines and cultivars was 0–30.87%, 0–23.06% for the second date, and 0–10.80% for the third. Only the line 'PFAU/MILAN//TROST/3/ PBW65/2*SERI.1B' did not present infected grain at any of the three dates.

Introduction. More than 100 species of fungi, including *Alternaria*, *Fusarium*, and *Helminthosporium* spp., can be isolated from newly harvested wheat grain. These fungi are most important in humid field environments, where they infect seed when relative humidity exceeds 90% and seed moisture content exceeds 20%. Rainfall during seed maturation and humid weather prevailing for a few days prior to harvest (Prescott et al. 1986; Watkins 2013) favor black point (BP). Expanding green kernels are most susceptible. Premature seed senescence also promotes BP, because many of the fungi are saprophytic (Wiese 1987). *Alternaria alternata* and *Bipolaris sorokiniana* are generally considered the primary causal agents of the disease (Mathur and Cunfer 1993). Infected ears may look normal, but there may be elliptical, brown to dark brown lesions on the inner side of the glumes. The disease is more pronounced as brown to dark brown or blackish, localized discolored areas, usually around the embryo end of seeds (Adlakha and Joshi 1974; Hanson and Christensen 1953; Rana and Gupta 1982; cited by Mathur and Cunfer 1993). The discoloration may also occur near the brush, in the crease, or any part of the seed and may be light or dark or with a distinct margin (Fig. 8). Severe infection causes discoloration and shrivelling of the whole seed (Adlakha and Joshi 1974). In southern Sonora, Mexico, black point is an endemic disease of durum and bread wheat, although incidence is variable from year to year. Wheat breeding programs select for disease resistance during seed evaluation after harvest, however, there is not a formal project on BP in Sonora. Our objective was to evaluate the reaction of selected durum and bread wheat advanced lines and commercial cultivars for their reaction to black point under natural conditions during the 2009–10 crop season.

Fig. 8. Symptoms of black point in wheat grain.

Materials and methods. Twenty advanced lines of durum wheat; the commercial durum wheat cultivars Samayoa C84, CEVY Oro C2008, CIRNO C2008, Patronato Oro C2008, Sáwali Oro C2008, Imperial C2008, and Chapultepec C2008; 21 advanced lines of bread wheat; and the commercial bread wheat cultivars Tacupeto F2001, Kronstad F2004, Navojoa M2007, Roelfs F2007, and Norman F2008 (Table 5, pp. 49-50), were evaluated for their reaction to black point under natural conditions, during the 2009–10 crop season at the Norman E. Borlaug Experimental Station in block 910 in a clay soil with pH 7.8, in the Yaqui Valley, Mexico. Sowing dates were 19, 25, and 30 November, 2009, using 8 g of seed for a 0.7-m long bed with two rows. Harvest was done manually, and the counting of healthy and infected seed was done by visual inspection. The agronomic management followed the technical recommendations by INIFAP (Figueroa-López et al. 2011).

Results. Durum wheat. The range of infected grain for the first sowing date for durum wheat lines and cultivars was 0–50.66%, with an average of 7.38; 0–17.75%, with an average of 0.99 for the second date; and 0–5.51%, with an

Table 5. Advanced lines and commercial durum and bread wheat cultivars evaluated for their reaction to black point in the field in three sowing dates, during the crop season 2009–10, in the Yaqui Valley, Sonora, Mexico.

	Pedigree and selection history
Durum wheats	
1.	Samayoa C2004
2.	CEVY Oro C2008
3.	CIRNO C2008
4.	Patronato Oro C2008
5.	Sawali Oro C2008
6.	1A.1D5+10-6/3*MOJO//RCOL/4/ARMENT//SRN_3/NIGRIS_4/3/CANELO_9.1 CDSS02Y00408S-0Y-0M-4Y-0Y
7.	GUAYACANINIA/POMA_2//SNITAN/4/D86135/ACO89//PORRON_4/3/SNITAN CDSS02B00562S-0Y-0M-2Y-1M-04Y-0B
8.	CMH83.2578/4/D88059//WARD/YAV79/3/ACO89/5/2*SOOTY_9/RASCON_37/6/1A.1D5+10-6/3*MOJO/3/AJAIA_12/F3LOCAL(SEL.ETHIO. 135.85)//PLATA_13 CDSS02B00720S-0Y-0M-8Y-1M-04Y-0B
9.	SOMAT_4/INTER_8//VERDI/10/PLATA_10/6/MQUE/4/USDA573//QFN/AA_7/3/ALBA-D/5/AVO/HUI/7/PLATA_13/8/RAFI97/9/MALMUK_1/SERRATOR_1 CDSS04Y01242T-0TOPB-4Y-0M-06Y-3M-1Y-0B
10.	TGBB/CANDEF//LALA/GUIL/3/BONVAL/4/TILO_1/LOTUS_4/5/TILO_1/LOTUS_4 CDSS02B01344T-0TOPB-0Y-0M-2Y-2M-04Y-0B
11.	MOHAWK/10/PLATA_10/6/MQUE/4/USDA573//QFN/AA_7/3/ALBA-D/5/AVO/HUI/7/PLATA_13/8/THKNEE_11/9/CHEN/ALTAR84/3/HUI/ POC//BUB/RUFO/4/FNFOOT CDSS04SH00022S-22Y-2M-5Y-1M-1Y-0B
12.	SILK_3/DIPPER_6/3/ACO89/DUKEM_4//5*ACO89/4/PLATA_7/ILBOR_1//SOMAT_3 CDSS02B00290S-0Y-0M-8Y-4M-04Y-0B
13.	CBC 509 CHILE/6/ECO/CMH76A.722//BIT/3/ALTAR 84/4/AJAIA_2/5/KJOVE_1/7/AJAIA_12/F3LOCAL(SEL.ETHIO.135.85)//PLATA_13/8/SOOTY_9/RASCON_37//WODUCK/CHAM_3 CDSS02B00596S-0Y-0M-5Y-1M-04Y-0B
14.	ARMENT//2*SOOTY_9/RASCON_37/4/CNDO/PRIMADUR//HAI-OU_17/3/SNITAN CDSS02B00643S-0Y-0M-1Y-4M-04Y-0B
15.	GUAYACAN INIA/GUANAY/10/LD357E/2*TC60//JO69/3/FGO/4/GTA/5/SRN_1/6/TOTUS/7/ENTE/MEXI_2//HUI/4/YAV_1/3/LD357E/2*TC60//JO69/8/SOMBRA_20/9/JUPARE C 2001 CDSS04Y00275S-8Y-0M-06Y-4M-1Y-0B
16.	SOMAT_4/INTER_8/4/GODRIN/GUTROS//DUKEM/3/THKNEE_11/5/CNDO/PRIMADUR//HAI-OU_17/3/SNITAN CDSS04Y00746T-0TOPB-4Y-0M-06Y-1M-1Y-0B
17.	PLATA_10/6/MQUE/4/USDA573//QFN/AA_7/3/ALBA-D/5/AVO/HUI/7/PLATA_13/8/RAFI97/9/MALMUK_1/SERRATOR_1/10/ARMENT//SRN_3/NIGRIS_4/3/CANELO_9.1/11/SHAG_21/DIPPER_2//PATA_2/6/ARAM_7//CREX/ALLA/5/ENTE/MEXI_2//HUI/4/YAV_1/3/LD357E/2*TC60//JO69 CDSS04Y00993T-0TOPB-16Y-0M-06Y-1M-1Y-0B
18	ALTAR 84/BINTEPE 85/3/STOT//ALTAR 84/ALD/4/POD_11/YAZI_1/5/VANRRIKSE_12/SNITAN/6/SOOTY_9/RASCON_37//WODUCK/CHAM_3 CDSS04Y01051T-0TOPB-21Y-0M-06Y-2M-1Y-0B
19.	CANELO_9.1/SNITAN/10/PLATA_10/6/MQUE/4/USDA573//QFN/AA_7/3/ALBA-D/5/AVO/HUI/7/PLATA_13/8/THKNEE_11/9/CHEN/ALTAR 84/3/HUI/POC//BUB/RUFO/4/FNFOOT CDSS02B00348S-0Y-0M-4Y-1M-04Y-0B
20.	YAV79/4/ARMENT//SRN_3/NIGRIS_4/3/CANELO_9.1/5/MINIMUS/COMB DUCK_2//CHAM_3/3/GREEN_19 CDSS02B01063T-0TOPB-0Y-0M-1Y-4M-04Y-0B
21.	DUKEM_1//SORA/2*PLATA_12/3/SOMAT_4/INTER_8 CDSS04Y00520S-18Y-0M-06Y-4M-1Y-0B
22.	RANCO//CIT71/CII/3/COMDK/4/TCHO//SHWA/MALD/3/CREX/5/SNITAN/6/YAZI_1/AKAKI_4//SOMAT_3/3/AUK/GUIL//GREEN CDSS04B00151S-3Y-0M-2Y-0M-2Y-0B
23.	GODRIN/GUTROS//DUKEM/3/THKNEE_11/4/DUKEM_1//PATKA_7/YAZI_1/3/PATKA_7/YAZI_1/5/AJAIA_12/F3LOCAL(SEL.ETHIO.135.85)//PLATA_13/3/ADAMAR CDSS04B00367T-0TOPY-10Y-0M-4Y-0M-4Y-0B
24.	MOHAWK/6/LOTUS_5/F3LOCAL(SEL.ETHIO.135.85)/5/CHEN/ALTAR 84/3/HUI/POC//BUB/RUFO/4/FNFOOT CDSS05Y00087S-64Y-0M-1Y-0M-1Y-0B
25.	AJAIA_3/SILVER_16//AJAIA_13/YAZI/4/ARMENT//SRN_3/NIGRIS_4/3/CANELO_9.1/5/GODRIN/GUTROS//DUKEM/3/THKNEE_11 CDSS05Y00512T-0TOPB-21Y-0M-4Y-0M-1Y-0B
26.	Imperial C2008
27.	Chapultepec C2008

Table 5. Advanced lines and commercial durum and bread wheat cultivars evaluated for their reaction to black point in the field in three sowing dates, during the crop season 2009–10, in the Yaqui Valley, Sonora, Mexico.

	Pedigree and selection history
Bread wheats	
1.	Tacupeto F2001
2.	Kronstad F2004
3.	Navojoa M2007
4.	Roelfs F2007
5.	TOBA97/PASTOR CMSS97M05756S-040M-020Y-030M-015Y-3M-1Y-3M-0Y
6.	KAMB1*2/BRAMBLING CGSS01B00069T-099Y-099M-099M-099Y-099M-20Y-0B
7.	BETTY/3/CHEN/AE.SQ//2*OPATA CMSW00WM00150S-040M-040Y-030M-030ZLM-3ZTY-0M
8.	WBLL1*2/BRAMBLING CGSS01B00062T-099Y-099M-099M-099Y-099M-12Y-0B
9.	BABAX/LR42//BABAX*2/4/SNI/TRAP#1/3/KAUZ*2/TRAP//KAUZ CGSS01B00045T-099Y-099M-099M-099Y-099M-26Y-0B
10.	BABAX/LR42//BABAX/3/ER2000 CMSA01Y00176S-040P0Y-040M-030ZTM-040SY-24M-0Y-0SY
11.	PFAU/MILAN/3/BABAX/LR42//BABAX CMSS02M00056S-030M-28Y-0M-040Y-25ZTB-0Y-01B-0Y
12.	THELIN/2*WBLL1 CGSS02Y00079T-099B-099B-099Y-099M-6Y-0B
13.	PBW343//CAR422/ANA/3/ELVIRA CMSS02M00409S-030M-1Y-0M-040Y-10ZTB-0Y-02B-0Y
14.	BABAX/LR42//BABAX/3/ER2000 CMSA01Y00176S-040P0Y-040M-030ZTM-040SY-30M-0Y-0SY
15.	TC870344/GUI//TEMPORALERA M 87/AGR/3/2*WBLL1 CMSA01Y00725T-040M-040P0Y-040M-030ZTM-040SY-10M-0Y-0SY
16.	ROLF07/YANAC//TACUPETO F2001/BRAMBLING CGSS05B00121T-099TOPY-099M-099NJ-4WGY-0B
17.	WAXWING*2/KRONSTAD F2004 CGSS04Y00020T-099M-099Y-099ZTM-099Y-099M-3WGY-0B
18.	WHEAR/KRONSTAD F2004 CGSS04Y00106S-099Y-099M-099Y-099M-9WGY-0B
19.	KEA/TAN/4/TSH/3/KAL/BB//TQFN/5/PAVON/6/SW89.3064/7/SOKOLL CMSS04Y00153S-099Y-099ZTM-099Y-099M-5WGY-0B
20.	CAL/NH//H567.71/3/SERI/4/CAL/NH//H567.71/5/2*KAUZ/6/WH576/7/WH 542/8/WAXWING CMSS04Y00364S-099Y-099ZTM-099Y-099M-2WGY-0B
21.	BECARD CGSS01B00063T-099Y-099M-099M-099Y-099M-33WGY-0B
22.	WHEAR/SOKOLL CMSS04Y00201S-099Y-099ZTM-099Y-099M-11WGY-0B
23.	PFAU/MILAN//TROST/3/PBW65/2*SERI.1B CMSS04M01426S-0TOPY-099ZTM-099Y-099M-3RGY-0B
24.	WHEAR/KRONSTAD F2004 CGSS04Y00106S-099Y-099M-099Y-099M-3WGY-0B
25.	CHEWINK CGSS03B00074T-099Y-099M-099Y-099M-6WGY-0B-3B
26.	Norman F2008

average of 0.48 for the third (Fig. 9A, p. 50). Only the line 'SOMAT_4/INTER_8//VERDI/10/PLATA_10 /6/MQUE/4/ USDA573//QFN/AA_7/3/ALBA-D/5/AVO/HUI/7/PLATA_13/8/RAFI97/9/ MALMUK_1/ SERRATOR_1' did not present infected grain at any of the three dates. Although all commercial cultivars in the overall average of infection, were in the 0.1–5.0% infection category (Fig. 10A, p. 51), at the first sowing date they had the highest potential for susceptibility, CIRNO C2008 had 13.93% infection, Samayoa C2004 12.13%, Patronato Oro C2008 6.03%, and Sáwali Oro C2008 5.85%. Lines with the highest levels of infection in the first sowing date were 'SOMAT_4/INTER_8/4/GODRIN/ GUTROS//DUKEM/3/THKNEE_11/5/CNDO/PRIMADUR//HAI-OU_17/3/SNITAN' with 50.66% and 'SILK_3/ DIPPER_6/3/ACO89/DUKEM_4//5*ACO89/4/ PLATA_7/ ILBOR_1//SOMAT_3' with 16.9%. SOMAT_4 showed a 17.75% infection at the second sowing date.

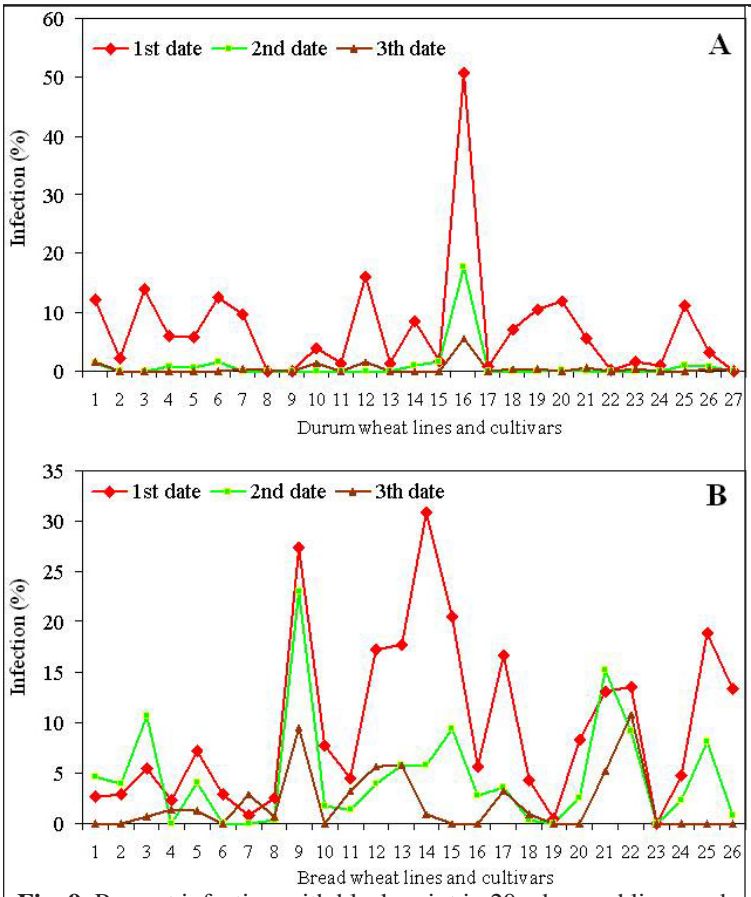

Fig. 9. Percent infection with black point in 20 advanced lines and seven commercial durum wheat cultivars (A) and in 21 advanced lines and five commercial bread wheat cultivars (B) in the Yaqui Valley, Sonora, Mexico, during the 2009–10 crop season.

infection with 13.93%. The overall average infection in the bread wheats was 5.43%, with a range of 0–30.87%. Among the bread wheat commercial cultivars, Norman F2008 had the highest infection at 13.29%.

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Bread wheat. The range of infected grain for the first sowing date for the bread wheat lines and cultivars was 0–30.87%, with an average of 9.69%; 0–23.06% for the second date, with an average of 4.60%; and 0–10.80% at the third date, with an average of 2.01% (Fig. 9B). Only the line 'PFAU/MILAN//TROST/3/PBW65/2*SERI.1B' did not present infected grain at any of the three dates. Commercial cultivars Tacupeto F2001, Kronstad F2004, Roelfs F2007, Norman F2008, and 10 advanced lines had 0.1–5.0% infection, and Navojoa M2007 and five lines had 5.1–10.0% infection (Fig. 9B). The highest infection were observed at the first sowing date in lines 'BABAX/LR42//BABAX/3/ER2000' with 30.87%, 'BABAX/LR42//BABAX*2/4/SNI/TRAP#1/3/KAUZ*2/TRAP//KAUZ' with 27.33%, and 'TC870344/GUI//TEMPORALERA M87/AGR/3/2*WBLL1' with 20.53%.

Conclusions. Black point is an endemic disease in southern Sonora, Mexico. The overall average infection in a selected group of advanced lines and commercial cultivars of durum wheat sown on three dates was 2.95%, with a range of 0–50.66%. Among the durum wheat commercial cultivars, CIRNO C2008 showed the highest

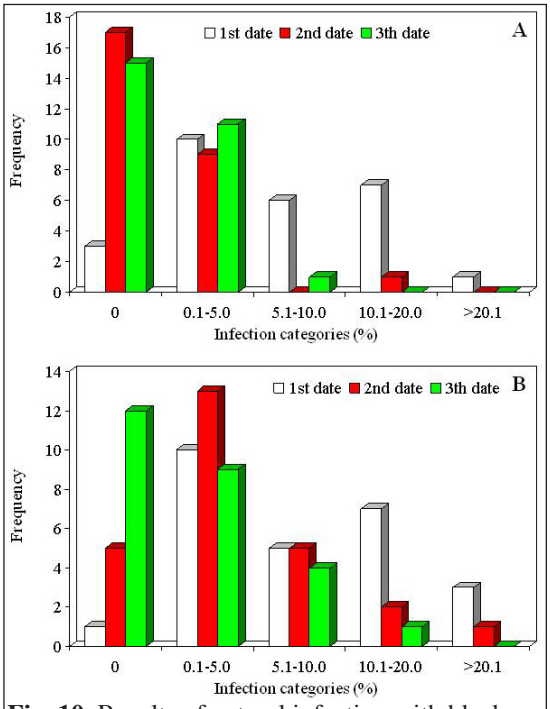

Fig. 10. Results of natural infection with black point in 20 advanced lines and seven commercial durum wheat cultivars (A) and in 21 advanced lines and five commercial bread wheat cultivars (B) in the Yaqui Valley, Sonora, Mexico, during the 2009–10 crop season.

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Reaction of bread wheat lines and cultivars to Karnal bunt under artificial inoculation during the 2011–12 crop season in the Yaqui Valley, Mexico.

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Abstract. Twenty-one advanced bread wheat lines and four commercial cultivars were evaluated for resistance to Karnal bunt by artificial inoculation during the 2011–12 crop season at two dates in the Yaqui Valley, Mexico. The infection range was 0.66-24.35% for the first date and 0.57–30.04% for the second date. Only lines 'FRET2/TUKURU//FRET2/3/MUNIA/CHTO//AMSEL/4/FRET2/TUKURU//FRET2' and 'KACHU/3/C80.1/3*BATAVIA//2*WBLL1/4/KACHU' were in the 2.6–5.0% infection category. Tepahui F2009 and seven lines had 5.1–10.0% infection and Roelfs F2007, Ónavas F2009, Villa Juárez F2009, and 12 lines had 10.1–30.0%. Commercial cultivars with the highest percent infection were Roelfs F2007 (10.19%) and Villa Juárez F2009 (18.53%). Lines that showed the highest infection were 'FRET2/TUKURU//FRET2/3/TACUPETOF2001*2/ KIRITATI' with 27.41% and 'ROLF07/4/BOW/NKT//CBRD/3/CBRD/5/FRET2/ TUKURU//FRET2' with 30.03%.

Introduction. Karnal bunt of wheat was first identified in India (Mitra 1931), and later in Mexico (Duran 1972), Pakistan (Munjal 1975), Nepal (Singh et al. 1989), Brazil, (Da Luz et al. 1993), the United States of America (APHIS 1996), Iran (Torarbi et al. 1996), and the Republic of South Africa (Crous et al. 2001). *Triticum aestivum* is most susceptible plant species to Karnal bunt (Fig. 11). Under artificial inoculation, some lines may show more than 50% infected grain (Fuentes-Dávila et al. 1992; 1993). The causal agent of this disease is the fungus *Tilletia indica* (Mitra 1931) (syn. *Neovossia indica*). Although *T. indica* may affect durum wheat and triticale (Agarwal et al. 1977), the level of infected grain is generally low. Control of this pathogen is difficult, because the teliospores are resistant to physical and chemical factors (Krishna and Singh 1982; Zhang et al. 1984; Smilanick et al. 1988). Chemical control can be accomplished by applying fungicides during flowering (Fuentes-Dávila et al. 2005), however, this measure is not feasible when quarantines do not allow tolerance levels for seed production. Resistant wheat cultivars are the best means to control this disease. Our objective was to evaluate 21 advanced bread wheat lines and four commercial cultivars for resistance to Karnal bunt.

Fig. 11. Karnal bunt (*Tilletia indica*) symptoms on the grain of bread wheat.

Materials and methods. Twenty-one advanced bread wheat lines and four commercial cultivars (Roelfs F2007, Tepahui F2009, Ónavas F2009, and Villa Juárez F2009) (Table 6, p. 53) were evaluated for resistance to Karnal bunt during the fall–winter of the 2011–12 crop season in block 910 in a clay soil with a pH 7.8, in the Yaqui Valley, Sonora, Mexico. Sowing dates were 30 November and 9 December, 2011, using a 1-m bed with two rows. Inoculum was prepared by isolating teliospores from infected kernels, followed by centrifugation in a 0.5% sodium hypochlorite solution and plating on 2% water-agar Petri plates. After teliospore germination, fungal colonies were transferred and multiplied on potato-dextrose-agar. Inoculations were carried out by injecting 1 mL of an allantoid sporidial suspension (10,000/mL) during the boot stage (Fig. 12) in 10 heads from each line and cultivar. High relative humidity in the experimental area was provided by a fine spray of water with back-pack manual sprayers. Harvest was done manually, and the counting of healthy and infected grains was done visually to determine the percentage of infection. Evaluated lines originated from the collaborative project between the International Maize and Wheat Improvement Center (CIMMYT) and the National Institute for Forestry, Agriculture and Livestock Research in Mexico (INIFAP).

Fig. 12. Artificial inoculation of bread wheat with a sporidial suspension of *Tilletia indica* by boot injection in the field.

Results. The range of infection for the first planting date was 0.66–24.35%, with a mean of 8.96 (Fig. 13, p. 54). The range of infection for the second planting date was 0.57–30.04%, with a mean of 12.44. The frequency of lines in the different infection categories in the two dates is shown in Fig. 14 (p. 54). The mean of the three highest percentage of

Table 6. Advanced bread wheat lines and commercial cultivars artificially inoculated with karnal bunt (*Tilletia indica*) in the field in two sowing dates, during the 2011–12 crop season, in the Yaqui Valley, Sonora, Mexico.

	Pedigree and selection history
1.	Roelfs F2007
2.	Tepahui F2009
3.	Onavas F2009
4.	Villa Juárez F2009
5.	PBW343//CAR422/ANA/3/ELVIRA CMSS02M00409S-030M-1Y-0M-040Y-10ZTB-0Y-02B-0Y
6.	MONR/3/GAV/ROM//BAR TC-060024-10R-0OAX-0R-6C
7.	MILAN/KAUZ//PRINIA/3/BAV92/4/PASTOR*2/BAV92/5/TACUPETO F2001*2/KUKUNA CMSA05Y01021T-040M-040ZTP0Y-040ZTM-040SY-14ZTM-01Y-0B
8.	SOKOLL*2/TROST CMSA05Y01186T-040M-040ZTP0Y-040ZTM-040SY-12ZTM-03Y-0B
9.	SOKOLL*2/3/BABAX/LR42//BABAX CMSA05Y01225T-040M-040ZTP0Y-040ZTM-040SY-12ZTM-01Y-0B
10.	WBLL1*2/4/SNI/TRAP#1/3/KAUZ*2/TRAP//KAUZ/5/KAUZ//ALTAR 84/AOS/3/MILAN/KAUZ/4/HUITES CMSS05B00060S-099Y-099M-099Y-099ZTM-14WGY-0B
11.	TACUPETO F2001/6/CNDO/R143//ENTE/MEXI_2/3/AEGILOPS SQUARROSA (TAUS)/4/WEAVER/5/PASTOR/7/ROLF07 CMSS06Y00716T-099TOPM-099Y-099ZTM-099Y-099M-3RGY-0B
12.	ROLF07/4/BOW/NKT//CBRD/3/CBRD/5/FRET2/TUKURU//FRET2 CMSS06Y00605T-099TOPM-099Y-099ZTM-099Y-099M-11WGY-0B
13.	WAXWING/4/SNI/TRAP#1/3/KAUZ*2/TRAP//KAUZ/5/KIRITATI//ATTILA*2/PASTOR CMSS07Y00288S-0B-099Y-099M-099Y-17M-0WGY
14.	WBLL1*2/KURUKU/4/PFAU/SERI.1B//AMAD/3/WAXWING CMSS07Y00338S-0B-099Y-099M-099Y-9M-0WGY
15.	STLN/MUNAL #1 CMSS07Y00452S-0B-099Y-099M-099Y-22M-0WGY
16.	PBW343*2/KUKUNA*2//FRTL/PIFED CMSS06Y00831T-099TOPM-099Y-099ZTM-099NJ-099NJ-5WGY-0B
17.	PBW343*2/KUKUNA*2//FRTL/PIFED CMSS06Y00831T-099TOPM-099Y-099ZTM-099NJ-099NJ-22WGY-0B
18.	WBLL1/FRET2//PASTOR*2/3/MURGA CMSS06Y00937T-099TOPM-099Y-099ZTM-099Y-099M-8WGY-0B
19.	FRET2/TUKURU//FRET2/3/MUNIA/CHTO//AMSEL/4/FRET2/TUKURU//FRET2 CMSS06Y00878T-099TOPM-099Y-099ZTM-099Y-099M-18WGY-0B
20.	KACHU/3/C80.1/3*BATAVIA//2*WBLL1/4/KACHU CMSS06Y01283T-099TOPM-099Y-099ZTM-099Y-099M-14WGY-0B
21.	BECARD/KACHU CMSS06B00169S-0Y-099ZTM-099Y-099M-13WGY-0B
22.	FRET2/TUKURU//FRET2/3/TACUPETO F2001*2/KIRITATI CMSS07B00081S-099M-099NJ-099NJ-12WGY-0B
23.	BECARD/QUAIU #1 CMSS07B00230S-099M-099NJ-099NJ-21WGY-0B
24.	ROLF07*2/5/REH/HARE//2*BCN/3/CROC_1/AE.SQUARROSA (213)//PGO/4/HUITES CMSS06B00704T-099TOPY-099ZTM-099Y-099M-23WGY-0B
25.	BECARD/AKURI CMSS06B00411S-0Y-099ZTM-099Y-099M-1WGY-0B

infection of the susceptible check was 100%. Only the lines 'FRET2/TUKURU// FRET2/3/MUNIA/CHTO//AMSEL/4/FRET2/TUKURU//FRET2' and 'KACHU/3/C80.1/ 3*BATAVIA//2*WBLL1/4/KACHU' were in the 2.6–5.0% infection category; lines with less than 5% infection are considered resistant (Fuentes-Dávila and Rajaram 1994). Tepahui F2009 and seven lines had 5.1–10.0% infection, and Roelfs F2007, Ónavas F2009, and Villa Juárez F2009 and 12 lines had 10.1–30.0% infection. Commercial cultivars with the highest infection were Roelfs F2007 (10.19%) and Villa Juárez F2009 (18.53%). Lines that showed the highest infection were 'FRET2/TUKURU//FRET2/3/TACUPETOF2001*2/KIRITATI' (27.41%) and 'ROLF07/4/BOW/NKT//CBRD/3/CBRD/5/FRET2/TUKURU//FRET2' (30.03%).

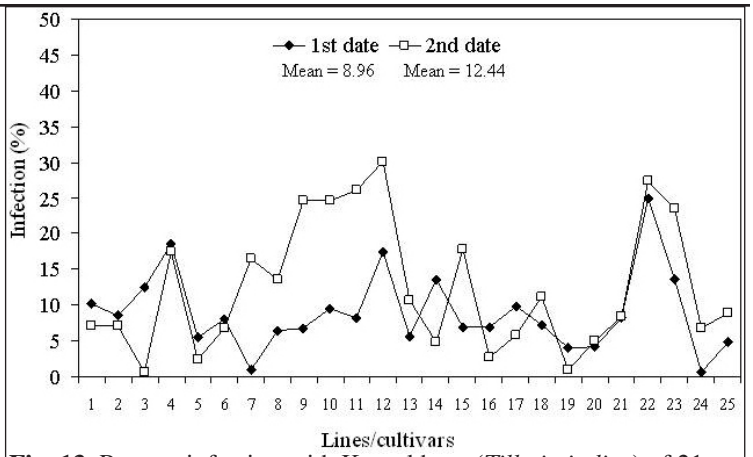

Fig. 13. Percent infection with Karnal bunt (*Tilletia indica*) of 21 elite advanced bread wheat lines and four commercial cultivars artifically inoculated in the field at two dates during the 2011–12 crop season in the Yaqui Valley, Sonora, Mexico.

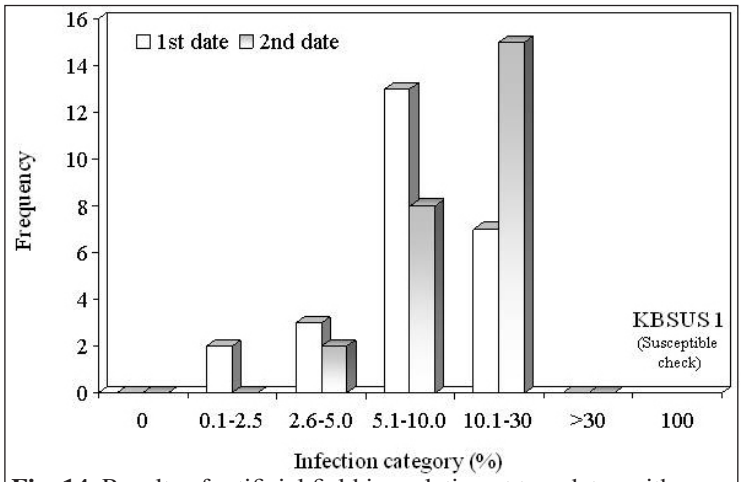

Fig. 14. Results of artificial field inoculation at two dates with Karnal bunt (*Tilletia indica*) of 21 elite advanced bread wheat lines and four commercial cultivars during the 2011–12 crop season in the Yaqui Valley, Sonora, Mexico. The infection level of KBSUS 1 is the mean of the three highest infection scores.

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Study on dissemination of Tilletia indica propagules from artificially inoculated wheat spikes that might cause infection to neighboring noninoculated spikes.

Guillermo Fuentes-Dávila.

Abstract. Experiments were conducted at the Norman E. Borlaug Experimental Station, in Mexico during the 1987–88 to 1990–91 crop seasons, to determine if there is spread of fungal propagules from artificially inoculated spikes to neighboring noninoculated ones. Inoculated spikes were injected during the boot stage with 1 mL of a sporidial suspension with a concentration of 10,000/mL. In 1987–88, data was collected from the susceptible cultivars Genaro T81, Glennson M81, and WL711, which were inoculated on 33 different dates. Approximately 50 spikes were collected around each inoculated spike. In crop season 1988–89, WL711 was inoculated on four dates, five in 1989–90 in five, and seven in 1990–91. The number of spikes evaluated were 40 in 1988–89, 1989–90 50 in, and 50 in 1990-91. The number of inoculated spikes per date were five in 1988–89, six in 1989–90, and ten in 1990–91. At maturity, spikes around the inoculated spikes were harvested and threshed by hand to determine the number of healthy and infected grains. The percent of spikes infected with Karnal bunt during crop seasons 1987–88 was 0.19 (3,033 spikes evaluated), 0.12 in 1988–89 (800 spikes), 0.40 in 1989–90 (1,500 spikes), and 1.74 in 1990–91 (3,450 spikes). The total percent affected spikes, 74 out of 8,783, was 0.84. The percent infected wheat grains with Karnal bunt during 1987–88 was 0.029 (126,647 spikes evaluated), 0.003 in 1988–89 (33,405 spikes), 0.10 in 1989–90 (67,468 spikes), and 0.14 in 1990–91 (135,457 spikes). The total percent of infected grains, 300 out of 362,977, was 0.082. Results of the experiments showed that dissemination of fungal propagules from inoculated spikes is nil or extremely low.

Introduction. *Tilletia indica* Mitra (syn. *Neovossia indica* (Mitra) Mundkur) is the causal agent of Karnal bunt disease of wheat, which was first identified in India (Mitra 1931), and later in Mexico (Duran 1972), Pakistan (Munjal 1975), Nepal (Singh et al. 1989), Brazil, (Da Luz et al. 1993), the United States of America (APHIS 1996), Iran (Torarbi et al. 1996), and the Republic of South Africa (Crous et al. 2001). Teliospores of this organism are resistant to extreme cold, heat, chemical treatments (Smilanick et al. 1985), and can survive up to three (Bonde et al. 2004) to four years in field soil (Krishna and Singh 1982), making control difficult. Fairly good chemical control with fungicide applications during flowering can be accomplished (Salazar-Huerta et al. 1997), however, in northwest Mexico, due to quarantine regulations (SARH 1987), this measure is still not profitable for commercial use. Evaluation of various *Triticum* species for resistance to *T. indica* has been on-going since the 1940s (Anonymous 1943: Bedi et al. 1949; Gautam et al. 1977; Singh et al. 1986; Singh et al. 1988). Aujla et al. (1986) reported the release of wheat cultivars WL1562, PBW120, and PBW65 as resistant in India, and Camacho-Casas et al. (1993) reported resistanc in cultivar Arivechi M92 in Mexico. The use of resistant cultivars offers the best alternative for farmers where Karnal bunt is a problem. Therefore, since the early 1980s, the Wheat Program of the International Maize and Wheat Improvement Center and the Mexican National Institute for Forestry, Agriculture, and Livestock Research implemented a project on breeding for resistance to *T. indica*. Because disease incidence is erratic in northwest Mexico, evaluation of experimental germ plasm can not rely on natural occurrence of the disease, therefore, this project is based primarily on artificial inoculation of experimental wheat germ plasm, which has brought up the concern of some government personnel and farmers that the fungus might be disseminated or spread from inoculated wheat spikes and if this activity might increase the inoculum level in the soil. Dhaliwal et al. (1988) reported trapping sporidia from underneath inoculated spikes under laboratory and field conditions and that these were infective on artificially inoculated wheat. They also reported that more plants had bunted kernels near inoculated spikes in the field, and that counts of such plants decreased with the increasing distance from the inoculated spikes. Later, Dhaliwal (1989) indicated that there was limited secondary spread of Karnal bunt around the inoculated spikes in spite of abundant sporidial production by them, suggesting that environmental conditions favorable for inoculum production may not necessarily be the same as for inoculum dissemination and establishment of infection. He found only 22 out of 60 infected grains in two groups of 20 uninoculated spikes. Those two groups were under sprinkler irrigation. Also, the possibility that infection was caused by natural inoculum can not be discarded. Bains and Dhaliwal (1989), reported that wheat spikes inoculated with Karnal bunt, produced secondary sporidia when later incubated (intact/detached) under moist conditions. Our objective was to determine if there is spread of *T. indica* propagules from artificially inoculated spikes that could cause infection in the neighboring noninoculated spikes. This work has not been published previously.

Materials and methods. Experiments were conducted at the Norman E. Borlaug Experimental Station, previously known as CIANO, located in the Yaqui Valley, Sonora, Mexico (27°20'N, 105°55'W, elevation 39 masl), during crop seasons 1987–88 to 1990–91. For inoculum preparation, one-year-old teliospores were taken from naturally infected wheat grains from various locations in the Yaqui Valley to assure a genetically heterogenous composite of the fungus population. To isolate teliospores, infected grains were agitated for about 15 seconds in a test tube containing a

water+tween solution using a vortex shaker, then sieved through a 60 um nylon microsieve to remove all debris. Teliospores were kept in the solution for 24 h to enhance germination. Thereafter, they were precipitated by centrifugation at 3,000 rpm, surface sterilized with sodium hypochlorite 0.5% a.i. for 2 min while centrifuging at 3,000 rpm, rinsed twice with distilled sterile water, and plated on water-agar (WA) 1.5%. Plates were incubated at room temperature (20–22°C) and teliospore germination was assessed starting five days after plating using a compound microscope at 10X. Pieces of agar with the fungus germinating were inverted onto the lids of potato-dextrose-agar (PDA) plates for ballistospore release and further multiplication. After 10 to 14 days, 2 to 3 mL of sterile distilled water were added to the plates and the colonies were scraped gently using a sterile spatula. Hyphae and sporidia were inoculated onto other plates with PDA using a sterile syringe, and the plates were incubated at 20°C in darkness for about nine days. After incubation, pieces of PDA with the different fungal propagules were transferred and placed upside down on the lids of sterile glass Petri plates, in order to induce production of allantoid secondary sporidia (Dhaliwal and Singh, 1989; Fuentes-Dávila et al., 1993). About three mL of sterile distilled water were added to the bottom of the plates. Then, the water-sporidia suspension was collected from the plates every 24 h; secondary allantoid sporidia were counted using a heamocytometer, and the concentration was adjusted to 10,000 per mL. Inoculation was performed by injection of 1 mL per spike in the boot (Zadoks et al. 1974), stages 48–49 and were identified with a colored plastic piece. Inoculations were carried out after 4 PM. To provide high relative humidity in order to assure a succesful infection, an overhead irrigation system was used after inoculation, and from 3–5 times/day for 15 min each time.

During the 1987–88 crop season, data was collected from inoculation of the susceptible cultivar Genaro T81, Glennson M81, and WL711, which had been artificially inoculated from 18 January to 3 March, giving a total of 33 different inoculation dates. Approximately 50 spikes were collected around each inoculated spike. For Genaro T81, 13 spikes were inoculated from 18 January to 10 February and 19 from 11–26 February; for Glennson M81, 17 spikes were inoculated from 19 January to 1 February; and for WL711, 12 spikes were inoculated from 11 Januray to 3 March. In the 1988–89 crop season, WL711 was inoculated on four dates (6, 11, 13, and 15 February), five in 1989–90 (24, 31 January, 8, 23 February, and 6 March) in 1989–90 in, and seven (18, 25 January, 1, 8, 15, 22 February, and 1 March) in 1990–91. The number of spikes evaluated were 40, 50, and 50 per inoculated spike per date in those three seasons. The number of inoculated spikes per date were 5 in crop season 1988-89, 6 in 1989-90, and 10 in 1990-91.

During 1987–88 to 1989–90, the experiment was carried out in material under an overhead irrigation system, and in 1990–91 without the system irrigation. The overhead irrigation was used to simulate rainfall and increase relative humidity, based on the fact that natural disease incidence is higher under those conditions (Aujla et al. 1977; Mundkur 1943; Bedi et al. 1949; Singh and Prasad 1978). However, because overhead irrigation might interfere with sporidial movement, the system was not used in 1991. At maturity, spikes around the inoculated ones were harvested and threshed by hand to determine the number of healthy and infected grains.

Results. The percent spikes infected with Karnal bunt (Fig. 15) was 0.19 in 1987–88 after evaluation of 3,033 spikes, 0.12 in 1988–89 (800 spikes evlautate), 0.40 in 1989–90 (1,500 spikes evaluated), and 1.74 in 1990–91 (3,450 spikes evaluated) (Table 7). The total infected spikes was 74 out of 8,783, or 0.84. The percent wheat grains infected with Karnal bunt to was 0.029 1987-88 (126,647 spikes evaluated), 0.003 in 1988–89 (33,405 spikes evaluated), 0.10 in 1989–90 (67,468 spikes evaluated), and 0.14 in 1990–91 (135,457 spikes evaluated) (Table 8, p. 57). The total percent of infected grains was 300 out of 362,977, or 0.082. The highest number of infected grains was obtained when no overhead irrigation was used, however, a greater number of spikes were evaluated, which increased the possibilities of finding

Fig. 15. Wheat spike showing infection by *Tilletia indica*.

Table 7. Number of infected spikes collected around spikes artificially inoculated with Karnal bunt (*Tilletia indica*) in the field during the 1987–88 to 1990–91 crop seasons at the Norman E. Borlaug Experimental Station, Yaqui Valley, Sonora, Mexico. The number of spikes evaluated in 1988 does not correspond to the expected, since some dates were repeated in three cultivars assessed.For inoculated spikes, the numerator indicates the numbe of inoculated spikes and the denominator the number of spikes collected around the inoculated spike.

	Inoculation		Spikes		
Season	Number	Spikes	Healthy	Infected	%
1987–88	33	1/50	3,033	6	0.19
1988–89	4	5/40	800	1	0.12
1989–90	5	6/50	1,500	6	0.40
1990–91	7	10/50	3,450	61	1.74

more. On the other hand, conducive environmental conditions for infection prevailed in the Yaqui Valley, which might have influenced a greater number of infected grains. Consequently, those grains might have been infected from inoculum coming from sources other than the artificially inoculated spikes. Despite the fact that the soil was not covered in the sorroundings of clusters of inoculated and noninoculated wheat plants, indications of sporidial spread or of any other fungal propagules is quite lacking, because the number of infected grains obtained was extremely low. If we assume that spread occurs in spikes closest to those that are inoculated, they would have the greatest chance of serving as points of inoculum arrival and, perhaps, would interfere with farther spread of propagules. The low frequency of spikes with a moderate number of infected grains can be considered as another indication of little or no spread of sporidia, considering also that the mean number of grains/spike (out of those evaluated) in cultivar WL711 is about 40. Results of the experiments show that dissemination of fungal propagules from inoculated spikes is nil or extremely low. In relation to the possibility of increasing inoculum levels in the soil by the activity of artificial inoculation in order to evaluate the reaction to *T. indica* of experimental germ plasm, because inoculated spikes are collected for evaluation, the possibilities of increasing inoculum levels in the soil are very low.

Table 8. Number of infected wheat grains obtained from spikes collected around spikes artificially inoculated with Karnal bunt (*Tilletia indica*) in the field during the 1987–88 to 1990–91 crop seasons at the Norman E. Borlaug Experimental Station, Yaqui Valley, Sonora, Mexico. The number of spikes evaluated in 1988 does not correspond to the expected, since some dates were repeated in three cultivars assessed.For inoculated spikes, the numerator indicates the numbe of inoculated spikes and the denominator the number of spikes collected around the inoculated spike.

Season	Inoculation		Grains		
	Number	Spikes	Healthy	Infected	%
1987–88	33	1/50	126,647	37	0.029
1988–89	4	5/40	33,405	1	0.003
1989–90	5	6/50	67,468	68	0.100
1990–91	7	10/50	135,457	194	0.143

Conclusions. Results of the experiments conducted during the 1987–88 to 1990–91 crop seasons showed that dissemination of fungal propagules from artificially inoculated spikes that could cause infection to neighboring noninoculated spikes is nil or extremely low.

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ITEMS FROM PAKISTAN

COMSATS INSTITUTE OF INFORMATION TECHNOLOGY (CIIT) Islamabad.

The effect of ACC deaminase (1-aminocyclopropane-1-carboxylate deaminase) in producing plant growth-promoting rhizobacteria on wheat under drought stress.

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Soil-borne microorganisms that help plants in promoting their growth and development are plant growth-promoting rhizobacteria (PGPR). They are abundantly present in the plant rhizosphere and survive on root exudates and lysates. Excessive production of ethylene by plants on exposure to certain biotic and abiotic stresses is known as 'stress ethylene'. Drought stress also has been suggested to alter tissue sensitivity to ethylene. In wheat, stress ethylene is increased by certain enzymes, such as benzyladenine (BA) and auxin, whereas ABA inhibits the biosynthesis of ethylene as in the unstressed condition. Thus, increased concentration of abscisic acid decreases ethylene in water-stressed wheat plants. A high level of ethylene slows down root elongation, root nodulation, and auxin transportation and encourages hypertrophies, paces up aging, and endorses the rate of senescence and abscission. For this reason, the severity of the infection from fungal or bacterial attack can significantly be reduced by inhibiting ethylene production. 1-aminocyclopropane-1-carboxylate deaminase (ACCD) was originally found inside a soil bacterium belonging to *Pseudomonas* sp. ACCD was recognized as an enzyme involved in the degradation of a cyclopropanoid amino acid (ACC) into α-ketobutyrate and ammonia. Ethylene and ACC (an intermediate) levels can be deceased up to 2–4 fold in wheat plants where the seedlings and roots are coated with ACCD-producing bacteria.

Collection, characterization for ACCD, and identification of bacterial strains. The bacterial strains were collected and characterized for the presence of ACCD. The ACCD-containing bacterial strains were identified by 16rRNA and were found to be of *Serratia* spp. and *Providencia* spp.

Sowing of wheat seeds coated with ACCD-producing bacteria. The experiment was conducted at the fields of Barani Agriculture Research Institute (BARI), Chakwal, which is considered a stress prone area. Field trial was conducted from October 2010 to May 2011. The experiment was laid out in randomized complete block design with four replications.

Root colonization assay. Twenty-five different bacterial isolates were collected from root rhizosphere and three were isolated from endosphere at 30 days postinoculation (Table 1). A total of 16 bacteria were isolated from root rhizosphere and three were isolated from endosphere. On the basis of their biochemical properties, the strains A, a, c, d, e, t, s, u, y, and z were selected for molecular characterization. ANOVA was applied to three bacterial isolates 30 days after inoculation and at harvest. The ACCD-producing *Serratia grimesii* strain z and *S. marcescens* strain t showed the best root colonization and persistence at 7.3 log CF U/g even at harvest. *Serratia grimesii* strain z also was found in the root endosphere at harvest. The root-colonizing ability of strains is given (Table 1).

Table 1. Statistical analysis of the root colonization assay for plant growth-promoting rhizobacteria. CFU = colony forming unit. All values are means of three replicates. Values with different letters are statistically different. LSD values at 0.05 = 0.2701).

Treatment	Log CFU
Cultivar	
Pasban-90	6.9 A
Sahar-2006	5.4 B
Inoculation	
Individual	7.5 C
Time	
30 days after sowing	6.7 E
Harvest	5.5 F
Strains	
A.1	7.3 G
A.2	7.3 G
F.11	3.8 H
Treatment interactions	
$V_1 I_1 T_1 S_1$	8.6 I
$V_1 I_1 T_1 S_2$	8.7 I
$V_1 I_1 T_1 S_3$	8.7 I
$V_1 I_1 T_2 S_1$	8.3 J
$V_1 I_1 T_2 S_2$	8.3 J
$V_1 I_1 T_2 S_3$	8.6 I
$V_1 I_2 T_1 S_1$	8.7 I
$V_1 I_2 T_1 S_2$	8.5 IJ
$V_1 I_2 T_1 S_3$	0.0 O
$V_1 I_2 T_2 S_1$	7.3 LM
$V_1 I_2 T_2 S_2$	7.1 M
$V_1 I_2 T_2 S_3$	0.0 O
$V_2 I_1 T_1 S_1$	7.5 KL
$V_2 I_1 T_1 S_2$	7.6 K
$V_2 I_1 T_1 S_3$	7.6 K
$V_2 I_1 T_2 S_1$	5.3 FN
$V_2 I_1 T_2 S_2$	5.3 FN
$V_2 I_1 T_2 S_3$	5.5 FN
$V_2 I_2 T_1 S_1$	7.5 KL
$V_2 I_2 T_1 S_2$	7.5 KL
$V_2 I_2 T_1 S_3$	0.0 O
$V_2 I_2 T_2 S_1$	5.3 FN
$V_2 I_2 T_2 S_2$	5.3 FN
$V_2 I_2 T_2 S_3$	0.0 O

Molecular characterization. PCR was by extracting DNA by boiling. After purification, the PCR product was sequenced and aligned (Fig. 1). The amplified 1,500-bp band was found to have close homology with two strains of *Serretia* spp. showing that the inoculated strain survived in the field.

Fig. 1. The bright band at 1,500 bp shows the amplification of the 16srRNA gene of selected strains from a root rhizosphere and endosphere population.

Field evaluation. The following parameters were evaluated at harvest.

Plant height. ANOVA was used to deduce that plant height was not affected by any treatment. Differences in cultivar plant height could not be due to the effect of the treatment (Table 2).

1,000-kernel weight. ANOVA deduced that 1,000-kernel weight also was not affected by any of the treatments individually but it had affects in combination (Table 2) and also a cultivar effect. Pasban-90 and Sahar-2006 had higher grain weights.

Grain yield. ANOVA revealed that grain yield was higher in most of the treatments and highest in treatment 4, which was the combination of all three treatments. Yield was least in treatments 5 and 6 (Table 2).

Table 2. Statistical analysis of treatments given and their effect on different parameters (NS = not significant; all values are means of three replicates; values with different letters are significantly different; LSD values for plant height at 0.05 = 9.638, 1,000-kernel weight at 0.05 = 5.463, and for grain yield at 0.05 = 0.2701).

Parameter	Plant height (cm)	1,000-kernel weight (g)	Grain yield (t/ha)
Treatment			
T_1 : *Serratia grimesii* strain A2	103.5	45.65	1. 96 a
T_2 : *Serratia marcescens* strain A3	98.78	45.17	1.82 b
T_3 : *Serratia liquefaciens* strain F11	99.78	45.77	1.99 b
T_4 : Consortium	103.0	47.09	2.81 a
T_5 : ACCD-negative strain R9	97.42	47.54	2.20 a b
T_6 : Control	98.34 NS	44.78 NS	2.05 b
Cultivar			
Pasban-90	97.50 a	37.40 a	1.91 c
Sahar-2006	102.80 b	54.61 b	2.37 d
Treatment combination			
$T_1 V_1$	98.53 c d e	36.74 c d	1.90 f g
$T_2 V_1$	98.45 c d e	36.60 c d	1.90 f g
$T_3 V_1$	96.24 d e	36.06 e	1.95 f g
$T_4 V_1$	99.90 d e	36.85 d e	2.50 e f
$T_5 V_1$	89.99 e	41.80 d	1.65 g
$T_6 V_1$	101.9 c d	36.35 d e	1.60 g
$T_1 V_2$	99.06 c d e	54.56 c	2.02 f g
$T_2 V_2$	101.1 c d	53.75 c	1.74 g
$T_3 V_2$	100.4 c d	55.49 c	2.0 b g
$T_4 V_2$	106.1 c	57.33 c	3.11 e
$T_5 V_2$	104.8 c d	53.28 c	2.75 e
$T_6 V_2$	105.2 c d	53.22 c	2.50 e f

Other parameters. Other parameters, such as germination %, plant height, spike length, grains/spike, and canopy temperature, also were recorded (Table 3, pp. 61). None showed a significant effect when inoculated with PGPR.

Table 3. List of parameters used to evaluate the effect of plant growth-promoting rhizobacteria on two wheat cultivars.

Cultivar	Treatment	Replication	Germination (%)	Plant height (cm)	Spike length (cm)	Grains/spike	Canopy temperature (°C)
Pasban-90	1	1	80	96.62	9.0	38.6	16.1
	2	1	80	95.40	7.9	31.7	15.7
	3	1	80	86.25	8.9	42.3	15.4
	4	1	75	90.52	9.2	37.3	17.5
	5	1	90	92.35	8.9	46.0	17.1
	6	1	90	96.62	8.9	33.6	16.5
Sahar-2006	1	1	80	104.54	8.1	44.0	16.0
	2	1	80	111.86	8.5	41.0	16.5
	3	1	85	109.72	8.6	37.3	16.7
	4	1	85	104.54	8.8	41.0	16.5
	5	1	90	105.76	9.1	42.3	16.2
	6	1	90	106.68	7.7	35.0	16.7
Pasban-90	1	2	80	98.45	10.8	55.3	16.3
	2	2	85	101.49	7.3	20.3	16.2
	3	2	85	94.48	8.5	38.0	15.5
	4	2	80	91.44	7.5	33.3	16.4
	5	2	95	93.26	8.0	39.0	15.8
	6	2	95	104.54	8.0	52.3	15.7
Sahar-2006	1	2	70	97.53	7.7	37.0	17.2
	2	2	70	98.45	7.5	54.3	17.6
	3	2	85	100.58	9.2	45.0	16.0
	4	2	80	100.58	8.1	40.6	17.5
	5	2	95	102.41	7.5	28.0	16.0
	6	2	95	107.59	9.8	60.3	17.3
Pasban-90	1	3	85	105.76	8.0	38.0	16.3
	2	3	90	104.54	7.8	37.3	16.8
	3	3	85	111.86	9.5	39.4	15.4
	4	3	75	108.81	9.7	37.0	16.8
	5	3	85	90.22	8.2	37.3	16.8
	6	3	90	105.76	8.0	26.6	16.5
Sahar-2006	1	3	85	88.39	9.0	44.3	15.9
	2	3	85	90.52	7.5	34.0	16.3
	3	3	75	86.86	10.0	56.0	18.0
	4	3	90	109.72	9.4	42.0	16.0
	5	3	90	107.59	8.8	39.7	16.6
	6	3	90	94.48	8.2	37.6	18.3
Pasban-90	1	4	90	93.26	8.0	38.0	16.0
	2	4	75	92.35	8.7	39.0	15.8
	3	4	85	92.35	8.4	36.3	15.7
	4	4	80	108.81	10.0	40.6	15.5
	5	4	90	84.12	7.4	37.3	17.2
	6	4	95	100.58	11.0	59.3	15.4
Sahar-2006	1	4	80	105.76	8.8	38.0	16.0
	2	4	85	103.63	9.0	37.0	16.3
	3	4	85	104.54	8.6	39.7	15.9
	4	4	85	109.72	9.8	43.3	17.1
	5	4	85	103.63	9.1	40.0	16.8
	6	4	95	111.86	11.5	56.0	14.0

Discussion. Ethylene is the major plant phytohormone and is produced by all almost all plants. Microorganisms present in the soil that produce ACCD promote plant growth by not only sequestering the ACC but also by lowering ethylene. Thus, lower ACCD levels make wheat plants more tolerant and resistant to stress. Soil in close proximity of plant roots is rich in ACCD-containing bacteria and more than 50% of these PGPR belong to *Pseudomonas* spp. and similar bacteria such as *Enterobacter* spp. This study gives information that none of the ACCD-positive strains belonged to either species.

In vivo evaluation was in the field. A root-colonization assay was done in order to check the survival of inoculated strains. Because different strains have different ability to colonize root rhizopshere. Colonization of these PGPR also depends upon the cultivar of wheat. The root-colonizing ability of PGPR was greater in Pasban-90 compared to that for Sahar-2006 (Table 1, p. 59), which can be due to the difference in root exudate composition in the rhizosphere that may vary due to the differences in cultivars. Rhizospheric conditions also vary greatly between cultivars. Elevation of the pH in the root rhizosphere was indicated in Sahar-2006. A soil analysis at BARI, Chakwal, showed that the soil was basic, the elevation in pH making it more alkaline and reducing the rhizophere population. The root-colonization assay indicated that in treatment 4, which was combination of all strains, F11 was not found, but was present in treatment 3, which consists of F11. We deduced that other bacteria, A2 and A3, may kill F11, so it was not found. F11 also was not found in the sequencing results. Sequencing of the bacterial isolates obtained from the root-colonization assay showed that the inoculated strains were present until harvest, because a *Serratia* sp. was obtained in the result. We concluded that these PGPR not only improve plant growth but also competitively survive, surpassing other bacteria, including pathogens. Because they were found throughout the experiment, they give a indication of their powerful nature to survive in any soil condition.

During the course of the experiments, 1,000-kernel weight was not affected by any of the treatments (Table 2, p. 60). Although grain weight is considered to be an important yield-contributing factor, individual grain weight was increased, reduced, or unaffected by the treatments or it may have relation with the number of grains obtained and the degree of environmental stress. Moreover, several reports indicate an inverse relationship between grain number and grain weight. The treatments yielding higher grain number have less grain weight, as was the case for Pasban-90 and Sahar-2006. Pasban-90 had higher grain number and less grain weight, whereas Sahar-2006 had a lower number of grains and their weight was greater. We also observed that these PGPR have different effects on different wheat cultivars, because their root-colonizing ability and survival in the root rhizosphere and endopshere are is cultivar-specific.

The effect of PGPR on individual wheat cultivars was significant in plant height but was not significant in combinations (Table 2, p. 60, and Table 3, p. 61). Plant height was greater for Sahar-2006 compared to that of Pasban-90. Many report that the plant height of Sahar-2006 generally is greater than that of Pasban-90, therefore, the effect of PGPR on plant height is not significant.

Wheat grain yield was higher in most of the treatments and was the best in treatment 4, which was the combination of treatments 1, 2, and 3. Yield was least in treatments 5 and 6. The improvement in grain yield under different treatments was different because of the effect of PGPR inoculated. Second, rhizobacteria containing the ACCD gene inhibit ethylene synthesis thus enhancing the growth of primary roots, seedling roots, and shoot growth, which are the major factors contributing to increased grain yield. ACCD-producing rhizobacteria decrease seedling ethylene, reducing stress in the seedlings. Treatment 4 may be the best because the combination of all the strains may have competitive nature, whcih suppresses other pathogenic strains that harmful for the plant. As a result, treatment 4 increased the number of productive tillers/m^2 and the number of grain/spike.

Conclusion. This research looks at the response of rhizobacteria on root growth and hormone signalling to positively enhance plant yield, thus increasing the plant water usage and harvest index. *Serratia* spp. proved to increase all yield components. However, the fact that *Serratia grimesii* strain A2, *S. marcescens* strain A3, and *S. liquefaciens* strain F11 remained in the root rhizosphere and endopshere throughout the trial proved that they influenced both systemic and local hormone signaling. These microorganisms containing the ACCD enzyme can be used in agriculture to promote plant growth, because they are both environment friendly and also cost effective. Plants inoculated with ACCD-containing bacteria proved to environmentally sociable and also cost effective compared to other technologies employed in agriculture against drought, high salt concentration, and high metals.

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WHEAT WIDE CROSSES AND CYTOGENETICS

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The Wheat Wide Crosses Program in Pakistan.

A. Mujeeb-Kazi and Alvina Gul Kazi.

The Wheat Wide Crosses Program around applied targets has its seat in Mexico at CIMMYT since early 1970s and was executed from 1979 until late 2004 by the senior author of this lead article. Upon retirement in late 2004, the CIMMYT program underwent an evolution and several areas were moved to Pakistan to be carried on in this new location but remaining under the leadership A. Mujeeb-Kazi, who was recruited by the Government of Pakistan.

The program is targeted heavily around prebreeding to harness genomic diversity encompassing basic, strategic, and applied tangents by utilizing the diversity potential of all three Triticeae gene pools. The focus is to address major biotic and abiotic stresses complimented by quality trait analyses crucial for nutritive value and grain export advantage.

The abiotic stresses influenced by climate change include tolerance to heat, drought, and salinity/sodicity. On a minor level, at this stage, is the threat of low temperature on which breeders have their sights set. The biotic stresses are stem, yellow, and leaf rust; Karnal bunt; and to a lesser level the new emergence of spot blotch, with sporadic presence in limited acreages of powdery mildew and barley yellow dwarf virus (BYDV).

Stresses are influenced by new pathogen emergence and the cropping systems. The first and most glaring example is the concern Pakistan faces from stem rust entry from the west via Iran around the race Ug99 and its variants. Cropping systems that follow rice and cotton delay wheat planting causing terminal heat to become a significant production constraint. In general, climate change has led to rainfall fluctuations that erratically influence output production in the rainfed areas. Hence, national total yields have stayed around 25 to 30 maunds/acre when the potential of such widely adapted germ plasm can approach 85 to 90 maunds/acre. National mean productivity levels as of the 2012 summer May harvest are 24.5 x 10^6 tons at 2.6 tons/ha. The yield gap remains huge and, if managed well, can add significantly to self-sufficiency, which can be addressed via management practices and varietal uniqueness where genetic diversity has a distinct role because the base of national cultivars is quite narrow. We have a heavy reliance on introduced germ plasm and the better adaptabilty of entries built on winter/spring combinations that possess the T1BL·1RS translocation all possessing a pedigree that have four wheats involved, i.e., Kavkaz, Buho, Kalyansona, and Bluebird. A prudent way forward would to diversify an effort the Wide Crossing Program is addressing.

Our efforts are a shift from the adaptive breeding dominance to take on recombination breeding using genetic resources that are not used in such creativity output by a majority of the national programs. Where we also follow the conventional breeding to a smaller degree, new modes are used like limited backcrossing and selected modified bulk as an efficient process with sights set on integrating doubled haploidy to achieve homozygosity on F_3 selections. The genetic diversity so far has exploited the D, A, and to a limited extent the B(S) genomes of the wheat diploid progenitor species/accessions based upon stocks that were brought in from CIMMYT, Mexico, where they were produced (Mujeeb-Kazi et al. 2004).

The current program in Pakistan is targeted around the production constraints mentioned above having the following major components:

- Exploiting the D- and A-genome, synthetic hexaploid wheats of up to 1,400 derivatives.
- Categorizing the national land races for all major stresses and their incorporation in cultivar development.
- Breeding specifically for yield components that target 1,000-kernel weight, overall spike parameters with a focus on large spikes and multiple ovaries, stay green, prostrate habit, and exploiting intraspecific diversity that includes resistant durums, multiple accessions of *T. turgidum* subsps. *dicoccum*, *dicoccoides*, and *carthlicum*, and, at the bread wheat level, investigate the potential of *T. aestivum* subsp. *spelta*.

— Maintaining global genetic stocks with high focus on wheat/alien chromosome translocations where those already available are getting transferred into national elite wheats via recurrent backcrossing and, in addition, producing new translocations with the salt tolerant and stem rust resistance genetic potential of *Thinopyrum bessarabicum* receiving the main attention. Global partnerships have allowed links to study other resources where *Haynaldia villosa*, *Leymus racemosus*, and *Secale cereale* take priority.

Molecular areas relate to adding efficiency to our major biotic and abiotic stresses targets in the program so far include QTL mapping, association mapping, extensive phenotyping to harness genotyping data (DArT) available globally derived from synthetic hexaploid wheats, use of allele specific markers for cereal quality (HMW, LMW, and grain hardiness), biotic stresses including plant growth aspects, e.g., stay green, dwarfing, and coleoptile length. The present genetic holdings of greater significance are in Table 1.

Table 1. Genetic holdings of greater significance to the Wide Crosses Program, National Agricultural Research Center, Islamabad, Pakistan.

Germ plasm	Entries	Source
Landraces	1,012	Pakistan
A -genome synthetics	194	CIMMYT, Mexico
B-genome synthetics	20	CIMMYT, Mexico
D-genome spring synthetics	1,400	CIMMYT, Mexico
D genome winter synthetics	84	CIMMYT, Mexico
Wheat/alien translocations	24	Kansas State University
CS monosomic, telosomic, di-telosomic lines	All	University of Missouri
Wheat/ alien translocations (Pavon)	67	University of California
Wheat/alien addition lines (*Th. bessarabicum*)	8	CIMMYT and Pakistan
Mapping populations	9	CIMMYT and Pakistan
Wheat/alien amphiploids	25	CIMMYT and Pakistan
Wheat/alien BC_1 self-fertile	19	CIMMYT and Pakistan
Aegilops tauschii	137	US (Idaho)
Aegilops speltoides	75	US (Idaho)
T. monococcum subsps. *monococcum* and *aegilopoides* and *T. urartu*	1,923	US (Idaho)
T. turgidum subsp. *dicoccum*	700	US (Idaho)
T. turgidum subsp. *dicoccoides*	800	US (Idaho)
T. turgidum subsp. *polonicum*	70	US (Idaho)
T. turgidum subsp. *sphaerococcum*	30	US (Idaho)
T. turgidum subsp. *carthlicum*	78	US (Idaho)
T. aestivum subsp. *spelta*	1,300	US (Idaho)
Secale cereale	153	US (Idaho)

Program modus operandi.

The national partners associated with the Wide Crosses Program are spread across all the provinces of Pakistan and are based upon multidisciplinary ties that aid our efforts to combat the target major and minor wheat production constraints.

In Sindh, links are with groups in Sakrand, Tandojam, Jamshoro, and Karachi, with these collaborators planting at diverse sites within the province. Objectives covered are local stem rust races, salinity, heat and drought tolerance, and cereal quality with complete rheology coverage. Advanced pre-bred germ plasm also is evaluated for varietal release potential via wheat breeders for this province.

In the Punjab province, germp lasm is evaluated for stem rust, leaf rust, spot blotch, salinity/sodicity, drought, heat, Karnal bunt, and BYDV. This hub also has a focus on evaluating advanced pre-bred lines for their varietal potential through key breeding programs of the public and private sectors. Within this province, the wide crosses group has partnerships with molecular geneticists who interact in areas of QTL mapping, association/nested association mapping, and

wheat transformation. Our program is located in this province and, thus, all wide crossing areas from basic, strategic, and applied sectors are conducted here with rapid flow of germ plasm to all partners nationally and internationally for stress testing. Specific areas are cytology, cytogenetics, molecular cytology, biochemical genetics, molecular genetics, tissue culture, interspecific/intergeneric/intraspecific hybridization, in vitro screening for abiotic stresses, micronutrient assays, recombination, and pre-breeding

In the Khyber Pakhtunkhwa province, pre-breeding ties are in place with the breeding partners with stress evaluations being conducted for yellow rust, BYDV, and drought tolerance.

Lastly, in Baluchistan, our focus is on sharing materials with partners for evaluation and utilization of wide cross products for varietal outputs aided by screening for yellow rust and drought tolerance.

Across all provinces are alliances with national universities aiding student and young human resource development through hands on experience and assist degree program research functions that lead to M.Phil. and Ph.Ds.

Brief write-ups from various national partners across their working disciplines reflect the strong, integrative ties that are vital for this nations wheat based food security 2050 vision. Our program has, in addition, alliances with international partners where CIMMYT plays a major role in sharing of expertise, training our young professionals, and providing user friendly germ plasm for rapid exploitation in breeding programs, and also direct cultivar releases via the adaptation/selection course. International alliances have been a major conduit to knowledge generation of which the recent major publications (Published or in Process) are supportive.

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QTL mapping of doubled haploids for physiological attributes under drought stress.

Mehmoona Ilyas, Abdul Waheed, Husne Sahar Zaidi, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Drought stress is one of the major environmental constraints to crop plants, including wheat, worldwide. Synthetic hexaploids can act as a vehicle for improving crop tolerance against various biotic and abiotic stresses. A doubled-haploid (DH) population consisting of 140 individuals derived from a cross between Opata and SH223 was used to identify genomic regions associated with various quantitative, physiological attributes. The DH mapping population was phenotyped for chlorophyll content, chlorophyll flouresence, osmotic adjustment, and proline and superoxide dismutase content under control and drought stress over two years. Genotyping utilized 261 polymorphic wheat microsatellites from Gaterslaben (Germany) and the Agriculture Research Center (Beltsville, MD USA) simple sequence repeats. The linkage map of the DH population comprised 19 linkage groups covering a map length of 2,626 cM was constructed using MapMaker software. Major and minor QTL associated with quantitative physiological traits were identified using QGene software. The phenotypic variation explained by QTL was under high control compared to those for drought stress depicting the complex genetics of drought. The identified QTL are important for high-resolution mapping in synthetic hexaploid wheat. The 'SH223/Opata' DH mapping population provides an excellent genetic stock for digesting genetically complex quantitative attributes under biotic and abiotic environmental constraints. Genomic synteny was observed in DHs with rice because of the occurrence of chlorophyll content QTL on chromosomes 2, 4, and 7, and with maize due to the presence of a chlorophyll fluorescence QTL on chromosome 7.

Major QTL for chlorophyll content (*QTc.wwc-1B-S11*) in the DH mapping population at anthesis during drought stress was mapped on chromosome 1B and exhibited 10.09% phenotypic variation at an LOD score of 5.5. The main

allele for this QTL was from Opata. Two more QTL (*QTc.wwc-5B-S9* and *QTc.wwc-7B-S9*) for this trait were mapped on chromosomes 5B and 7B, respectively, under drought stress at anthesis and explained 10.09% and 12.30%, respectively, of the phenotypic variation. The positive allele for QTL on chromosome 5B was contributed by SH223, whereas the QTL on chromosome 7B was from Opata. Seven major and minor QTL for PCFK in the DHs were identified on chromosomes 1B, 7A, and 7D under control and drought stress conditions at anthesis. Only one minor QTL for osmotic adjustment in the DH mapping population was identified on chromosome 7A under drought stress at anthesis. The positive allele for this attribute was governed by SH223 and showed only 8.5% of phenotypic variation at LOD value of 2.5. QTL for chlorophyll content, osmotic adjustment, and chlorophyll fluorescence of the DHs were mapped on homoeologous group 7 under both control and drought stress conditions.

QTL mapping of doubled haploids for phenological attributes under drought stress.

Mehmoona Ilyas, Abdul Waheed, Nosheen Ilyas, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Drought, one of the major multidimensional environmental constraints to plant growth and productivity, impacts wheat yield in arid and semi-arid regions of the world. Synthetic hexaploids (SH) are considered a novel source of germ plasm under hostile environments because of their potential to cope with biotic and abiotic stresses. One hundred forty double haploids (DH) were evaluated for different phenological attributes at anthesis under drought stress (Table 2). Four minor and four major QTL for 1,000-kernel weight in a DH mapping population were identified on chromosomes 3D, 7B, 5A, 5B, and 3B under drought stress at anthesis. Six QTL for grain number in the DH mapping population were identified on chromosomes 1B, 2A, 3B, 5A, 7A, and 7B under preanthesis drought stress given over two years. Two minor and four major QTL for days-to-heading were identified in the population under drought stress at preanthesis. One major QTL (*QDh.wwc-5A-S9*) under drought stress explained 12.76% of the phenotypic variation at an LOD score of 4.54, flanking in the vicinity of *Xbarc10–Xwms71*. A second and third major QTL under drought stress were identified on chromosomes on 7A and 3B, respectively, and collectively explained 21.08% of the phenotypic variation.

Table 2. QTL identified in a doubled-haploid mapping populaton for phenological attributes under drought stress.

Parameter	1A	1B	1D	2A	2B	2D	3A	3B	3D	4A	4B	5A	5B	5D	6A	6B	6D	7A	7B	7D
1,000-kernel weight		X						X	X		X	X	X						X	
Grain number		X		X	X		X	X		X		X						X	X	
Spikelets/spike				X		X			X			X	X							
Days-to-heading		X		X				X				X		X				X	X	
Days-to-physiological maturity	X			X	X			X		X	X	X	X			X			X	
Spike length	X	X				X	X			X	X	X	X		X	X			X	
Plant height	X			X	X	X		X			X							X	X	
Total chlorophyll content		X			X			X		X			X					X	X	
Osmotic adjustment																		X		
Fo		X																		
Fv/Fo																		X		X
Fv/Fm																				X

The first major QTL for spike length (*QSl.wwc-2D-C9*) in the DH mapping population under controlled conditions was on chromosome 2D. This QTL explained 10.23% of the phenotypic variation at an LOD score of 6.5. Chromosomes 5A and 5B are considered important genomic regions for QTL for 1,000-kernel weight, plant height, physiological maturity, spike length, and days-to-heading under drought stress. QTL detected under control and drought stress conditions not only provide information for the polygenic inheritance and their interaction with the environment but also helps to identify molecular markers closely linked with quantitative traits.

Phenological evaluation and RGAP-based molecular characterization of some exotic wheat genetic stocks.

Mehreen Naz, Sobia Tabassum, Muhammad Ashraf, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Plants feed ~93% of the people of the world, two-thirds of which is contributed by the cereals (wheat, maize, rice, barley, sorghum, and millet). Approximately 80% of the global cereal production is wheat, maize, and rice. Among the cereals, wheat is the largest, being cultivated in 27 countries of the developing world. Of the two principle types of wheat, i.e., bread and durum, 90% is bread wheat. Wheat cultivation encompasses a major production area of 8.303 x 10^6 ha, thereby engaging 33% of the cultivated area of the country each year with a production around 21.7 x 10^6 tons in the last wheat season. The annual average volume of world wheat trade has been about 106 x 10^6 tons between 1999 and 2003. By 2020, the world's demand for wheat is expected to be 40% higher than that in 1990. The demand for wheat, based on production and stock changes, is estimated to increase from a current level of roughly 625 x 10^6 tons to around 813 x 10^6 tons in 2030 and to more than 900 x 10^6 tons by 2050.

Wheat production is threatened by various factors. Apart from abiotic factors, including scarcity of water, drought, and salinity, biotic factors, such as the rust diseases, are the oldest known to humans. Stripe or yellow rust is one wheat disease that has caused yield loss. These diseases of cereals cause low germination, slower growth, shorter height, foliar damage, reduced floret set, stumpy forage quality, and reduced grain yield. Like other developing countries, agriculture is the most important sector of Pakistan's economy and food. Wheat is the country's most important agricultural product, cultivated by 80% of the farmers and contributing to roughly a quarter of the total crop sector value added. Of the total wheat production area of 8.303 x 10^6 ha in Pakistan, 70% is prone to stripe rust, an area of approximately 5.8 x 10^6 ha. In Pakistan, wheat production is threatened due to rust diseases, especially stripe rust. Stripe rust is significant in southern Pakistan, the foothills, northern areas, and Baluchistan. Molecular markers are being used to characterize genetic diversity, population structure, phylogenetic relationships, and pathotypes in the pathogen populations and to study the disease epidemiology and verify an extensive effective control of the disease. A variety of molecular marker systems have been used to study the stripe rust pathogen. Genetic diversity between and within diverse populations of wheat has been analyzed using molecular markers such as isozymes, restriction fragment length polymorphism (RFLP), random amplified polymorphic DNA (RAPD), amplified fragment length polymorphism (AFLP), simple-sequence repeats (SSR), and resistant gene analogue polymorphism (RGAP). Among these, RGAP markers have been developed from conserved domains of plant resistance genes and distributed in the genome to assess their validity for determining genetic diversity and genetic relationships. In addition, the RGAP approach has been utilized successfully to identify markers associated with resistance genes in wheat for stripe rust. SSRs have the capability to differentiate among closely related individuals for diversity and allelic variation across a broad range of germ plasm and have the advantage, over other markers, to trace plants pedigrees. In this study, we identified ergonomically suitable land races of wheat based upon phenological parameters and applied SSR and RGAP markers on wheat stripe rust for genetic diversity (Table 3, pp. 68-70).

To spot molecular markers linked to rust resistance gene(s) in wheat, RGAP and SSR techniques were used determine genetic diversity. Seven SSR primers were screened; primers CFD-13 and CFD-81 showed 29% genetic diversity and polymorphism. CFD-13 showed a high level of polymorphism. The co-efficient ranged from 0.44 to 1.00 for the SSR cluster analysis. Genetic diversity also was accessed using five RGAP primers. Only one RGAP primer, XLRR, gave positive results and showed a 20% polymorphism. Seven genotypes showed 6.25% polymorphism. The co-efficient ranged from 0.55 to 1.00. The SSR markers showed more genetic diversity compared to the RGAP markers. Based upon morphological characteristics, nine genotypes are the most appealing based on their higher number of grains/spike, lower inter-node distance, normal height, and grain quality and color. Out of 112 landraces, 14 were highly polymorphic (12.5% polymorphism). A number of exceptional lines, 7, 14, 26, 44, 45, 85, 107, and 110, were identified on the basis of higher number of grains/spike and lower internode distances ranging from 0.3 to 0.4 cm. The height of these lines is normal, lower than 100 cm, with well-filled grains.

Stripe rust is considered the major constraint in meeting the challenges of sustainable production and can cause yield loss worldwide. Constant growing of cultivars with major genes increases vulnerability, so there is a need to construct a breeding program that uses major genes in combination with minor genes. The competent monitoring of disease and the expansion of genetic diversity may result in valuable control of the disease and reduce major grain losses resulting from the stripe rust pathogen. So, it is clear now that wider the genetic makeup, the wider will be the diversity, and, therefore, the possibility of the crop affected by the pathogen will be reduced.

Table 3. A phenological characterization of 112 landraces of wheat (pubescence lacking (–) and present (+)).

Line	Pubes-cence	Plant height (cm)	Awns	Spikes/ plant	Grains/ spike	Spike length (cm)	Seed color	Internode distance (cm)	1,000—kernel weight (g)
1	—	97.5	gold	12	33	10	amber	0.3	33
2	—	85	gold	15	32	10	brown	0.4	31
3	—	70	gold	12	40	5	amber	0.4	40
4	—	97.5	gold	13	39	10	gold	0.4	39
5	—	92.5	gold	11	44	10	gold	0.5	36
6	—	82.5	gold	14	33	7.5	brown	0.3	30
7	—	72.5	gold	15	67	7.5	amber	0.5	28
8	—	77.5	gold	17	39	5	amber	0.3	39
9	—	80	gold	20	33	7.5	amber	0.3	36
10	—	75	gold	17	20	5	amber	0.3	41
11	—	97.5	gold	20	35	7.5	gold	0.3	43
12	—	87.5	gold	17	24	10	brown	0.2	37
13	—	75	gold	23	25	7.5	amber	0.3	30
14	—	90	—	22	45	7.5	gold	0.3	32
15	—	102	—	12	33	9.5	amber	0.2	37
16	+	82.5	—	19	37	7.5	amber	0.3	36
17	+	95	—	25	32	5	brown	0.3	29
18	—	97.5	gold	11	34	10	gold	0.3	39
19	—	105	gold	18	49	5	amber	0.5	42
20	—	97.5	gold	17	42	7.5	brown	0.5	37
21	—	102.5	gold	16	23	10	brown	0.6	42
22	—	92.5	gold	14	42	7.5	amber	0.4	37
23	—	120	gold	18	33	10	amber	0.4	32
24	—	102.5	—	17	23	12.5	amber	0.4	35
25	—	95.5	—	18	31	8	amber	0.4	36
26	—	92.5	gold	16	48	7.5	amber	0.5	41
27	—	93	—	16	32	8	amber	0.4	31
28	—	97.5	—	16	37	7.5	amber	0.5	42
29	—	97.5	gold	30	21	7.5	amber	0.6	63
30	—	97.5	gold	16	35	7.5	amber	0.5	24
31	—	87.5	—	20	31	7.5	amber	0.4	45
32	—	77.5	gold	14	41	7.5	amber	0.5	32
33	—	88	—	24	33	8	amber	0.4	41
34	—	87.5	—	20	34	7.5	amber	0.4	35
35	—	100	gold	15	33	10	amber	0.4	43
36	—	92.5	gold	18	38	7.5	amber	0.3	20
37	—	85	gold	12	13	7.5	amber	0.4	38
38	—	83	gold	10	31	8	brown	0.5	22
39	—	92.5	gold	7	35	7.5	amber/ white	0.4	36
40	—	100	—	14	41	7.5	amber/ brown	0.5	28
41	—	85	—	4	24	7.5	amber	0.4	14
42	—	80	—	9	38	7.5	amber	0.3	34
43	—	77.5	—	11	36	7.5	amber	0.4	39
44	—	131.5	gold	11	51	11.5	amber	0.4	31
45	—	115	—	13	64	12.5	amber	0.5	46

Table 3. A phenological characterization of 112 landraces of wheat (pubescence lacking (–) and present (+)).

Line	Pubes-cence	Plant height (cm)	Awns	Spikes/ plant	Grains/ spike	Spike length (cm)	Seed color	Internode distance (cm)	1,000—kernel weight (g)
46	—	112.5	—	15	30	10	amber	0.5	56
47	—	87.5	—	17	34	7.5	amber/ white	0.4	36
48	—	95	—	13	34	7.5	amber	0.5	33
49	—	80	—	14	40	7.5	amber	0.6	33
50	—	78	—	10	43	8	amber	0.5	22
51	—	86	—	17	39	7.5	amber	0.4	38
52	—	97	gold	18	40	9.5	amber	0.4	38
53	—	73	—	14	30	7.5	amber	0.4	36
54	+	78	gold	7	32	8	amber	0.4	59
55	+	90	—	20	46	7.5	amber	0.5	31
56	+	77.5	—	14	37	7.5	amber	0.6	37
57	+	80	—	15	40	7.5	amber	0.4	34
58	+	85	—	16	32	7.5	white/ amber	0.4	36
59	+	113	—	24	36	7.5	white/ amber	0.5	48
60	+	85	—	17	31	7.5	amber	0.5	39
61	+	77	—	16	39	7	amber	0.3	36
62	+	83	gold	17	31	7.5	amber	0.4	43
63	+	87.5	gold	12	42	7.8	white/ amber	0.5	30
64	+	108	gold	16	36	12.5	amber	0.4	41
65	—	83	gold	10	37	7.5	amber	0.3	34
66	—	97.5	gold	16	32	7.5	amber	0.4	38
67	—	97.5	gold	14	37	7.5	amber	0.3	38
68	—	93	gold	10	37	7.5	white/ amber	0.4	33
69	—	85	gold	18	31	10	amber/ brown	0.6	31
70	—	82.5	gold	9	47	10	amber	0.4	35
71	+	87.5	gold	12	49	7.5	amber	0.4	41
72									
73	+	102.5	gold	12	45	7.5	white/ amber	0.4	48
74	+	92.5	gold	8	48	7.5	amber	0.2	38
75	+	75.3	gold	10	38	7.8	amber	0.2	52
76	+	76	gold	7	57	7.5	amber	0.4	46
77	+	97	—	15	34	9.5	amber	0.3	35
78	—	90	—	17	40	7.5	amber	0.3	36
79	+	80	—	9	41	7.5	amber	0.3	41
80	+	95	—	15	35	7.5	amber	0.4	40
81	+	77.5	—	16	26	7.5	amber	0.3	35
82	—	92.5	—	14	30	7.5	brown	0.3	40
83	+	83	gold	20	42	8	amber	0.4	29
84	+	87.5	—	11	35	7.5	amber	0.4	33
85	+	85	gold	13	60	7.5	amber	0.3	43

Table 3. A phenological characterization of 112 landraces of wheat (pubescence lacking (–) and present (+)).

Line	Pubes-cence	Plant height (cm)	Awns	Spikes/ plant	Grains/ spike	Spike length (cm)	Seed color	Internode distance (cm)	1,000—kernel weight (g)
86	+	93	—	12	41	8	white/ brown	0.3	34
87	+	107.5	—	11	52	10	white/ brown	0.5	40
88	+	100	—	17	46	10	amber	0.3	34
89	+	97.5	—	18	39	7.5	amber	0.4	37
90	+	97.5	gold	17	37	7.5	amber	0.3	47
91	+	98	—	18	33	10.5	amber	0.3	30
92	—	108	—	13	45	8	amber	0.4	36
93	—	115	gold	10	30	10	white/ amber	0.7	29
94	+	111.5	gold	19	27	11.5	amber	0.6	36
95	+	121.5	gold	17	29	11.5	amber	0.5	41
96	+	118	gold	9	56	11	amber	0.4	36
97	—	107.5	gold	16	19	12.5	amber	0.5	43
98	+	103	gold	11	19	10.5	amber	0.4	22
99	+	104	gold	10	30	15	amber	0.4	27
100	+	100	gold	13	33	10	amber	0.5	34
101	+	107.5	gold	17	17	7.5	amber	0.4	37
102	+	98.5	gold	12	37	8.5	amber	0.5	42
103	+	89	gold	9	55	8.5	amber	0.5	37
104	—	85	gold	14	34	7.5	amber	0.5	37
105	+	83	gold	14	36	7.5	amber	0.6	45
106	—	106	gold	10	45	12.5	amber	0.4	24
107	+	85	gold	12	51	10	amber	0.4	37
108	—	77.5	gold	16	32	7.5	amber	0.4	44
109	+	100	gold	13	36	7.5	amber	0.3	42
110	+	73	gold	8	68	10.5	white/ amber	0.4	35
111	—	110	gold	3	19	10	amber	0.5	38
112	—	112	gold	13	23	5	amber	0.4	33

Detection of QTL for drought tolerance in a doubled-haploid mapping population.

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A doubled-haploid (DH) mapping population, with the pedigree 'Doy–1//*Aegilops tauschii* (458)/5/Opata', was planted under control (field, fully irrigated) and stress (rain shelter) conditions. The parents of mapping population, SH-349 (a D-genome based drought tolerant synthetic hexaploid wheat) and the bread wheat Opata M-85 (drought susceptible), also were grown individually in the field and shelter. A biochemical analysis included osmolyte determination, i.e., soluble sugar, proline, antioxidant superoxide dismutase, and cell membrane stability followed by molecular diagnostics, including DNA extraction, PCR, and capillary electrophoresis. Yield and yield components determined were days-to-heading, days-to-physiological maturity, plant height (cm), spike length (cm), number of spikelets/spike, 1,000-kernel weight (g), pubescence, awn color, and growth habit. The treatments were arranged in a complete randomized design, and comparison of means was by least significant difference. JoinMap4 and MapQTL5 were used for molecular diagnostics. Drought tolerant wheat lines were identified, and QTL controlling drought tolerance in the wheat mapping population were determined. Eight drought-tolerant lines were identified for different traits. A linkage map was constructed using SSR

markers; 141 markers used to screen the Opata, drought-sensitive, and SH-349, drought-tolerant, parents. Seventy-nine polymorphic markers were identified and applied on the mapping population. We identified 61 markers that showed linkage and 16 linkage groups. The map covered 14 chromosomes. The length of genetic linkage map was 665.3 cM with a density of 10.91 cM/marker. Interval mapping and multiple QTL mapping were used to detect 120 QTL. Many major QTL were identified (Table 4).

Table 4. QTL detected by multiple QTL mapping in a 'synthetic hexaploid/ Opata' doubled-haploid mapping population.

Line	QTL name	Chromosome	Trait	Environment
1	*QDH.C.1.MQ.wwc-2D*	2D	DH-1st	field
2	*QDH.S.1.MQ.wwc-2D*	2D	DH-1st	rain shelter
3	*QDPM.C.1.MQ.wwc-2D*	2D	DPM-1st	field
4	*QDH.C.2.MQ.wwc-7D*	7D	DH-2nd	field
5	*QDH.S.2.MQ.wwc-6A*	6A	DH-2nd	rain shelter
6	*QDPM.C.2.MQ.wwc-7A*	7A	DPM-2nd	field
7	*QDPM.S.2.MQ.wwc-7D*	7D	DPM-2nd	rain shelter
8	*QPH.S.2.MQ.wwc-7A*	7A	PH-2nd	rain shelter
9	*QSp.L.C.1.MQ.wwc-4A*	4A	Sp-L-1st	field
10	*QSp.L.S.1.MQ.wwc-7A*	7A	Sp-L-1st	rain shelter
11	*QPub.C.2.MQ.wwc-1B*	1B	Pub-2nd	field
12	*QTGW.S.1.MQ.wwc-5A*	5A	TGW-1st	rain shelter
13	*QAC.C.1.MQ.wwc-1B*	1B	AC-1st	field
14	*QAC.S.1.MQ.wwc-2D*	2D	AC-1st	rain shelter
15	*QAC.C.2.MQ.wwc-7D*	7D	AC-2nd	field
16	*QAC.S.2.MQ.wwc-7D*	7D	AC-2nd	rain shelter
17	*QG/S.C.1.MQ.wwc-6A*	6A	G/S-1st	field
18	*QG/S.S.1.MQ.wwc-1B*	1B	G/S-1st	rain shelter
19	*QG/S.C.2.MQ.wwc-6A*	6A	G/S-2nd	field
20	*QG/S.S.2.MQ.wwc-6A*	6A	G/S-2nd	rain shelter
21	*QPros.C.1.MQ.wwc-7D*	7D	Pros-1st	field
22	*QPros.S.2.MQ.wwc-7D*	7D	Pros-2nd	rain shelter
23	*QRL.C.MQ.wwc-2D*	2D	RL-C	hydro
24	*QSL.S.MQ.wwc-7A*	7A	SL-S	hydro
25	*QChlro.S.MQ.wwc-2A*	2A	Chlro-S	hydro
26	*QCMS.MQ.wwc-2A*	2A	CMS	hydro
27	*QPhoto.C.MQ.wwc-7D*	7D	Photo-C	hydro
28	*QPhoto.S.MQ.wwc-7D*	7D	Photo-S	hydro
29	*QProlin.S.MQ.wwc-7A*	7A	Proline-S	hydro
30	*QSugar.S.MQ.wwc-4A*	4A	Sugar-S	hydro
31	*QSOD.C.MQ.wwc-6A*	6A	SOD-C	hydro

In conclusion, our results show that some drought-tolerant or resistant genes in both parents (Opata and SH-349) can be transferred to susceptible genotypes using marker-assisted selection. A variety of factors may affect the outcome of a QTL analysis; for instance, the selection of the cross, the population structure and size, the number of measured replications and environments, and the density of markers. The magnitude of the QTL effect and accurate chromosome map location also are important for verifying the identified QTL. Overall, many QTL were detected across multiple traits, and the wild or synthetic line contributed both positively and negatively to these traits. We have some indication that the wild species Ae. tauschii donated some alleles, because QTL for some traits were associated with the D genome and with the synthetic line. This study provided additional evidence that QTL strategy is useful and is able to enhance the performance of existing cultivars. Further QTL studies will assist in the contribution of positive allelic diversity in the future.

Grain morphology and quality characterization of synthetic hexaploids to enhance its breeding value.

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Considerable genetic diversity resides in the wild progenitor species of bread wheat. A large collection of wild accessions of *Triticum turgidum* subsp. *dicoccum* and *Aegilops tauschii* has been captured in the primary synthetic wheats developed by the CIMMYT Wide Crosses Program. Diversity to various biotic (Mulki et al. 2012, Mol Breed 31:299-311) and abiotic stresses (Ogbonnaya et al. 2013, Plant Breed Rev 37:35-122) has been reported. Favorable genes for grain yield must be present in synthetic hexaploids, because several cultivars have been released in China, Spain, Ecuador, and Mexico from synthetic derivatives. However, no good strategy in place to identify and narrow down the wide array of synthetics based on grain quality and yield and attributes that enhance their true breeding value. The current study emphasizes a detailed genetic potential analysis at various grain quality loci and analyzing seed morphology of synthetic hexaploids using digital imaging technology.

Grain quality. The properties of processing quality were determined primarily by high- and low-molecular-weight gluten protein subunits in the flour that are associated with dough rheological properties determining bread-making quality. Polyphenol oxidase activity, yellow pigment content, grain hardiness (PINA, PINB, and PINBV2), lipoxygenase acitivity, zeta-carotene desaturase, and waxy protein have significant influence on the quality of end-use products. Several functional markers are available that cover the allelic variation at all contributing loci. A list of markers and the complete methodology is available at University of California–Davis website maintained by WheatCAP project (http://maswheat.ucdavis.edu/protocols/FunctionalMarkers/FM_quality.htm). We used these markers to elucidate allelic variation in 230 D-genome, synthetic hexaploids at major quality contributing loci.

Grain morphology. Useful and novel variation in untapped germ plasm needs state-of-the-art phenotypic characterization prior to their utilization. Maximizing yield is possible by enhancing the genetic gain from synthetic hexaploids by effective evaluation and exploitation. Grain yield primarliy depends on kernel weight, which is contributed to by several of the seed shape descriptors. The progress in phenotyping is not at par with high-throughput genotyping. More accurate, efficient, and high-throughput avenues for phenotyping need to be pursued in order to get maximum benefit from low-cost genotyping resources. In this scenario, photometric measurements or digital imaging provide more concise and cheaper phenotypic information and better elucidate the individual components of complex traits. The present study focuses on the state-of-the-art phenotyping of grain morphology using digital imaging to describe major kernel dimensions and elliptic fourier descriptots contributing to grain size and weight. A population comprised of 230 D-genome, synthetic hexaploids was genotyped for 1,500 DArT markers, facilitating a genome-wide association of kernel traits and the different kernel dimensions that contribute to size and weight. The population structure analysis and genetic diversity measurements, using DArT markers, indicated that these synthetics are diverse and will broaden the genetic base of crop if exploited for wheat genetic improvement. This technique also narrowed down the D-genome synthetic hexaploids as having the potential to improve grain yield, which will facilitate the targeted exploitation of these synthetics to improve grain yield practices in Pakistan under the umbrella of a wheat wide crosses program.

Comparative assessment of high-molecular-weight glutenin composition and their relationship with grain quality traits in bread wheat and synthetic derivatives.

Ahmad Ali, Muhammad Arshad, Abdul Aziz Napar, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Various synthetic hexaploid wheats (*T. turgidum* subsp. *durum*/*Ae. tauschii*) have resulted in significantly superior combinations for biotic/abiotic resistance/tolerances (Mujeeb-Kazi 2003). At the same time, baking industry exigencies and wide consumer preferences have driven wheat breeders to incorporate grain quality-related traits as an important preference in current research. Hence, the development of wheat cultivars with good bread-making quality is a challenging task in many wheat breeding programs (Oury and Godin 2007), particularly if adaptive to a stress environment. Gluten proteins are the key endosperm components that are mainly emphasized regarding end-use quality traits (Payne et al. 1987). The glutenins are long chains of polypeptides linked by disulfide bonds and comprised of low-molecular-weight (LMW-GS) and high-molecular-weight subunits (HMW-GS) (Payne and Lawrence 1983). The HMW-GSs designated as *Glu-A1*, *Glu-B1*, and *Glu-D1* are encoded by multi-allelic genes located on the long arms of chromosomes 1A, 1B, and 1D, respectively. The HMW-GSs constitute about 10% of the wheat endosperm storage proteins, compared to 40% LMW-GSs, but still have a major influence on the bread-making properties of flour (Payne et al. 1987). This study investigated a collection of drought-tolerant wheat genotypes, comprised of D-genome synthetic hexaploid derivatives and conventional bread wheat germ plasm, for key grain quality parameters and their genetic composition based on the HMW-GS profiles. The experimental germ plasm consisted of a core collection of 55 drought-tolerant wheats comprising three groups: i) D-genome synthetic hexaploid derivatives, ii) conventional bread wheat lines, and iii) elite check cultivars.

The frequency of HMW-GS alleles identified in the germ plasm is presented (Table 5, p. 73). *Glu-1* loci encoded 11 different alleles across the three genomes in these genotypes. At the *Glu-A1* locus, three alleles were observed, of which the Ax null allele was found predominantly in 46 (83.64%) genotypes. At the *Glu-B1* locus, subunit 17+18, encoded by *Glu-B1b*, was found in the most genotypes (54.44%). Similarly, *Glu-D1d*, which encodes the Dx5+Dy10 subunit, was observed in 63.64% of the genotypes, an important good quality subunit encoding a HMW-GS allele. Maximum allelic diversity was found at the *Glu-B1* (0.61) locus, followed by *Glu-D1* (0.50), and *Glu-A1* (0.29), primarily due to the allelic richness (five alleles) observed at the *Glu-B1* locus. However, *Glu-A1* and *Glu-D1* showed the same allelic

Table 5. Allelic variation at the *Glu-1* (high-molecular-weight subunit) loci in a core collection of 55 drought-tolerant wheats comprising three groups: i) D-genome synthetic hexaploid derivatives, ii) conventional bread wheat lines, and iii) elite check cultivars.

Locus	Allele	Subunit	Accessions	Frequency (%)	H (Nei's Index)
Glu-A1	A	1	3	5.45	
	B	2*	6	10.91	
	C	Null	46	83.64	0.29
Glu-B1	A	7	2	3.64	
	B	7 + 8	30	54.55	
	D	6 + 8	3	5.45	
	I	17 + 18	16	29.09	
	F	13 + 16	4	7.27	0.61
Glu-D1	A	2 + 12	17	30.91	
	D	5 + 10	35	63.64	
	Z	3 + 10	3	5.45	0.50

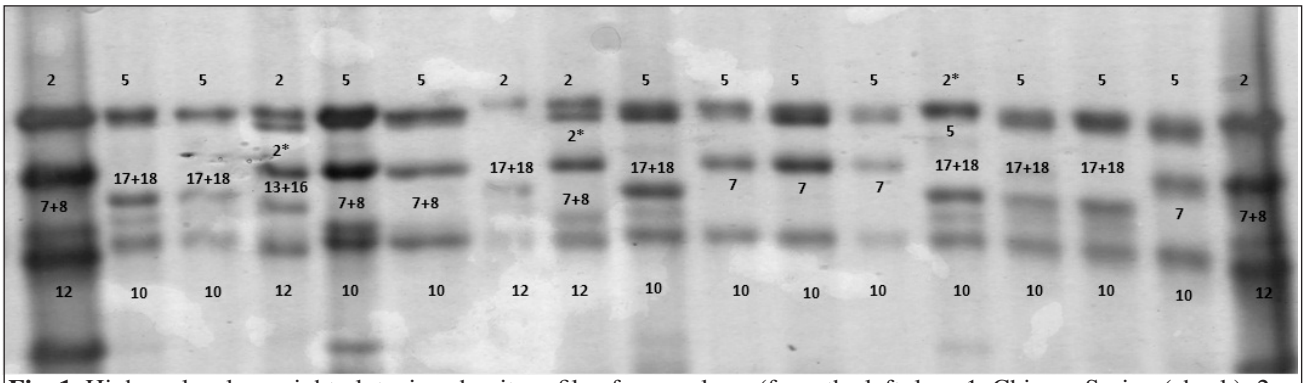

Fig. 1. High-molecular-weight glutenin subunit profile of germ plasm (from the left; lane 1, Chinese Spring (check); 2, AA11; 3, AA15; 4, AA37; 5, AA17, 6, AA18; 7, AA27; 8, AA31; 9, AA38; 10, AA22; 11, AA213; 12, C291 (check); 13, Pavon (check); 14, AA41; 15, AA45; 16, C591 (check); and 17, Chinese Spring (check).

richness (three alleles for both loci), but the distribution of allele frequencies at the *Glu-D1* locus contribute towards more diversity than that of alleles at the *Glu-A1* locus. The HMW-GS observed are presented (Fig. 1).

Results from previous studies on the relationship between glutenin subunits and end-use quality have confirmed that HMW-GS are highly correlated with bread baking quality (Payne et al. 1987). Glutenins and gliadins also were found to influence the bread-making quality (Payne et al. 1987) but with inconsistent results, mostly due to the use of different genetic materials with various genetic backgrounds in different studies. For example, when measuring the effect of 2* subunits on SDS-sedimentation volume, a value equal to 1 was observed from using the British-grown wheat cultivars (Payne et al. 1987), whereas a value greater than 1 was found by Mao et al. (1995) using different wheat cultivars, and a value of less than 1 was obtained by Liu et al. (2005) using 251cultivars and advanced lines.

The key quality parameters studied in this germ plasm include protein contents (%), SDS-sedimentation volume, and carotenoids and their values in individual genotypes are given (Table 6, pp. 74-75). Protein contents ranged from 11.2% to 19.9% with an average of 13.6%. The highest protein content was observed in AA53 (Chakwal-50) which is a rainfed cultivar released in Pakistan. The lowest protein content was found in AA55, which also is cultivated in Pakistan. Eighteen genotypes (32.7%) were found to have more than 14% protein content, which is significantly a promising result. The SDS sedimentation volume in the germ plasm ranged from 2.4 mL to 5.0 mL with an average 3.4 mL. Chakwal-50, which is known to have good glutenin composition and protein contents had SDS-sedimenation value of 5.0 mL. At the same time, other lines, including AA20 and AA48, which are synthetic derivatives, exhibited SDS-sedimenation volume of 4.3 mL and 4.1 mL, respectively. Carotenoids ranged from 4.4 ppm to 9.9 ppm with an average of 6.5 ppm. Results with NIR spectroscopy for grain quality traits of wheat were found promising and economical. Wheat flour SDS sedimentation volume, together with gluten strength, is correlated with dough rheology (Mondal et al. 2009). Because wheat is focused mainly from end-use quality, a better understanding of the genetics underlying specific quality parameters is essential to enhance selection during the breeding process (Carter et al. 2012). The results help us improve under-

Table 6. High-molecular-weight glutenin subunit and quality characteristics of the studied wheat genotypes (SBW = synthetic-derived bread wheat, CBW = conventional bread wheat, and CBW* = check cultivar).

Genotype (AA)	Type	High-molecular-weight glutenin subunit			Protein (%)	Carotenoid (ppm)	SDS-sedimentation (mL)
		Glu-1A	*Glu-1B*	*Glu-1D*			
1	CBW	Null	7+8	2+12	13.3	6.2	3.1
2	CBW	Null	13+16	2+12	13.3	8.1	3.5
3	CBW	Null	7+8	5+10	12.5	6.6	3.1
4	CBW	Null	17+18	5+10	13.2	8.1	3.2
5	SBW	Null	6+8	2+12	13.4	6.5	3.3
6	CBW	Null	7+8	5+10	13.0	8.3	3.3
7	CBW	Null	7+8	5+10	13.9	7.4	3.5
8	CBW	Null	7+8	5+10	12.3	4.5	3.0
9	CBW	Null	17+18	5+10	12.9	5.9	2.4
10	CBW	Null	7+8	2+12	12.0	7.2	2.9
11	CBW	Null	17+18	5+10	12.7	8.4	2.7
12	SBW	Null	13+16	3+10	13.5	9.9	3.6
13	SBW	Null	7+8	3+10	12.0	5.2	3.5
14	SBW	Null	7+8	5+10	12.5	5.5	3.1
15	CBW	Null	17+18	5+10	12.4	8.0	3.2
16	SBW	Null	7+8	2+12	13.7	5.5	3.9
17	SBW	Null	7+8	5+10	13.3	7.2	3.7
18	SBW	Null	7+8	5+10	15.0	8.3	3.4
19	SBW	Null	7+8	5+10	13.4	5.2	3.2
20	SBW	Null	7+8	5+10	14.5	6.1	4.3
21	CBW	Null	7+8	5+10	14.2	8.7	3.3
22	CBW	Null	7	5+10	13.6	8.5	3.4
23	CBW	Null	7	5+10	13.4	7.7	4.1
24	SBW	Null	7+8	2+12	13.7	7.0	3.2
25	CBW	Null	7+8	5+10	13.5	7.0	3.6
26	SBW	Null	6+8	2+12	14.9	5.7	3.6
27	SBW	Null	17+18	2+12	14.3	6.9	3.7
28	SBW	2*	17+18	5+10	15.3	8.2	3.6
29	SBW	Null	7+8	5+10	15.3	5.3	3.6
30	CBW	Null	17+18	2+12	15.3	5.9	3.4
31	SBW	2*	7+8	2+12	13.6	6.9	3.4
32	SBW	1	7+8	2+12	14.4	5.8	3.5
33	SBW	1	6+8	2+12	13.9	6.2	3.3
34	SBW	Null	7+8	5+10	12.7	6.2	2.9
35	CBW	Null	7+8	5+10	13.0	5.3	2.8
36	SBW	Null	7+8	5+10	12.0	6.6	3.3
37	CBW	Null	13+16	2+12	13.3	6.4	3.5
38	CBW	Null	17+18	5+10	12.9	6.5	3.8
39	SBW	2*	17+18	5+10	13.0	6.3	3.5
40	CBW	2*	17+18	2+12	12.7	5.1	3.0
41	SBW	Null	17+18	5+10	12.8	5.1	3.5
42	CBW	Null	7+8	5+10	13.5	6.0	3.0
43	CBW	Null	7+8	5+10	13.8	5.5	3.4
44	SBW	2*	17+18	5+10	14.8	7.7	3.6
45	SBW	Null	17+18	5+10	13.4	6.4	3.6
46	SBW	Null	7+8	2+12	13.8	5.9	3.4
47	SBW	Null	7+8	3+10	14.1	6.0	3.6

Table 6. High-molecular-weight glutenin subunit and quality characteristics of the studied wheat genotypes (SBW = synthetic-derived bread wheat, CBW = conventional bread wheat, and CBW* = check cultivar).

Genotype (AA)	Type	High-molecular-weight glutenin subunit			Protein (%)	Carotenoid (ppm)	SDS-sedimentation (mL)
		Glu-1A	*Glu-1B*	*Glu-1D*			
48	SBW	Null	13+16	2+12	14.6	6.0	4.1
49	CBW	Null	17+18	5+10	15.3	5.9	3.3
50	CBW	Null	7+8	5+10	14.1	7.0	4.1
51	CBW*	Null	17+18	2+12	14.0	6.7	3.5
52	CBW*	Null	7+8	5+10	14.7	6.0	3.2
53	CBW*	Null	7+8	5+10	19.9	5.1	5.0
54	CBW*	Null	17+18	5+10	12.2	4.4	3.0
55	CBW*	2*	7+8	5+10	11.2	5.3	3.2
Average					13.6	6.5	3.4
σ					1.28	1.18	0.41
CV (%)					9.37	18.04	11.89
Maxium					19.9	9.9	5.0
Minimum					11.2	4.4	2.4

standing of the relationships among glutenin compositions and grain quality traits (Tabasum et al. 2011). Quality traits and diversity for glutenin alleles were compared between conventional bread wheat germplasm and D-genome synthetic hexaploid derivatives (Table 7). In synthetic derivatives, more diversity for *Glu-A1* (0.38) and *Glu-D1* (0.59) was observed, whereas no comparison was found for *Glu-B1*. Similarly, protein content (13.8±0.9) and SDS-sedimentation volume (3.5±0.3) were slightly higher in synthetic derivatives compared to bread wheat cultivars. Carotenoids were slightly lower in synthetic derivatives, which is statistically not significant.

Table 7. Comparison of quality traits and glutenin diversity between D-genome synthetic derivatives (SBW) and conventional bread wheat (CBW).

Trait	SBW (n=26)	CBW (n=29)
Protein (%)	13.8 ± 0.90	13.5 ± 1.50
SDS sedimentation	3.5 ± 0.30	3.3 ± 0.40
Carotenoids	6.5 ± 1.10	6.6 ± 1.25
Diversity (H) at:		
Glu-A1	0.38	0.19
Glu-B1	0.59	0.60
Glu-D1	0.59	0.36

Conclusively, this germ plasm set, which is known to have drought-tolerant characteristics, possessed desirable glutenin alleles and grain quality characteristics. The diverse origin of the genotypes in this germ plasm set will enhance the genetic base of breeding programs and contribute to new alleles from D-genome synthetic hexaploids for both drought and grain quality.

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Comparative assessment of low molecular weight glutenin composition and their relationship with grain quality traits in bread wheat and synthetic derivatives.

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Common wheat, being one of the most important food crops worldwide, needs extensive research with major emphasis on yield improvement as well as its adaptation to various biotic and abiotic stresses. Because only a few accessions of the donor species were involved in the evolution of common wheat, genetic diversity was introduced into common wheat by the 'bridge' of a synthetic hexaploid (SH) wheat derived from artificial synthesis of hexaploid wheat (*T. turgidum*/*Ae. tauschii*) in a manner analogous to the evolution of hexaploid wheat (Mujeeb-Kazi et al. 1996). The quality and quantity of wheat gluten proteins give elasticity and extensibility necessary for bread making, contributes about 80–85% of the total flour protein (Shewry et al. 1995), and is comprised of two prolamine groups, gliadins, and glutenin. The glutenins, in turn, are comprised of high-molecular-weight (HMW-GS) and low-molecular-weight (LMW-GS) glutenin subunits. Utilization of wheat is highly dependent on its end-use quality, which, in turn, relies on protein content, carotenoid con-

Table 8. Low-molecular-weight glutenin subunits of wheat the genotypes (SBW = synthetic-derived bread wheat, CBW = conventional bread wheat, and CBW* = check cultivar).

Genotype (AA)	Type	Low-molecular-weight subunit		Genotype (AA)	Type	Low-molecular-weight subunit	
		Glu-A3	*Glu-B3*			*Glu-A3*	*Glu-B3*
1	CBW	b	J	29	SBW	c	f
2	CBW	b	J	30	CBW	f	J
3	CBW	b	g	31	SBW	c	h
4	CBW	d	f	32	SBW	b	e
5	SBW	d	i	33	SBW	b	h
6	CBW	c	h	34	SBW	b	h
7	CBW	f	h	35	CBW	c	h
8	CBW	c	h	36	SBW	b	b
9	CBW	c	i	37	CBW	f	d
10	CBW	f	J	38	CBW	c	e
11	CBW	g	d	39	SBW	b	b
12	SBW	d	i	40	CBW	b	i
13	SBW	b	f	41	SBW	c	h
14	SBW	g	J	42	CBW	b	d
15	CBW	b	d	43	CBW	f	h
16	SBW	c	i	44	SBW	c	J
17	SBW	f	c	45	SBW	c	i
18	SBW	c	h	46	SBW	c	e
19	SBW	c	h	47	SBW	c	h
20	SBW	c	f	48	SBW	b	i
21	CBW	b	J	49	CBW	c	e
22	CBW	a	d	50	CBW	c	j
23	CBW	c	f	51	CBW*	c	g
24	SBW	b	i	52	CBW*	g	g
25	CBW	a	d	53	CBW*	c	c
26	SBW	c	f	54	CBW*	c	c
27	SBW	f	e	55	CBW*	c	g
28	SBW	c	f				

tent, and SDS-sedimentation volume. Recently, Tang et al. (2008, 2010) suggested that grain quality improvement also is possible through the utilization of SHs in breeding programs. Our objective was to investigate a collection of drought-tolerant, wheat genotypes of D-genome SH derivatives and conventional bread wheat germ plasm for key grain quality parameters and their genetic composition based on the LMW-GS profiles.

Results and discussion. Given the discrepancy reported in some studies about the correct identification of the alleles coded at LMW-GS loci using SDS-PAGE (Ikeda et al. 2008), we identified alleles using a PCR-based analysis. In effect, the recent development of allele-specific markers for *Glu-A3* (Wang et al. 2010) and *Glu-B3* (Wang et al. 2009) alleles have profoundly increased the efficiency, accuracy, and reduced the cost for allelic characterization in bread wheat germ plasm. The alleles present, their frequency, and the genetic diversity of alleles at each LMW-GS locus are reported (Tables 8, p. 76, and Table 9). The PCR profiles of LMW-GS alleles are presented (Fig. 2). Six different alleles at *Glu-A3* locus and eight alleles at *Glu-B3* locus were identified by allele-specific markers. At *Glu-A3*, the allele *Glu-A3c* was present in majority of the genotypes (45.45%), whereas *Glu-A3a* was present only in two genotypes (AA39 and AA41). The predominant frequency of *Glu-A3c* has been observed in Indian cultivars (Ram et al. 2011), and other studies also showed its presence in diverse wheat genotypes representing different regions (Wang et al. 2010; Zhang et al. 2004). The frequency of other alleles, *Glu-A3b*, *Glu-A3d*, *Glu-A3f*, and *Glu-A3g*, was found to be 27.27%, 5.45%, 12.73%, and 5.45%, respectively. The major

Table 9. Allelic variation at the *Glu-3* (low-molecular-weight subunit) loci in a core collection of 28 D-genome synthetic derivatives (SBW) and conventional bread wheat (CBW).

Locus	Allele	Accessions	Frequency (%)	H (Nei's Index)
Glu-A3	a	2	3.64	0.70
	b	15	27.27	
	c	25	45.45	
	d	3	5.45	
	f	7	12.73	
	g	3	5.45	
Glu-B3	b	2	3.64	0.87
	d	6	10.91	
	e	5	9.09	
	f	7	12.73	
	g	4	7.27	
	h	12	21.82	
	i	8	14.55	
	j	8	14.55	

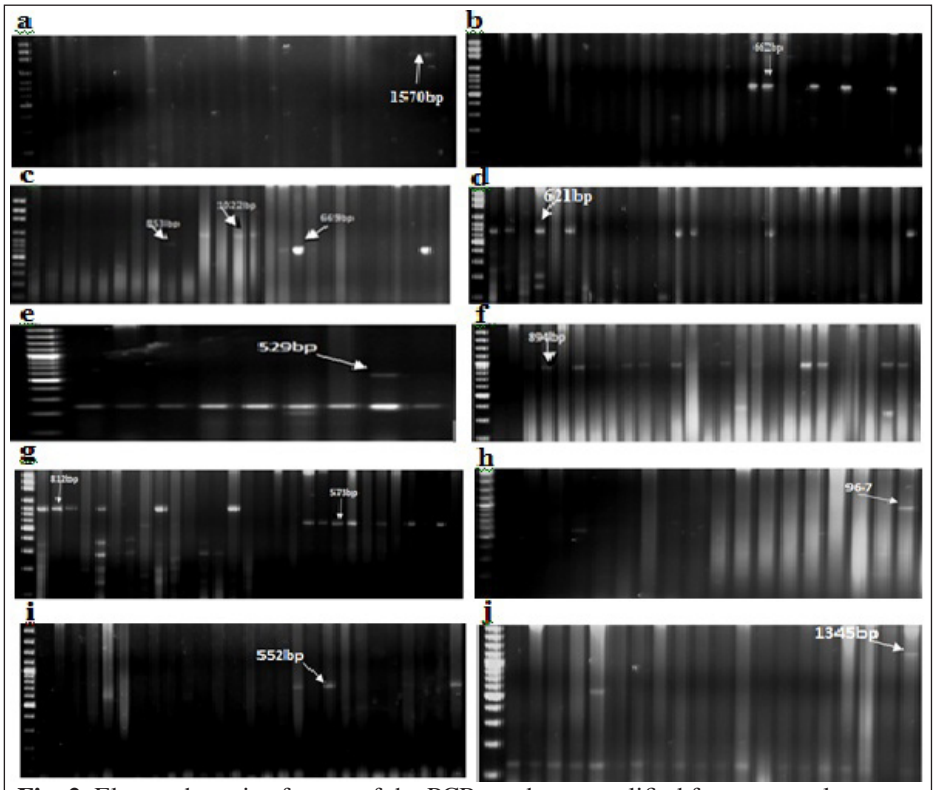

Fig. 2. Electrophoresis of some of the PCR products amplified from germ plasm on agarose gels using allele-specific markers for a, *gluB3b*; b, *gluB3d*; c, *gluB3g gluB3h gluB3e*; d, *gluB3i*; e, *gluA3a*; f, *gluA3b*; g, *gluB3f gluA3c*; h, *gluA3d*; i, *gluA3f*; and j, *gluA3g*. Materials used as PCR templates were (a) B3b- AA36, AA39; (b) B3d-Aa11, AA15, AA22, AA25, AA37; (c) B3g-AA29, B3h-AA18, AA19, AA31, B3e-AA27, AA32, AA46; (d) B3i- AA5, AA9, AA12, AA16, AA24, AA40, AA45, AA48; (e) A3a-AA22; (f) A3b-AA1, AA3, AA13, AA15, AA21, AA32, AA33, AA34, AA36, AA39, AA40, AA42, AA48; (g) B3f-AA4, AA13, AA20, AA26, AA28, AA29; A3c-AA16,.AA18, AA19, AA20, AA26, AA28, AA29, AA31; (h) A3d-AA30; (i) A3f-AA7, AA10, AA27, and (j) A3g-AA11.

allele found at the *Glu-B3* locus was *Glu-B3h*, which appeared in 12 genotypes (21.81%), followed by *Glu-B3i* and *Glu-B3j* (14.54%). Maximum allelic diversity was found at the *Glu-B3* (0.87) locus, followed by *Glu-A3* (0.70), which is primarily due to the allelic richness (eight alleles) observed at the *Glu-B3* locus. Many reports indicate different allelic frequencies representing the *Glu-B3* locus in genotypes from different regions. Among Indian cultivars, the frequency of *Glu-B3b* was the highest (29.3%), followed by *Glu-B3j* (27.1%) and *Glu-B3h* (13.8%).

Results from previous studies on the relationship between glutenin subunits and end-use quality have confirmed that LMW-GS are highly correlated with bread baking quality. Therefore, genotype selection based on HMW-GS and LMW-GS for bread-making quality trait is reliable and considered as a crucial analysis of the germ plasm (Xiyong et al. 2012).

Quality traits and diversity for glutenin alleles were compared between conventional bread wheat germ plasm and D-genome synthetic hexaploid derivatives (Table 10). In the synthetic derivatives, more diversity for *Glu-B1* (0.84) was observed but was equivalent to conventional wheats for *Glu-A3* (0.64). Similarly, protein contents (13.8±0.9) and SDS-sedimentation volume (3.5±0.3) were slightly higher in the synthetic derivatives compared to the bread wheat cultivars. Wheat flour SDS sedimentation volume together with gluten strength is correlated with dough rheology (Mondal et al. 2009).

Table 10. Comparison of quality traits and glutenin diversity between D-genome synthetic derivatives (SBW) and conventional bread wheat (CBW).

Trait	SBW (n=26)	CBW (n=29)
Protein (%)	13.8 ± 0.90	13.5 ± 1.50
SDS sedimentation	3.5 ± 0.30	3.3 ± 0.40
Carotenoids	6.5 ± 1.10	6.6 ± 1.25
Diversity (H) at:		
Glu-A3	0.64	0.64
Glu-B3	0.82	0.84

From the evaluation of ~200 synthetics, the *Ae. tauschii* parent appears to have a much larger influence on the quality of the synthetic hexaploid than the durum parent (Van Ginkel and Ogbonnaya 2007). About 20% of the SHs could be classified as attaining the Australian hard and Australian prime hard quality classification based on examination of the polymeric glutenins proteins (Van Ginkel and Ogbonnaya 2007). This study reports the glutenin diversity and good end-use quality profile of synthetic derivatives, which is the practical example for improving quality traits of bread wheat using SHs in breeding programs. Our results are consistent with those of Lage et al. (2006), who reported significant genetic variation among SHs for protein content and quality, grain weight, and plumpness. In recent studies at CIMMYT, synthetic derivatives carrying excellent bread-making quality can indeed be bred if the common bread wheat parent(s) in the cross has good bread quality traits. HMW- and LMS-GS profiles of the primary SHs (Rasheed et al. 2012) can be used to determine promising crosses and to identify the best quality lines in their progeny.

PCR-based, allele-specific markers were used to identify allelic variation at the *Glu-3* loci (LMW-GS), having a significant effect on visco-elastic properties of wheat dough. Several combinations of favorable LMW-GS alleles were observed at both *Glu-A3* and *Glu-B3* loci. Key quality parameters, such as protein, sedimentation volume and carotenoids, differed significantly within genotypes (Table 10). Higher values for desirable quality traits were found in synthetic derived genotypes as well as in conventional bread wheat varieties. Our results established significant variability in quality characteristics and glutenin composition among D-genome SH wheat derivatives as compared to conventional bread wheat germ plasm, suggesting their use in improving quality traits in bread wheat.

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Effect of pentosans on the peak viscosity and time to reach peak viscosity of wheat flour.

Saqib Arif, Qurrat ul ain Afzal, Mubarik Ahmed, Abid Hasnain, Uzma Sitara, Shazia Arif, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

The influence of added pentosans irrespective of cultivar was insignificant on peak viscosity (Fig. 3). The effects of cultivar differences, water-extractable pentosans (WEP), and water-unextractable pentosans (WUP) on the peak viscosity (PV) of hard white spring wheat (HWSP) flour was determined by an analysis of variance. The results indicate that cultivar differences, WEP, WUP, and the cultivar-by-WEP interaction contribute significantly in the variation of PV of HWSW flours

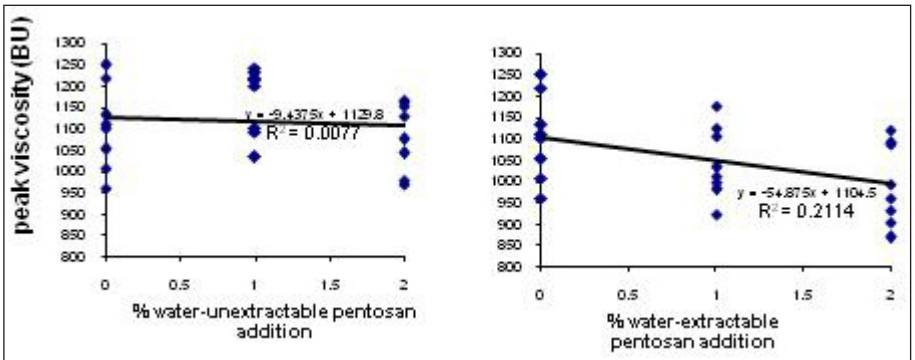

Fig. 3. Effect of pentosans on peak viscosity of wheat flour.

Table 11. Analysis of variance for peak viscosity of eight hard white spring wheat cultivars ($R^2 = 0.695$; adjusted $R^2 = 0.596$).

Source	Type III sum of squares	df	Mean square	F	Significance
Corrected model	1,605,866.975	39	41,176.076	7.009	0.000
Intercept	65,195,962.225	1	65,195,962.225	11097.453	0.000
Cultivar	385,704.775	7	55,100.682	9.379	0.000
Water-unextracted pentosan (WUP)	114,686.333	2	57,343.167	9.761	0.000
Water-extracted pentosan (WEP)	192,679.000	2	96,339.500	16.399	0.000
Cultivar x WUP	136,737.000	14	9,766.929	1.662	0.072
Cultivar x WEP	239,682.333	14	17,120.167	2.914	0.001
Error	704,983.000	120	5,874.858		
Total	189,146,590.000	160			
Corrected total	2310849.975	159			

(Table 11, p. 79). Lin and Czuchajowska (1997) also found a significant effect of cultivar on peak viscosity. Our results reveal that the supplementation level of pentosan influences the PV of wheat flour.

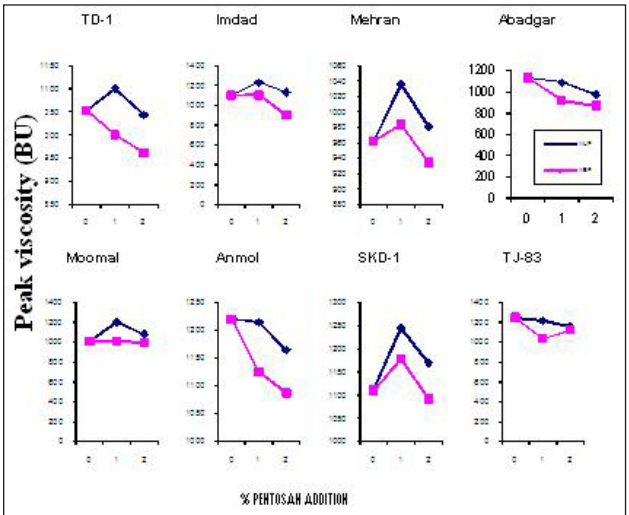

Fig. 4. The effect of water-extractable and water-unextractable pentosans on the peak viscosity of flours from different hard white spring wheat cultivars.

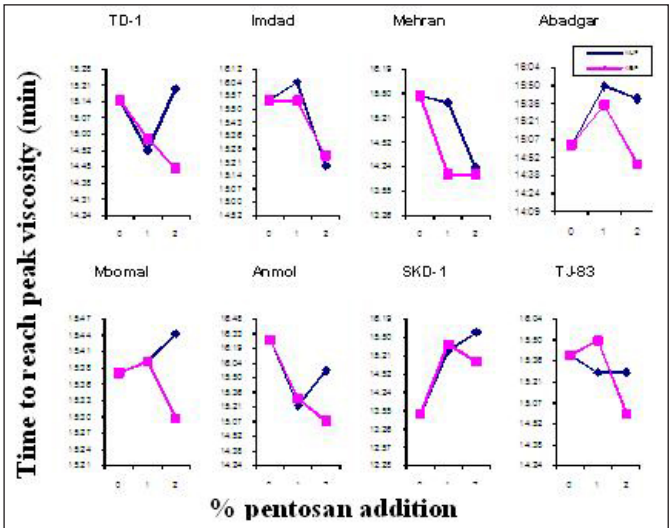

Fig. 5. The effects of water-extractable and water-unextractable pentosans on the time to reach peak viscosity of flours from different hard white spring wheat cultivars.

Because different cultivars were used in this study, we compared the influence of WEP and WUP on the PV of each cultivar at same concentration level (Fig. 4). At a concentration of 1%, both pentosans had the same influence (whether increase or decrease) on all cultivar flours except TD-1, but the magnitude of influence was different for the same cultivar. At a concentration of 2%, WEP had negative impact on all cultivars, whereas the influence of WUP was variable on PV. El-Wakeil et al. (1975) demonstrated a positive influence of pentosan content on amylograph peak viscosity and found greater effect with the addition of water-soluble pentosans than for water-insoluble pentosan.

The analysis of variance to determine the effects of varietal differences, WEP and WUP on time to reach peak viscosity (TPV) of HWSW flour (Table 12). Cultivar differences had highly significant and WEP imparted significant effect on TPV. Whereas, WUP caused negligible variation (F=0.360, P=0.698). 'Cultivar x WUP' and 'cultivar x WEP' interactions contributed significant variation in TPV.

Table 12. Analysis of variance for time to reach peak viscosity of eight hard white spring wheat cultivars ($R^2 = 0.479$; adjusted $R^2 = 0.310$)

Source	Type III sum of squares	df	Mean square	F	Significance
Corrected model	43.387	39	1.112	2.831	0.000
Intercept	13,244.141	1	13244.141	33,705.178	0.000
Cultivar	12.928	7	1.847	4.700	0.000
Water-unextracted pentosan (WUP)	0.283	2	0.142	0.360	0.698
Water-extracted pentosan (WEP)	2.845	2	1.423	3.620	0.030
Cultivar x WUP	18.152	14	1.297	3.300	0.000
Cultivar x WEP	13.006	14	0.929	2.364	0.006
Error	47.153	120	0.393		
Total	37,053.900	160			
Corrected total	90.540	159			

The effect of both pentosans on TPV of each cultivar is shown (Fig. 5). Both pentosans had a different influence on TPV of all cultivar flours, and the influence of both pentosans was found to be inconsistent in terms of type and magnitude.

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The influence of pentosan type on the cold and hot paste viscosity of wheat flour.

Qurrat ul ain Afzal, Saqib Arif, Mubarik Ahmed, Abid Hasnain, Hadi Bux, Abdul Aziz Napar, Alvina Gul Kazi, and Abdul Mujeeb Kazi.

An analysis of variance was used to determine the effects of cultivar differences, water-extractable pentosans (WEP), and water-unextractable pentosans (WUP) on cold-paste viscosity of hard white spring wheat (HWSW) flour (Table 13). Cultivar, WEP, WUP, and the 'cultivar x WEP' interaction significantly influence the CPV of HWSW flours. However, WEP contributed much greater variation in CPV of HWSW flours when compared to cultivar and WUP.

Table 13. Analysis of variance for cold paste viscosity of eight hard white spring wheat cultivars ($R^2 = 0.896$; adjusted $R^2 = 0.863$)

Source	Type III sum of squares	df	Mean square	F	Significance
Corrected model	7,963,089.375	39	204,181.779	26.643	0.000
Intercept	40,010,000.625	1	40,010,000.625	5,220.699	0.000
Cultivar	695,599.975	7	99,371.425	12.966	0.000
Water-unextracted pentosan (WUP)	211,140.333	2	105,570.167	13.775	0.000
Water-extracted pentosan (WEP)	3,284,460.083	2	1,642,230.042	214.286	0.000
Cultivar x WUP	111,446.333	14	7,960.452	1.039	0.420
Cultivar x WEP	290,802.583	14	20,711.613	2.710	0.002
Error	919,647.000	120	7,663.725		
Total	45,394,512.000	160			
Corrected total	8,882,736.375	159			

The effects of WEP and WUP on CPV of each cultivar are shown (Fig. 6). We found that both pentosan fractions exhibited different impacts on CPV. The WEP considerably reduced the CPV of all flours, whereas WUP induced irregular changes in CPV values.

An analysis of variance determined the effects of varietal differences, WEP, and WUP on hot-paste viscosity of HWSW flour (Table 14, p. 82). Cultivar, WEP, WUP, and cultivar-by-WEP interaction significantly influenced the HPV of HWSW flours. However, WEP contributed much greater variation in HPV of HWSW flours when compared to cultivar and WUP.

Both pentosans had different impacts on HPV of same cultivar flours (Fig. 7, p. 82). WEP was found to reduce the HPV of all cultivar flours, whereas WUP induced irregular changes in HPV. The possible reason could be the hydrophilic nature of WEP that makes it capable of facilitating the rupturing of starch granules during isothermal phase.

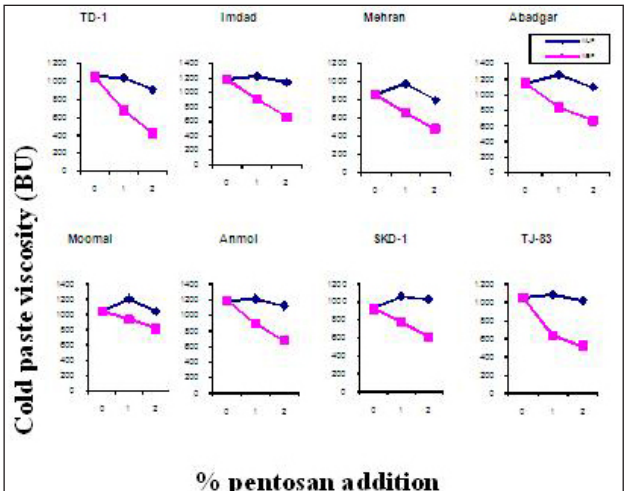

Fig. 6. The effect of water-extractable and water-unextractable pentosans on hot paste viscosity of wheat flours from different hard white spring wheat cultivar flours.

Table 14. Analysis of variance for hot paste viscosity of eight hard white spring wheat cultivars ($R^2 = 0.901$; adjusted $R^2 = 0.869$).

Source	Type III sum of squares	df	Mean square	F	Significance
Corrected model	4,922,296.775	39	126,212.738	28.083	0.000
Intercept	7,998,619.225	1	7,998,619.225	1,779.755	0.000
Cultivar	272,967.375	7	38,995.339	8.677	0.000
Water-unextracted pentosan (WUP)	189,022.583	2	94,511.292	21.029	0.000
Water-extracted pentosan (WEP)	1,993,657.583	2	996,828.792	221.802	0.000
Cultivar x WUP	69,742.083	14	4,981.577	1.108	0.357
Cultivar x WEP	232,213.083	14	16,586.649	3.691	0.000
Error	539,307.000	120	4,494.225		
Total	37,382,786.000	160			
Corrected total	5,461,603.775	159			

On the other side, WUP, being insoluble in water, failed to interact with the aqueous slurry.

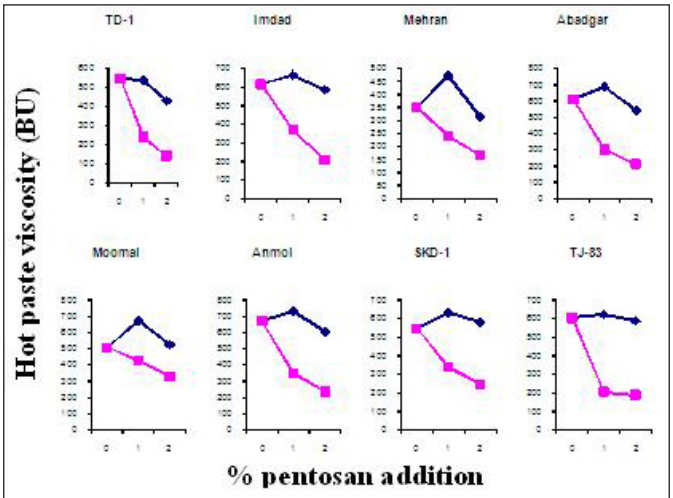

Fig. 7. Effect of water-extractable and water-unextractable pentosans on the cold paste viscosity of different hard white spring wheats cultivar flours.

Influence of water-unextractable pentosan concentration on the pasting temperature and the time required to reach the pasting temperature of wheat flours.

Saqib Arif, Qurrat ul ain Afzal, Mubarik Ahmed, Abid Hasnain, Najmus Sahar, Shazia Arif, Sumaira Farrakh, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Pasting temperature (PT) is one of the most important parameters of wheat flour pasting properties. Moreover, the time required to reach peak viscosity also is important to determine. Many factors, including intrinsic and extrinsic, are known to determine the PT of wheat flour. The addition of pentosan (water unextractable in nature) to the flour may influence the PT of wheat flour. Our study was undertaken by adding water-unextractable pentosan (WUP) at two concentrations (1 % and 2%) to eight different wheat flours in order to examine the shifting of PT at different levels.

Pasting temperature. The effect of WUP at 1% and 2% was variable on the PT of flour. The influence of WUP, in terms of percent change (whether increase or decrease) in PT, from the respective control flour is shown (Fig. 8). Few cultivars showed an increase in PT, whereas others had a lower PT compared to their control flour. The largest reduction in PT was observed in the flours of the cultivar Abadgar, followed by SKD-1, at both 1% and 2% WUP concentration. The reduction is valuable in bread-making, because it implies an earlier start of starch gelatinization and, in turn, an increase in the availability of starch enzyme substrate during the baking period (Rojas et al. 1999). The PT of flour of Moomal remained unchanged in the presence

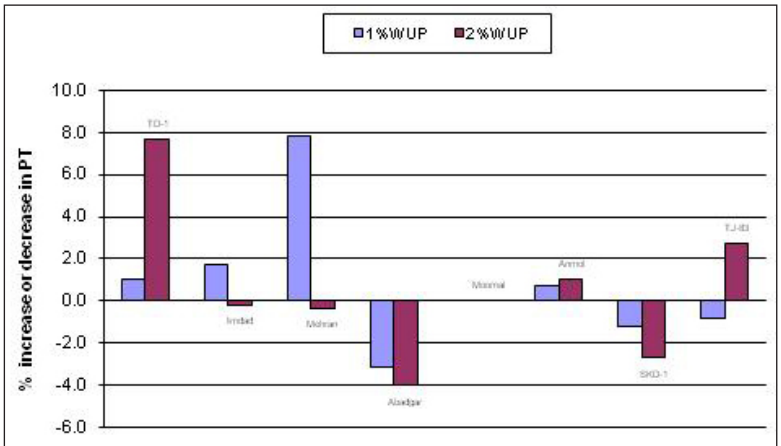

Fig. 8. The magnitude of increase or decrease (%) in pasting temperature of hard white spring wheat flours due to the addition of water-unsoluable pentosan (WUP).

of WUP. The greatest delay in PT was observed in the flour of Mehran at 1% WUP, but a slight decrease in PT was found when the WUP level increased to 2%. The PT of flour of TD-1 also was delayed at both levels of WUP, although the degree was higher at 2%.

Time to reach peak viscosity (TPV). For the same cultivar, both levels of supplementation of WUP imparted a similar type of effect (whether increase or decrease) with the exception of TD-1 and Imdad (Fig. 9). However, the magnitude of influence was different among all cultiarts except TJ-83. The maximum delay was found in TPV of SKD-1 upon addition of both concentration levels, whereas the greatest reduction (85 s) in TPV was found in the flour of Mehran.

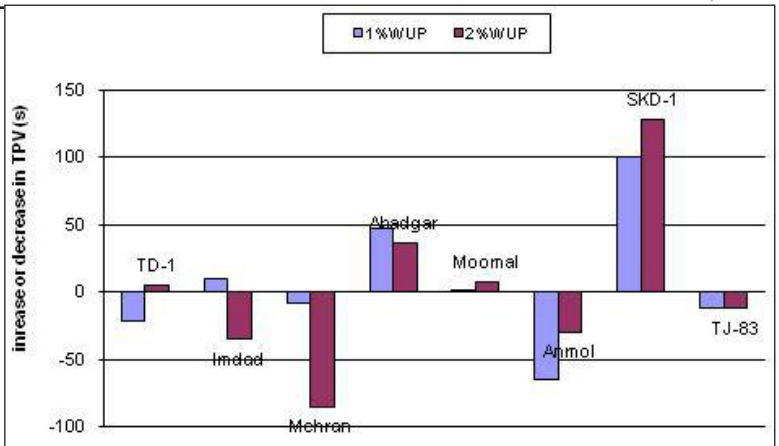

Fig. 9. The increase or decrease in time to reach peak viscosity (TPV) of hard white spring wheat flours due to the addition of water-unextractable pentosan (WUP).

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Effects of water-extractable (WEP) and water-unextractable pentosan (WUP) on the setback viscosity of wheat flour.

Saqib Arif, Qurrat ul ain Afzal, Mubarik Ahmed, Abid Hasnain, Najmus Sahar, Shazia Arif, Sumaira Farrakh, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

An analysis of variance was used to determine the effects of cultivar differences, WEP, and WUP on setback viscosity (SB) of hard white spring wheat (HWSW) flour (Table 15). Cultivar, WEP, and WUP contributed significant variation in SB viscosity, but WEP caused a much greater effect on setback when compared to the cultivar and WUP.

Table 15. Analysis of variance for setback viscosity of eight hard white spring wheat cultivars ($R^2 = 0.843$; adjusted $R^2 = 0.792$)

Source	Type III sum of squares	df	Mean square	F	Significance
Corrected model	1,504,984.000	39	38,589.333	16.503	0.000
Intercept	14,292,202.500	1	14,292,202.500	6,112.263	0.000
Cultivar	79,487.900	7	11,355.414	4.856	0.000
Water-unextracted pentosan (WUP)	63,643.083	2	31,821.542	13.609	0.000
Water-extracted pentosan (WEP)	561,932.583	2	280,966.292	120.159	0.000
Cultivar x WUP	42,667.583	14	3,047.685	1.303	0.215
Cultivar x WEP	36,482.750	14	2,605.911	1.114	0.352
Error	280,594.000	120	2,338.283		
Total	44,056,938.000	160			
Corrected total	1,785,578.000	159			

Both types of pentosan influence setback values of the same cultivar flour differently (Fig. 10, p. 80). WEP reduced the setback viscosity, whereas WUP increased the setback value of flour. The reduction in setback viscosities of flours indicates that WEP reduces the staling tendency of wheat flours. WEP, being hydrophilic in nature, possibly binds to the water molecules, which interacts with solubilized amylose chains to curtail the reassociaion tendency of amylose

chains. Based on the low firmness values, pentosans reduce the starch retrogradation and bread staling (Kim and D'Appolonia 1997a, b; Jankiewicz and Michniewicz 1987). Schiraldi et al. (1996) explored the role of water-soluble pentosans besides other ingredients in bread staling through DSC and confirmed their indirect role anti-staling. Gul and Dizlek (2008) also reported that pentosan decreases the staling rate of bread by retarding the starch retrogradation.

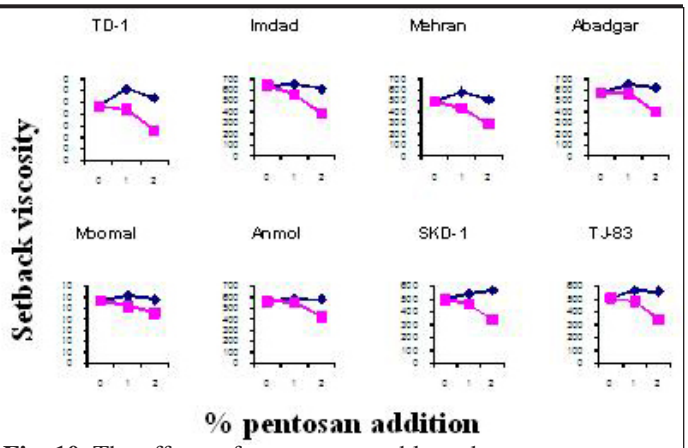

Fig. 10. The effects of water-extractable and water-unextractable pentosans on the setback viscosity of flours of different hard white spring wheat cultivars.

The relationship among pasting parameters in the presence of water-unextractable pentosan.

Saqib Arif, Qurrat ul ain Afzal, Mubarik Ahmed, Abid Hasnain, Shazia Arif, Ahmad Ali, Nosheen Shafqat, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Pasting parameters may relate differently with each other in the presence of added water-unextractable pentosan (WUP) in wheat flour. We checked this hypothesis by determining relationships among pasting parameters of WUP added flours. The relationship of pasting temperature (PT) with other pasting parameters upon the addition of WUP were analyzed by Pearson correlation coefficients (Table 16). The relationship was not in line with that of the control flours. The PT of WUP-substituted flours was weakly related with peak viscosity, time to reach peak viscosity, hot and cold past viscosity, breakdown, and setback of WUP-added flours.

Table 16. Correlation coefficients between pasting temperature and other pasting parameters of water-unextractable pentosan substituted flours.

	Peak viscosity	Time to reach peak viscosity	Hot paste viscosity	Cold paste viscosity	Breakdown viscosity	Setback viscosity
Pasting temperature	–0.115	–0.282	–0.197	–0.306	0.103	–0.163

Peak viscosity of WUP-substituted flours had a significantly positive relationship with hot-paste viscosity (r=0.693**) and cold-paste viscosity (r=0.574**), whereas, peak viscosity did not relate with breakdown (r=0.219) or setback (r=0.102) values. The time to reach peak viscosity of WUP-substituted flours positively related with their hot-paste viscosity (r=0.404*), but did not relate with setback (r=0.214) or breakdown (r=-0.223). A strong relationship was found between cold-paste and host-paste viscosity in the presence of WUP (Table 17). Hot-paste viscosity moderately related with peak viscosity, time to reach peak viscosity, breakdown, and setback.

Table 17. Correlation coefficients between hot paste viscosity and other pasting parameters of water-unextractable pentosan substituted flours.

	Pasting temperature	Peak viscosity	Time to reach peak viscosity	Cold paste viscosity	Breakdown viscosity	Setback viscosity
Hot paste viscosity	–0.197	0.693**	0.404*	0.919**	–0.552**	0.458**

Correlation coefficients between cold-paste viscosity and other pasting parameters in the presence of WUP indicate that cold paste viscosity relates almost in a similar manner in the presence of WEP with as in its absence (Table 18).

Table 18. Correlation coefficients between cold paste viscosity and other pasting parameters of water-unextractable pentosan substituted flours.

	Pasting temperature	Peak viscosity	Time to reach peak viscosity	Hot paste viscosity	Breakdown viscosity	Setback viscosity
Cold paste viscosity	–0.306	0.574**	0.443*	0.919**	–0.580**	0.666**

The relationship between breakdown viscosity and other pasting parameters in the presence of WUP is shown (Table 19). Compared to the control flours, the WUP flours had similar pattern of relationship between breakdown viscosity and other pasting properties.

Table 19. Correlation coefficients between breakdown viscosity and other pasting parameters of water-unextractable pentosan substituted flours.

	Pasting temperature	Peak viscosity	Time to reach peak viscosity	Hot paste viscosity	Cold paste viscosity	Setback viscosity
Breakdown viscosity	0.130	0.219	–0.223	–0.552**	–0.580**	–0.505**

Setback viscosity positively related with cold- and hot-paste viscosity and the time to reach peak viscosity, but was negatively related with breakdown viscosity in WUP-substituted flours (Table 20). Setback viscosity did not relate with pasting temperature or peak viscosity. However, the strength of relationships, as interpreted by their r values, was found to be weaker compared to WEP substituted flours.

Table 20. Correlation coefficients between setback viscosity and other pasting parameters of water-unextractable pentosan substituted flours.

	Pasting temperature	Peak viscosity	Time to reach peak viscosity	Hot paste viscosity	Cold paste viscosity	Breakdown viscosity
Setback viscosity	–0.163	0.102	0.214	0.458**	0.666**	–0.502**

The influence of water-extractable and water-unextractble pentosans on the pasting temperature of wheat flour.

Qurrat ul ain Afzal, Saqib Arif, Mubarik Ahmed, Abid Hasnain, Uzma Sitara, Shazia Arif, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

The influence of added pentosans on pasting temperature (PT) of flours (irrespective of cultivar) is shown (Fig. 11). We found that substituting flours with pentosans up to 2% did not impart any significant change in the PT of wheat flour. The reason for insignificant changes in PT may probably be due to the reason that pentosan interacts with the wheat flour during later stage of heating cycle. The pasting temperature affects the availability of starch granules for amylolytic enzymes. Low pasting temperature is therefore considered desirable for yeast leavened breads (Rojas et al. 1999). Since substitution of pentosans did not affect pasting temperature, it is revealed that the biopolymer can be substituted up to a 2% level without affecting the availability of starch granules during bread making.

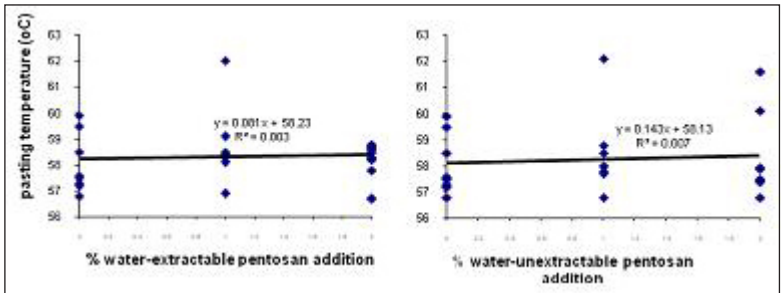

Fig. 11. The effect of pentosan addition on pasting temperature of wheat flour.

An analysis of variance to determined the effects of cultivar differences, water-extractable (WEP) and water-unextractable (WUP) pentosans on the PT of hard white spring wheat (HWSW) flour (Table 21, p. 86). The results indicated that neither WEP nor WUP had significant effect on PT of these flours. However, the significant variation in PT was caused by the cultivar differences and cultivar-by-WEP and cultivar-by-WUP interactions.

We observed slight differences in the effects of both pentosans (both in terms of type and magnitude) for the individual cultivars (Fig. 12, p. 86). We compared the influence (whether increase or decrease) of both types of pentosans at the same level of supplementation on the PT of each cultivar. The trend of both pentosans was similar only in the cultivars Imdad, Abadgar, and Anmol. In the other five, pentosans did not impart consistent impact and varied with the type.

Table 21. Analysis of variance for the effect of water-extractable (WEP) and water-unextractable (WUP) pentosans on the pasting temperature of eight hard white spring wheat cultivars ($R^2 = 0.451$; adjusted $R^2 = 0.272$).

Source	Type III sum of squares	df	Mean square	F	Significance
Corrected model	266.945	39	6.845	2.526	0.000
Intercept	436,388.175	1	436,388.175	161,056.086	0.000
Cultivar	86.823	7	12.403	4.578	0.000
Water-unextracted pentosan (WUP)	2.931	2	1.465	0.541	0.584
Water-extracted pentosan (WEP)	7.833	2	3.916	1.445	0.240
Cultivar x WUP	130.439	14	9.317	3.439	0.000
Cultivar x WEP	67.701	14	4.836	1.785	0.048
Error	325.145	120	2.710		
Total	544,671.040	160			
Corrected total	592.090	159			

Reference.

Rojas JA, Rosell CM, and Bendito de Barber C. 1999. Pasting properties of different wheat flour-hydrocolloidal systems. Food Hydrocolloids 13:27-33.

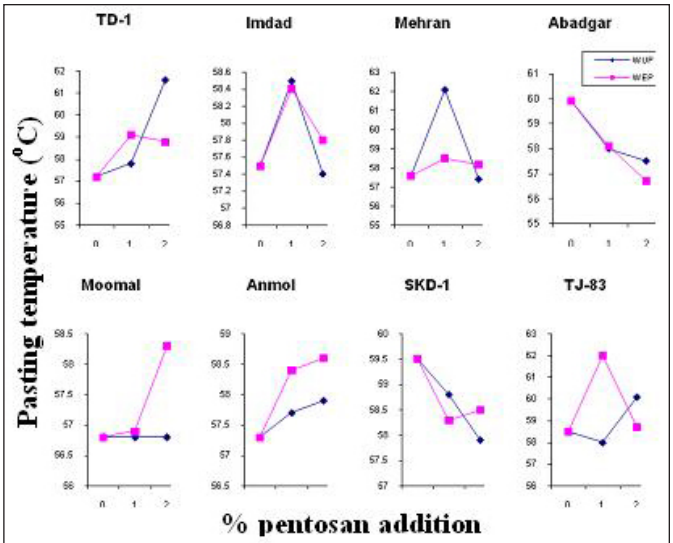

Fig. 12. The effects of water-extractable (WEP) and water-unextractable (WUP) pentosanson pasting temperature of different hard white spring wheat flours.

Comparing the influence of water-extractable pentosan and water-unextractable pentosan on the breakdown viscosity of wheat flour.

Qurrat ul ain Afzal, Saqib Arif, Mubarik Ahmed, Abid Hasnain, Zia ul Hassan, Shazia Arif, Abdul Aziz Napar, Hadi Bux, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Two broad types of pentosans, water-extractable (WEP) and water-unextractable pentosan (WUP), exist in wheat flour. This study was to determine the different role of both pentosans in the breakdown viscosity of wheat flour. For this purpose, a portion of flours was replaced with pentosans at 1% and 2% levels in order to assess the influence of added WUP and WEP on breakdown viscosity of flour. WEP increased the breakdown viscosity of wheat flour (Fig. 13) but WUP did not. Sasaki et al. (2000) studied the influence of nonstarch polysaccharides on gelatinization properties of wheat starch. They found higher breakdown values in nonstarch polysaccharide starch compared to starch alone. The increase in breakdown values was evident with the addition of nonstarch polysaccharides (from rice and ragi) at levels of 0.25 and 0.5% to the wheat flour (Rao et al. 2007).

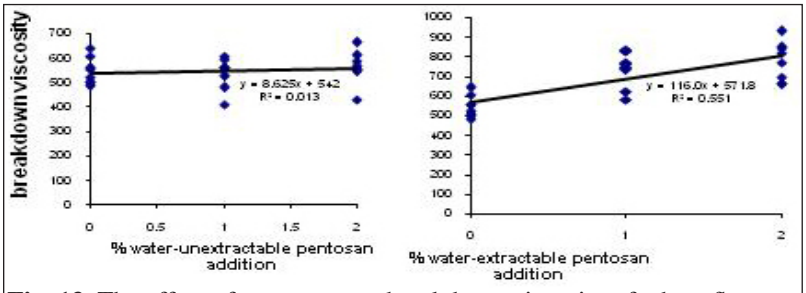

Fig. 13. The effect of pentosans on breakdown viscosity of wheat flour.

An analysis of variance determined the effects of cultivar differences, WEP, and WUP, on breakdown viscosity of hard white spring wheat flour (Table 21, p. 87). The cultivar and WEP contributed significant variation in breakdown viscosity of the flours. WUP and the cultivar-by-WEP, and cultivar-by-WUP interactions did not cause significant variation in breakdown viscosity.

Table 21. Analysis of variance for the effect of water-extractable (WEP) and water-unextractable (WUP) pentosans on the breakdown viscosity of eight hard white spring wheat cultivars ($R^2 = 0.809$; adjusted $R^2 = 0.748$).

Source	Type III sum of squares	df	Mean square	F	Significance
Corrected model	2,528,796.775	39	64,840.943	13.073	0.000
Intercept	27,458,147.025	1	27,458,147.025	5,536.075	0.000
Cultivar	335,671.575	7	47,953.082	9.668	0.000
Water-unextracted pentosan (WUP)	11,753.083	2	5,876.542	1.185	0.309
Water-extracted pentosan (WEP)	972,853.083	2	486,426.542	98.073	0.000
Cultivar x WUP	111,620.250	14	7,972.875	1.607	0.087
Cultivar x WEP	119,498.250	14	8,535.589	1.721	0.060
Error	595,183.000	120	4,959.858		
Total	67,302,602.000	160			
Corrected total	3,123,979.775	159			

The pentosans influenced the breakdown values of each cultivar flour differenctly (Fig. 14). The influence of WEP was consistently positive, whereas WUP was variable depending on the concentration of WUP and genotype of wheat flour. Moreover, the degree of the effect of WEP was found to be greater (up to 63%) than that of WUP.

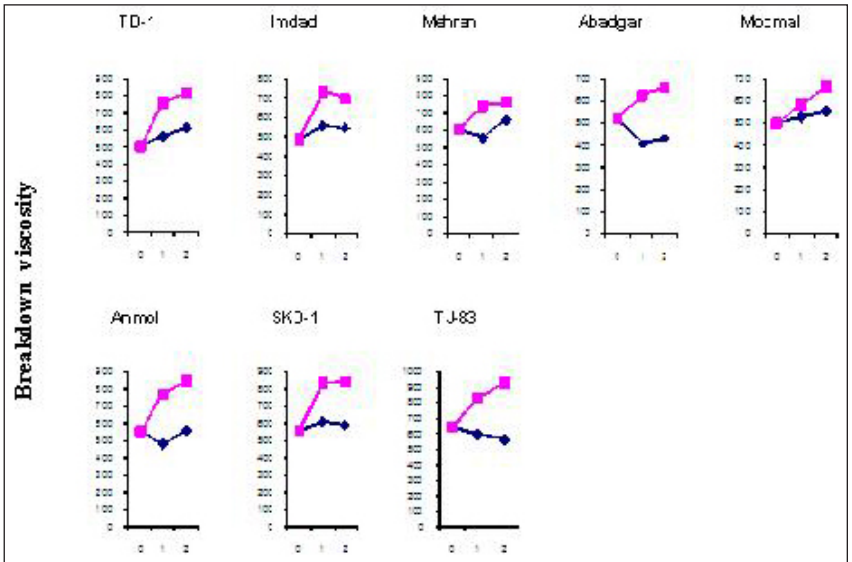

Fig. 14. The effect of water-extractable (WEP) and water-unextractable pentosan (WUP) on the breakdown viscosity of eight different hard white spring wheat cultivar flours.

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Rao RSP, Manohar RS, and Muralikrishna G. 2007. Functional properties of water-Soluble non-starch polysaccharides from rice and ragi: Effect on dough characteristics and baking quality. Food Sci Tech 40(10):1678-1686.

Cultivar and location-wise differences in arabinoxylan levels of wheat.

Qurrat ul ain Afzal, Saqib Arif, Mubarik Ahmed, Abid Hasnain, Akhlaq Ahmed, Shazia Arif, Abdul Aziz Napar, Hadi Bux, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Eight wheat cultivars were analyzed to determine the differences in their arabinoxylan (AX) content. The study also included location-wise differences in total (TOAX) and water-unextractable (WEAX) and water-extractable (WUAX) arabinoxylan content (Fig. 15, p. 88).

Cultivar differences. Duncan's test was used to determine the mean separation of AX content among cultivars and across locations (Fig. 15, p. 88). The cultivar TJ-83 was found to be significantly ($P<0.05$) different in terms of TOAX, WEAX, and WUAX content from all the other cultivars. Higher AX content was found in SKD-1, followed by Anmol, Imdad, and Moomal. The values were insignificant ($P>0.05$) between the cultivars. Variation in WEAX fraction due to the genotype effect has been reported (Hong et al. 1989; Saulnier et al. 2007; Li et al. 2009). Cultivar variability in the total AX fraction also has been reported in durum wheat cultivars (Lempereur et al. 1997; Ciccoritti et al. 2011). The

variability of WEAX contents in durum wheat was found to be higher compared to common wheat cultivars of Australia (Turner et al. 2008). Ramseyer et al. (2011) noted that cultivar influences WUAX content in wheat.

Location-wise differences. The AX content significantly ($P<0.05$) differed across the three cropping locations. The overall performance of cultivars in terms of AX content was found to be better in Hyderabad, followed by Dadu and Nawabshah. Location differences in WEAX content of barley has been reported (Henry 1986; Mikyska et al. 2002; Holtekjølen et al. 2006). The contribution of environmental differences in the variation of WUAX also has been reported for durum wheat (Ciccoritti et al. 2011).

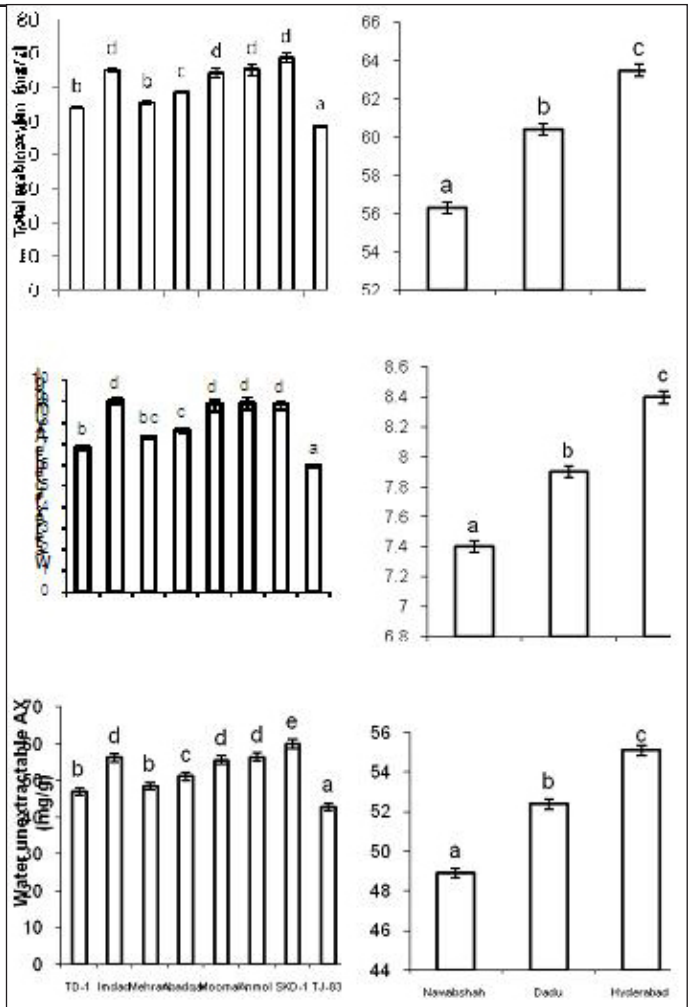

Fig. 15. Cultivar and location-wise differences in arabinoxylan (AX) content in wheat meal.

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Total arabinoxylan levels in whole meal flour of hard wheat cultivars.

Qurrat ul ain Afzal, Saqib Arif, Mubarik Ahmed, Abid Hasnain, Akhlaq Ahmed, Shazia Arif, Sumaira Farrakh, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Arabinoxylans (AX) are the nonstarch polysaccharides present in wheat and are well- known for their impact on processing and product formulation. Eight prevalent Pakistani hard white spring wheat varieties (TJ-83, Abadgar, Anmol, TD-1, Moomal, Imdad, SKD-1, and Mehran) were selected for the study. These cultivars were grown at three different locations in the Sindh province (Nawabshah, Hyderabad, and Dadu) for three consecutive crop years (2005-06, 2006-07, and 2007-08). All wheat samples were ground into whole meal flour (referred to as meal) using a cyclone sample mill (UDY Corp, USA) fitted with a 0.5 mm screen. A modified colorimetric method was used to determine total arabinoxylans (TOAX) content in the meal samples (Table 22).

The TOAX contents irrespective of wheat cultivar, crop year, and location was found to range between 46.3 and 77.8 with an average of 59.7 mg/g. The maximum TOAX content was found in SKD-1 (68.7 mg/g) and the minimum in TJ-83 (48.7 mg/g). Three cultivars, Anmol, Imdad, and Moomal, had a mean TOAX above 60 mg/g. Three other cultivars, TD-1, Mehran, and Abadghar, had a TOAX between 50 and 60 mg/g. TJ-83 was the only cultivar that fell below 50 mg/g. The cultivars grown in Hyderabad expressed the maximum TOAX value (62.4 mg/g), irrespective of the crop year. The average minimum TOAX value in the cultivars grown in Nawabshah was 56.3 mg/g. The variation in TOAX content, in terms of percentage, ranged from -22.44 to + 30.32. The coefficient of variance (CV) among the cultivars was between 9.0% and 16.3% in the same year and between the three crop years. Within the cultivar, there was less variation (CV =2 .7 and 4.6%) in five of the cultivars, whereas three cultivars, Moomal, SKD-1, and Anmol, were less stable across locations and crop years (CV = 10.0-11.8%). The mean CV between the cultivars, irrespective of crop year and location, was 13.2% and 6.4% within the cultiar. Other studies also have found TOAX in wheat meal to range between 40 to 78 mg/g (Saulnier et al. 1995; Lempereur et al. 1997; Wang et al. 2006; Barron et al. 2007).

Table 22. Total arabinoxylan content in meal of Pakistani hard white spring wheat cultivars grown in three locations for three crop years. Values are expressed as mg xylose equivalents/g of sample and were the average of three replications ± SE. Different lowercase letters within the same column are significantly different at $P < 0.05$. Different capital letters within same rows are significantly different at $P < 0.05$.

Location	Year	Cultivar								CV (%)
		TD-1	Imdad	Mehran	Abadgar	Moomal	Anmol	SKD-1	TJ-83	Mean	
Nawabshah	2006	52.4±0.10 aA	63.0±0.12 aB	53.9±0.10 aA	57.5±0.10 aC	56.6±1.19 aC	57.8±0.07 aC	62.7±0.09 aB	47.8±0.17 aD	56.5	9.0
	2007	52.9±0.07 aA	61.8±1.55 aB	53.0±0.53 aA	56.0±1.01 aC	59.3±1.29 aB	58.5±0.01 aC	61.9±0.03 aB	46.3±0.52 aD	56.2	9.5
	2008	52.8±0.12 aA	64.0±0.32 aB	53.5±0.03 aA	56.7±0.07 aC	57.9±0.03 aC	56.9±1.33 aC	61.1±1.22 aB	47.2±0.07 aD	56.3	9.2
Hyderabad	2006	54.5±0.01 bA	67.0±0.16 bB	58.4±0.07 bC	61.7±0.22 bD	66.4±0.24 bB	67.3±1.58 bB	77.8±3.28 bE	50.8±0.10 bF	63.0	13.5
	2007	56.1±1.36 bA	68.9±3.38 bB	57.6±1.34 bA	62.3±1.09 bC	72.4±5.38 cD	75.5±4.45 cD	74.2±0.06 cD	51.6±0.09 bE	64.8	14.1
	2008	55.4±0.07 bA	65.1±0.16 cB	59.4±0.12 bC	60.0±0.24 bC	69.5±0.07 cD	71.4±0.10 dD	70.5±3.13 cD	49.8±0.71 bE	62.6	12.5
Dadu	2006	52.7±0.15 aA	65.0±0.21 cB	53.4±0.21 aA	56.6±0.12 aC	58.0±0.14 aC	67.5±0.54 bB	77.6±0.56 bD	47.4±0.31 aE	59.8	16.3
	2007	52.4±0.22 aA	68.7±0.83 bB	57.7±0.91 bC	57.3±0.28 aC	66.6±0.39 bB	57.9±0.20 aC	70.4±0.82 cB	48.0±0.29 aD	59.9	13.3
	2008	55.1±0.21 bA	63.3±0.49 aB	53.7±0.35 aA	60.3±0.39 bC	72.1±0.80 cD	75.2±0.41 cE	62.2±0.42 aB	49.4±0.62 bF	61.4	14.5
Data for three years across three location	Mean	53.8	65.2	55.6	58.7	64.3	65.3	68.7	48.7		
	CV (%)	2.7	3.9	4.6	4.1	10.0	11.8	10.0	3.7		

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Lempereur I, Rouau X, and Abecassis J. 1997. Genetic and Agronomic variation in arabinoxylan and ferulic acid contents of durum wheat (*Triticum durum* L.) grain and its milling fractions. J Cereal Sci 25:103-110.

Saulnier L, Peneau N, and Thibault JF. 1995. Variability in grain extract viscosity and water-soluble arabinoxylan content in wheat. J Cereal Sci 22(3):259-264.

Wang M, Sapirstein HD, Machet AS, and Dexter JE. 2006. Composition and distribution of pentosans in millstreams of different hard spring wheats. Cereal Chem 83(2):161-168.

Level of water-extractable and unextractable arabinoxylans in whole meal flour of hard wheat cultivars.

Qurrat ul ain Afzal, Saqib Arif, Mubarik Ahmed, Abid Hasnain, Zia ul Hassan, Shazia Arif, Abdul Aziz Napar, Hadi Bux, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Arabinoxylans can be divided into two main groups, water extractable and water unextractable. Each group was analyzed for their content in Pakistani wheat cultivars.

Water-extractable arabinoxylan (WEAX). The WEAX content in meals of different wheat cultivars is given (Table 23). The results more or less reflect the same pattern as that for TOAX content; they have a highly significant correlation (r = 0.929**) with each other. WEAX ranged between 6.8 and 9.0 mg/g (= 7.87 mg/g) irrespective of cultivar, year, or location. The average minimum and maximum WEAX content of 7.4 and 8.3 mg/g were found in Hyderabad and Nawabshah, respectively. The mean CV values between the cultivars in different years at the two locations were found to be 18.0% and 11.4% at Hyderabad and Nawabshah, respectively The average maximum WEAX contents were 8.8–9.0mg/g in cultivars Imdad Anmol, Moomal, and SKD-1. Cultivars Mehran, Abadghar, and TD-1 showed values between 6.8 and 7.6 mg/g, the lowest value of 5.9 mg/g was found in TJ-83. The variation within the cultivar, irrespective of crop year and location, were the least in Imdad (3.0%) and Mehran (4.0%) and the greatest in Anmol (13.4%) and Moomal (12.7%). The mean CV between cultivars was 15.6% and within cultivars was 7.29%. WEAX content ranged from 4.75 to 9.19 mg/g in the meal of 25 hard spring wheat varieties grown under three different environmental conditions in the United States (Li et al. 2009).

Water-unextractable arabinoxylan (WUAX). WUAX is the larger proportion, consisting of about 75% of the total arabinoxylans. Some workers have reported the influence of WUAX on the functional properties of wheat flour. We determined the level of WUAX in wheat flour in various wheat cultivars (Table 24, p. 91).

WUAX constitutes the major portion of TOAX. The WUAX fraction was approximately 7-fold greater than that of the WEAX, which agrees with Finnie et al. (2006). Both of these arabinoxylan fractions showed a highly significant

Table 23. Water-extractable arabinoxylan content in meal of Pakistani hard white spring wheat cultivars grown in three locations for three crop years. Values are expressed as mg xylose equivalents/g of sample and were the average of three replications ± SE. Different lowercase letters within the same column are significantly different at P < 0.05. Different capital letters within same rows are significantly different at P < 0.05.

Location	Year	Cultivar								CV (%)
		TD-1	Imdad	Mehran	Abadgar	Moomal	Anmol	SKD-1	TJ-83	Mean	
Nawabshah	2006	6.4±0.12aA	8.7±0.03aB	7.0±0.16aC	7.7±0.21abD	7.2±0.89aC	7.7±0.03aD	7.9±0.21aD	5.7±0.21aE	7.3	12.8
	2007	6.7±0.27aA	8.5±0.39aB	7.1±0.18aC	7.2±0.65aC	7.9±0.21bD	7.7±0.15aD	8.1±0.03aB	6.5±0.21bA	7.5	9.3
	2008	6.5±0.07aA	8.8±0.19aB	7.0±0.03aC	7.3±0.03aC	7.5±0.03abC	7.8±0.51aBC	8.3±0.51aB	6.1±0.03bA	7.4	12.1
Hyderabad	2006	6.9±0.39aA	9.2±0.03aB	7.6±0.03bC	7.9±0.03bC	9.6±0.15cB	9.1±0.28bB	10.1±0.80bB	5.8±0.03aD	8.3	17.8
	2007	7.6±0.21bA	9.1±0.47aB	7.5±0.18bA	8.0±0.24bC	9.9±1.28cBD	10.7±0.71cD	9.4±0.03cB	6.2±0.09bE	8.6	17.2
	2008	7.4±0.03bA	9.3±0.38aB	7.7±0.19bA	7.8±0.27bA	9.7±0.03cB	9.3±0.03bB	8.8±0.12dC	5.5±0.21aD	8.2	16.9
Dadu	2006	6.6±0.21aA	9.2±0.28aB	6.9±0.12aA	7.4±0.07aC	7.8±0.20bC	9.2±0.21bB	10.0±0.73b	6.0±0.16bD	7.9	18.1
	2007	6.3±0.09aA	9.1±0.47aB	7.4±0.12abC	7.8±0.15bC	9.5±0.16cB	7.8±0.10aC	8.7±0.21dB	5.6±0.16aD	7.8	17.2
	2008	7.2±0.22bA	8.9±0.16aB	7.2±0.16abA	7.6±0.18abA	9.7±0.21cC	10.5±0.53cC	8.3±0.12aB	5.7±0.17aD	8.1	19.0
Data for three years across three location	Mean	6.8	9.0	7.3	7.6	8.8	8.9	8.8	5.9		
	CV (%)	6.8	3.0	4.0	3.7	12.7	13.4	9.2	5.5		

Table 24. Water-unextractable arabinoxylan content in meal of Pakistani hard white spring wheat cultivars grown in three locations for three crop years. Values are expressed as mg xylose equivalents/g of sample and were the average of three replications ± SE. Different lowercase letters within the same column are significantly different at P < 0.05. Different capital letters within same rows are significantly different at P < 0.05.

Location	Year	Cultivar								CV (%)
		TD-1	Imdad	Mehran	Abadgar	Moomal	Anmol	SKD-1	TJ-83	Mean	
Nawabshah	2006	46.0±0.14aA	54.3±0.09aB	46.9±0.12aA	49.8±0.18aC	49.4±0.48aC	50.0±0.03aC	54.8±0.12aB	42.1±0.30aD	49.2	8.6
	2007	46.2±0.21aA	53.3±1.21aB	46.0±0.40aA	48.8±0.50aC	51.4±1.50aD	50.8±0.15aD	53.8±0.15aB	39.8±0.05bE	48.8	9.5
	2008	46.2±0.09aA	55.2±0.41bB	46.4±0.03aA	49.3±0.03aC	50.4±0.05aC	49.1±1.19aC	52.8±0.94aD	41.0±0.03aE	48.8	8.9
Hyderabad	2006	47.6±0.39aA	57.8±0.17cB	50.8±0.06bC	53.7±0.19bD	56.8±0.36bB	58.2±1.30bB	67.7±2.52bE	45.0±0.07cF	54.7	13.0
	2007	48.6±1.21bA	59.7±3.00dB	50.1±1.48bC	54.3±0.91bD	62.5±4.11cE	64.8±3.77cF	64.8±0.03cF	45.4±0.06cG	56.3	13.7
	2008	48.0±0.10abA	55.8±0.33bB	51.7±0.31bC	52.2±0.36bC	59.7±0.03dD	61.5±0.07dE	61.8±3.24cE	44.3±0.64cF	54.4	11.8
Dadu	2006	46.1±0.21aA	55.8±0.27bB	46.5±0.10aA	49.2±0.07aC	50.2±0.30aC	58.3±0.53bD	67.6±0.22bE	41.4±0.24aF	51.9	16.1
	2007	46.1±0.21aA	59.6±0.83dB	50.3±0.98bC	49.6±0.18aC	57.1±0.42bB	50.1±0.10aC	61.8±0.78cB	42.4±0.4aD	52.1	13.0
	2008	47.9±0.43aA	54.4±0.37aB	46.5±0.26aA	52.7±0.28bB	62.4±0.60cC	64.7±0.21cC	53.9±0.33aB	43.7±0.70aD	53.3	13.9
Data for three years across three location	Mean	47	56.2	48.4	51.1	55.5	56.4	59.9	42.8		
	CV (%)	2.2	4.1	4.8	4.2	9.6	11.5	10.2	4.5		

correlation (r=0.899**) with each other. The average maximum and minimum content, irrespective of cultivar, crop year, or location, were 67.7 and 39.8 mg/g, respectively. Like TOAX, the maximum quantity of WEAX was found in cultivars grown in Hyderabad (53.7 mg/g), followed by those grown in Dadu (52.0 mg/g). Five cultivars had a WUAX content less than 50 mg/g. The CV values ranged from 8.6 to 16.1% (=12.05%) between the cultivars and 2.2 to 10.5% (=6.39%) within the cultivars.

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Finnie SM, Bettge AD, and Morris CF. 2006. Influence of cultivar and environment on water-soluble and water-insoluble arabinoxylans in soft wheat. Cereal Chem 83:617-623.
Li S, Morris CF, and Bettge AD. 2009. Genotype and environment variation for arabinoxylans in hard winter and spring wheats of the us pacific northwest. Cereal Chem 86(1):88-95.

The proportion of water-extractable arabinoxylans in total arabinoxylans and the ratio of water-extractable to water-unextractable arabinoxylans in wheat.

Qurrat ul ain Afzal, Saqib Arif, Mubarik Ahmed, Abid Hasnain, Akhlaq Ahmed, Shazia Arif, Hadi Bux, Abdul Aziz Napar, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Water-extractable arabinoxylan (WEAX) is an important proportion of arabinoxylan. Soluble in water, WEAX has tremendous nutritional importance as a dietary fiber and is known to affect the functional properties of wheat flour. The proportion of WEAX in wheat flour is important for determining the behavior of dough during processing and assessing the quality of finished product. However, the proportion of WEAX varies with genotype and location. The variation in the amount of WEAX can be assessed with the values of percentage of WEAX in total arabinoxylan (TOAX) and the ratio of WEAX to WUAX (water-unextractable arabinoxylan). We determined the levels of WEAX in some of the wheat cultivars grown at different locations in southern Pakistan. Eight wheat cultivars grown at three locations for three crop years were tested for their WEAX levels. The tests were performed on wheat meal obtained by grinding wheat grains with a UDY cyclone mill.

The percent WEAX in TOAX and WEAX:WUAX ratio across the locations and crop years is presented (Fig. 16, p. 92). The percent WEAX in TOAX was found to be independent of TOAX; it was weakly related (r=0.384) with the amount of TOAX in the wheat meal. The water-extractable fraction ranged from 12.1 to 13.8%. A narrow range of percent WEAX in TOAX in meals is possibly because all the cultivars under study were hard white spring wheats. Li et al. (2009) found that the percent WEAX content ranged

from 11.7 to 23% in TOAX in both spring and winter wheats. Faurot et al. (1995) found that WEAX, as a fraction of TOAX in wheat, was higher (up to 30%). A higher molecular weight WEAX fraction in TOAX may contribute to the desirable technological properties for dough and bread-making quality characteristics of wheat (Courtin and Delcour 2002).

The WEAX:WUAX ratio (also called the extractability ratio of AX) was narrow, ranging between 0.14 and 0.17 and was the same across all three locations and crop years. We also found that the WEAX:WUAX ratio did not depend on the AX content; the ratio had a weak relationship with TOAX (r=0.390) and WUAX (r=0.326). Others have also reported that the WEAX:WUAX extractability ratio is independent of the TOAX content (Lempereur et al. 1997; Ciccoritti et al. 2011).

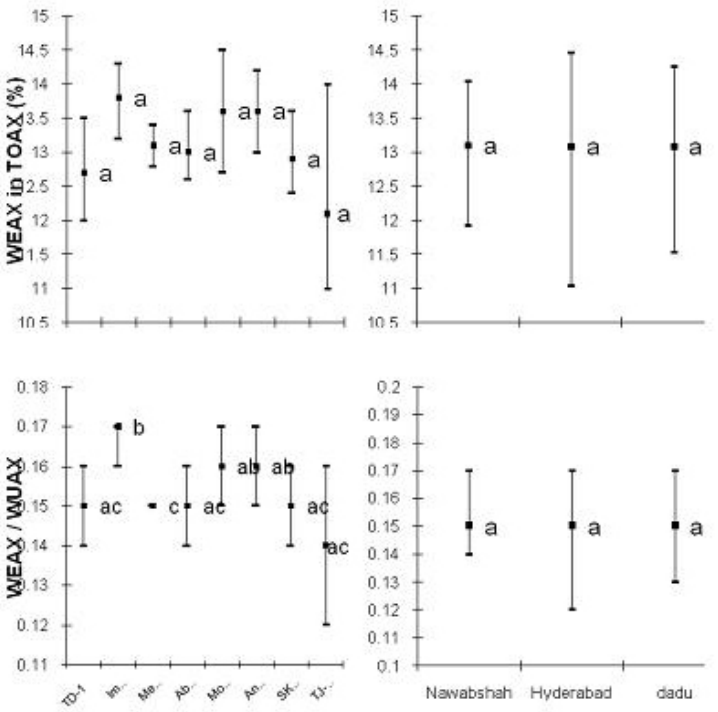

Fig. 16. Minimum, maximum, and mean values of % water-extractable arabinoxylan (WEAX) in total arabinoxylan (TOAX), and WEAX/ WUAX (water-unextractable arabinoxylan) ratio of wheat cultivars.

References.

Ciccoritti R, Scalfati G, Cammerata A, and Sgrulletta D. 2011. Variations in content and extractability of durum wheat (*Triticum turgidum* var. *durum*) arabinoxylans associated with genetic and environmental factors. Internat J Mol Sci 12(7):4536-4549.

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Li S, Morris CF, and Bettge AD. 2009. Genotype and environment variation for arabinoxylans in hard winter and spring wheats of the us pacific northwest. Cereal Chem 86(1):88-95.

Relationship of arabinoxylans with breakdown, setback, and other paste viscosities of wheat flour.

Saqib Arif, Qurrat ul ain Afzal, Mubarik Ahmed, Abid Hasnain, Uzma Sitara, Shazia Arif, Hadi Bux, Sumaira Farrakh, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

The relationship of arabinoxylan (AX) content (naturally present in flour) with the pasting properties of wheat flour was estimated through Pearson's correlation. The AX content, consisting of total arabinoxylan (TOAX), water-extractable arabinoxylan (WEAX), and water-unextractable arabinoxylan (WUAX), were negatively related with breakdown viscosity (BD) (Fig. 17). The percentage of WEAX in TOAX was moderately correlated with BD value.

The relationship of setback viscosity with AX content (naturally present) was esti-

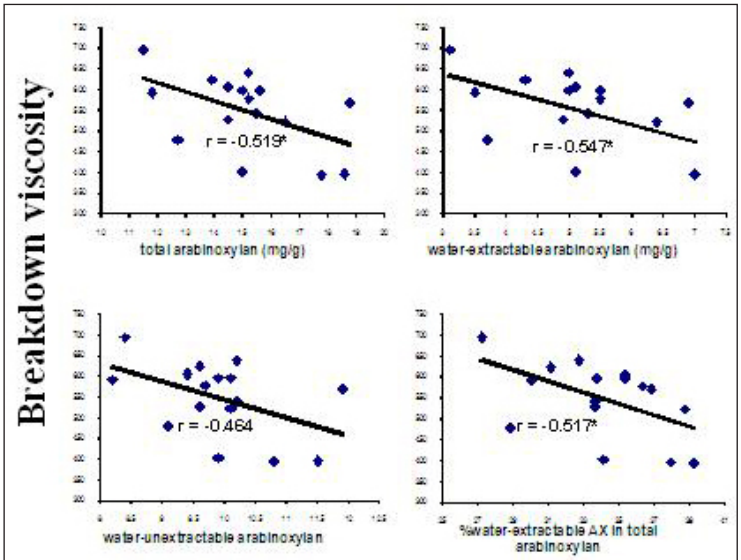


Fig. 17. Relationship of breakdown viscosity with arabinoxylans.

mated. The setback value positively relates with AX content and % WEAX in TOAX (Fig. 18). Flours containing a higher AX content have larger setback values and are more susceptible to bread staling. The flours investigated were extracted from sound wheats, because they had setback values less than 650BU.

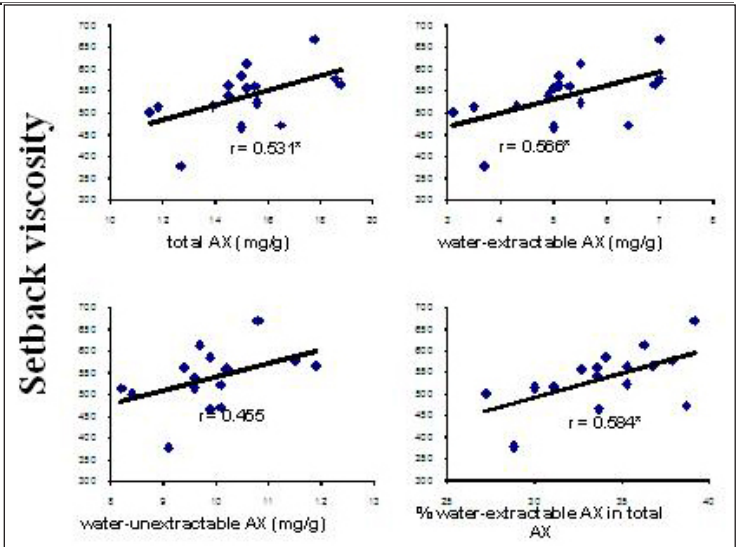


Fig. 18. The relationship between setback viscosity and arabinoxylans (AX).

The AX content naturally present in wheat flour does not relate with HPV. The correlation coefficients of HPV with TOAX, WEAX, WUAX, and %WEAX in TOAX are 0.172, 0.166, and 0.057, respectively, due to the low amount of AX (1.2-1.7%) in wheat flour. The AX content and % WEAX in TOAX weakly relate with peak viscosity (PV) of wheat flour. Iriki et al. (2003) found that AX content significantly correlated with the apparent viscosity of heat-treated flour paste. In our study, the relationship of PV is not statistically significant with TOAX (r=-0.262), WEAX(r=-0.289), WUAX(r=-0.203), or % WEAX in TOAX (r= -0.369).

In order to understand the dependence of time to reach peak viscosity (TPV) on native AX content, the relationship of TTP and AX was determined. TPV did not relate with TOAX (r=0.038), WEAX (r=-0.037), WUAX (r=0.132), or %WEAX in TOAX (r=-0.194).

Reference.

Iriki N, Yamauchi H, Takata K, Nishio Z, Ichinose Y, and Yoshihira T. 2003. Effects of genotype and growth consitions on apparent viscosity of heat-treated flour paste and their correlation with certain flour properties in wheat produced in Hokkaido. Food Sci Technol Res 9(1):104-109.

The impact of weather conditions on arabinoxylan content in wheat.

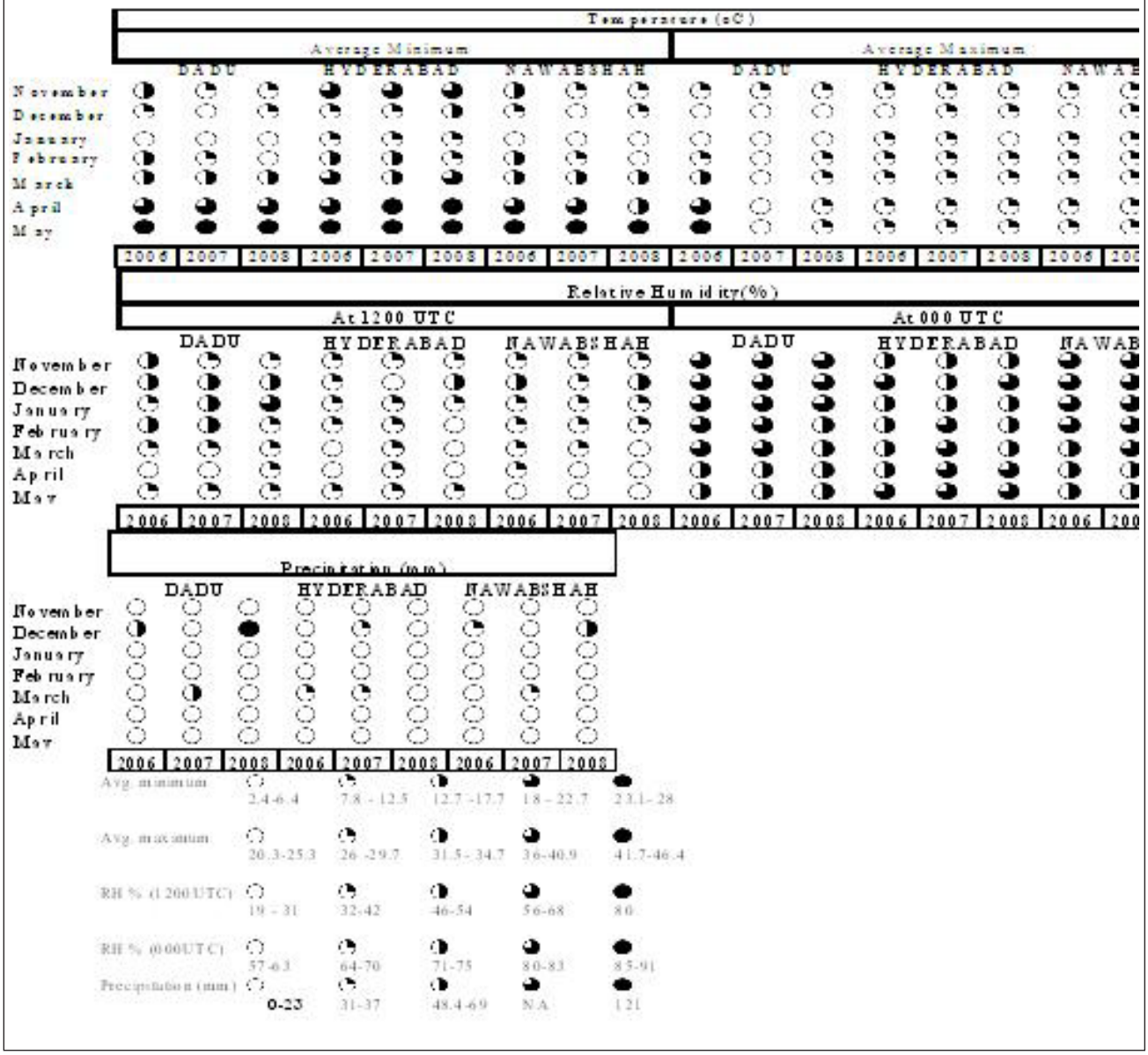
Qurrat ul ain Afzal, Saqib Arif, Mubarik Ahmed, Abid Hasnain, Akhlaq Ahmed, Shazia Arif, Abdul Aziz Napar, Hadi Bux, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

The impact of weather conditions during growth period on arabinoxylan content of wheat was studied. The overall weather in all the crop producing areas was dry and the prime source of water was canal irrigation (Table 25, p. 94). Climatic conditions during the crop year and between heading and harvest were analyzed separately for their correlation with arabinoxylan (AX) fractions (Table 26, p. 95).

Temperature. The minimum temperature across the crop year was significant and positively correlated with AX content. The maximum temperature between heading and harvest showed a negative relationship with AX content. The average temperature recorded throughout the crop year had a moderately positive influence on the amount of AX in wheat meal. Shewry (1999) found that WEAX was negatively correlated with the average temperature during heading to harvest. This relationship was in the flour and bran fractions of winter wheat at heading-to-harvest temperatures between ~10-25oC. In our study, no relationship was found between the average temperature during heading-to-harvest in any of the AX fractions. The differences in findings may be the impact of different temperature values, the nature of samples, and the wheat fraction.

Relative humidity and precipitation. Our investigations relate to wheat cultivation under dry environment with less precipitation. The average precipitation during heading-to-harvest was a moderate negative relationship with % WEAX in TOAX and the WEAX:WUAX ratio. Li et al. (2009) indicated a negative relationship between rainfall and WEAX content and the amount of WEAX in TOAX. Other investigators reported positive relationship of rainfall with TOAX

Table 25. Temperature, relative humidity, and precipitation at the preharvest stage of wheat cultivation in different localities during 2006–08.



(Ciccoritti et al. 2011) and WEAX (Dornez et al. 2008). The amount of the AX fraction had a positive correlation with the average relative humidity during the crop year, but no relative humidity value between heading-to-harvest influenced the AX level. Thus, a humid environment during early growth period of the crop may increase the accumulation of AX in wheat grain.

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Ciccoritti R, Scalfati G, Cammerata A, and Sgrulletta D. 2011. Variations in content and extractability of durum wheat (*Triticum turgidum* L. var. *durum*) arabinoxylans associated with genetic and environmental factors. Internat J Mol Sci 12(7):4536-4549.

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Table 26. Correlation of arabinoxylan parameters with weather conditions during the crop year and during heading-to-harvest (*correlation is significant at the 0.05 level (2-tailed); TOAX = total arabinoxylan; WEAX = water-extractable arabinoxylan; WUAX = water-unextractable arabinoxylan).

	TOAX	WEAX	WUAX	% WEAX in TOAX	WEAX:WUAX ratio
Across crop year					
Temperature maximum	–0.309	–0.281	–0.306	0.063	0.044
Temperature minimum	0.711*	0.684*	0.7178*	0.042	0.016
Temperature mean	0.477	0.465	0.484	0.058	0.030
Precipitation	0.313	0.305	0.309	0.044	0.054
Humidity at 0:00	–0.269	–0.211	–0.275	0.254	0.218
Humidity at 12:00	–0.186	–0.197	–0.186	–0.112	–0.144
Humidity average	0.653	0.679*	0.647	0.336	0.280
Across heading-to-harvest					
Temperature maximum	0.673*	–0.671*	–0.677*	–0.189	–0.145
Temperature minimum	0.426	0.398	0.426	–0.038	0.028
Temperature mean	–0.012	–0.034	–0.014	–0.133	–0.055
Precipitation	–0.008	–0.086	0.008	–0.466	–0.463
Humidity at 0:00	0.077	0.114	0.084	0.245	0.249
Humidity at 12:00	0.164	0.182	0.163	0.154	0.118
Humidity average	0.172	0.200	0.173	0.225	0.194

Effects of cultivar, location, and year on the arabinoxylan content of wheat.

Qurrat ul ain Afzal, Saqib Arif, Mubarik Ahmed, Abid Hasnain, Akhlaq Ahmed, Shazia Arif, Hadi Bux, Abdul Aziz Napar, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

The amount of arabinoxylan in wheat is reported to be influenced by the cultivar and location. However, the extent of influence varied with the change of genotypes and locations and, therefore, it is important to analyze the effects of these factors on the arabinoxylan (AX) content of wheat grown in Pakistan. An analysis of variance (ANOVA) was used to analyze the sources of variation of AX fractions (Table 27, p. 96). The ANOVA model explained 58-96% of the total variation in AX content, %WEAX (water-extractable arabinoxylan) in TOAX (total arabinoxylan) and the WEAX:WUAX (water-unextractable arabinoxylan) (extractability ratio) in wheat meal due to the effects of cultivar, location, harvest year, and their interaction.

TOAX, WEAX, and WUAX varied significantly at $P<0.001$ by the both cultivar and location. The F-value for cultivar was 2-fold and ~1.4-fold greater than that for the location for TOAX and WUAX and WEAX, respectively. Li et al. (2009) reported environmental conditions as the main variance contributor for WEAX. Saulnier et al. (1995) and Finnie et al. (2006) have reported the genotypic variance as the primary source of variation. In our study, TOAX, WEAX, and WUAX content varied insignificantly ($P>0.05$) by harvest year. The effects of 'cultivar × location' and 'cultivar × year' were significant for TOAX, WEAX, and WUAX. Differing results in the variation in crop year can be attributed to the different agronomic practices during the different crop years but further study is required. The interaction of climate with agronomic inputs and soil type dictates variations in AX content (Gebruers et al. 2010). For breeding prospects, the crop year could be an important factor imparting variation in the genotypes. Significant variation was identified in TOAX and WUAX due to 'location × year' interaction, although it was insignificant for WEAX. TOAX, WEAX, and WUAX varied significantly due to the interaction of 'cultivar × location × year', but WEAX variation was significantly lower ($P< 0.01$). Although significant, the effect of 'cultivar × location × year' was found to be lower than the individual effects of cultivar and location. The results are inline with the those of Li et al. (2009).

Table 27. Analysis of variance of arabinoxylan content (TOAX (total arabinoxylan), WEAX (water-extractable arabinoxylan), and WUAX (water-unextractable arabinoxylan)) in the meal of different hard white spring wheat cutivars (F values used type-III mean squares; ns = nonsignificant; * = P<0.05; ** = P<0.01; and *** = P<0.001).

Source	TOAX	WEAX	WUAX	%WEAX in TOAX	WEAX:WUAX ratio
R^2	0.956	0.887	0.955	0.578	0.587
Corrected model *F* value	44.261***	15.923***	43.189***	2.774***	2.881***
Cultivar *F* value	308.958***	118.158***	295.313***	15.239***	16.313***
Location *F* value	221.269***	55.301***	225.980***	0.025 ns	0.055 ns
Year *F* value	1.377ns	0.978ns	1.251ns	0.342 ns	0.571 ns
Cultivar * location *F* value	12.239***	5.828***	11.849***	2.638**	2.789**
Cultivar * year *F* value	10.445***	2.627**	11.049***	1.035 ns	0.955 ns
Location * year *F* value	5.657***	1.997ns	5.779***	0.939 ns	1.109 ns
Cultivar * location * year *F* value	6.940***	2.303**	7.181***	1.229 ns	1.151 ns

Cultivar and the 'cultivar × location' interaction had a significant impact on %WEAX in TOAX and the WEAX:WUAX ratio. Location, 'cultivar × year', and the 'cultivar × location × year' interaction were insignigicant. In contrast, the environmental conditions had a greater influence on the quantity of WEAX in TOAX. A genotypic influence was found for both spring and winter wheat classes (Li et al. 2009). The influence of individual cultivars was much larger than the impact of interaction on AX content.

Our results show that the variance ratios of cultivar-to-location (σ2 cultivar/σ2 location) were 1.9, 4.6, and 1.7 for TOAX, WEAX, and WUAX, respectively. Higher values of 'σ2 cultivar/σ2 location' for WEAX revealed that it remained stable across the locations. The variance ratios of 4.4 for TOAX and 4.9 for WEAX has been reported (Lempereur et al. 1997). Hong et al. (1989) reported 'σ cultivar/ σ location' as 1.6 and 2.4 for WEAX and TOAX, respectively, which shows a greater variance ratio of TOAX than WEAX, in contrast to our results. The 'σ2 cultivar/σ2 location' values for %WEAX in TOAX and the WEAX:WUAX ratio are overestimates because there was insignificant variation at the different locations. Genotypic variance ratios (σ2 cultivar/σ2 culitvar + σ2 location + σ cultivar x location) were 0.66, 0.82, and 0.63 for TOAX, WEAX, and WUAX, respectively.

The percent contributions of variation in AX contents due to cultivar and location are illustrated (Fig. 19). The cultivar is the major variance contributor to TOAX, WEAX, and WUAX content. Percent WEAX in TOAX and the WEAX:WUAX ratio showed approximately 97% variation due to the cultivar effect.

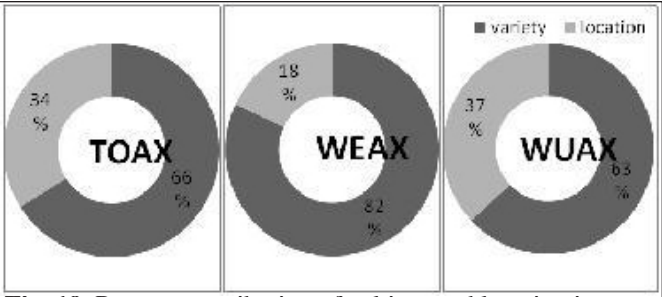

Fig. 19. Percent contribution of cultivar and location in the variability of arabinoxylan (TOAX (total arabinoxylan), WEAX (water-extractable arabinoxylan), and WUAX (water-unextractable arabinoxylan)) content in wheat.

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Saulnier L, Peneau N, and Thibault JF. 1995. Variability in grain extract viscosity and water-soluble arabinoxylan content in wheat. J Cereal Sci 22:259-264.

The effect of water-unextractable pentosans on peak viscosity and time to reach peak viscosity of hard wheat flours.

Saqib Arif, Qurrat ul ain Afzal, Mubarik Ahmed, Abid Hasnain, Shazia Arif, Hadi Bux, Abdul Aziz Napar, Nosheen Shafqat, Ahmad Ali, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Peak viscosity. Peak viscosity (PV) is one of the most important pasting attributes useful for distinguishing starch properties. This study investigates the changes in PV of different wheat flours after the addition of water-unextractable pentosan (WUP) (Fig. 20). Duncan's test was used to further analyze the impact of WUP on the PV of the flour of each cultivar. We found that the changes induced by WUP in PV were statistically insignificant.

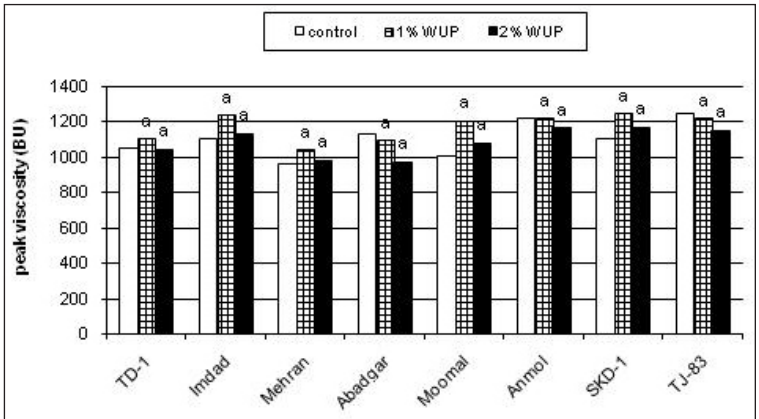

Fig. 20. Effect of water-unextractable pentosan (WUP) on the peak viscosity of flours from different hard white spring wheat cultivars. Bars labeled with an 'a' indicate an insignificant difference from the control at P<0.05.

The PV of 1% WUP-substituted flours varied between 1,036 and 1,245 BU, with the lowest value in the flour of cultivar Mehran and the highest in SKD-1 (Fig. 21). The PV of Mehran was significantly different from those of all the other cultivars except TD-1 and Abadgar. The PV of 2% WUP-substituted flours was between 973 and 1,170 BU. The differences were statistically insignificant among the PV of 2% WUP-substituted flours.

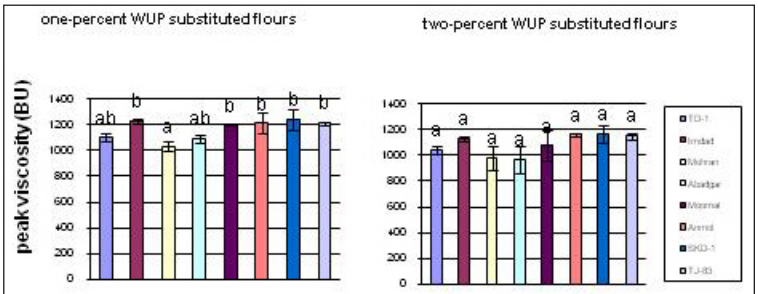

Fig. 21. Peak viscosities of water-unextractable pentosan (WUP)-substituted flours. Bars labeled with different letters are significantly different at P<0.05.

Time to reach peak viscosity. Time to reach peak viscosity (TPV) is an indication of the time required for cooking. Kuo et al. (2001) related the amylographic parameters with the eating quality of rice and found negative correlation between TPV and palatability score. The flour suspension took less time to reach peak viscosity as compared to starch suspension even when both were extracted from the same source of wheat (Mira et al. 2005).

The impact of WUP addition on the time to reach peak viscosity of flours from different wheat cultivars is shown (Fig. 22). WUP imparted insignificant changes in TPV of all cultivar flours. The reason may be due to the lack of interference in the extent of granule swelling. No doubt, WUP competes for water with other constituents of flour, but, possibly due to sufficient availability of water, WUP does not hinder the extent of granule swelling.

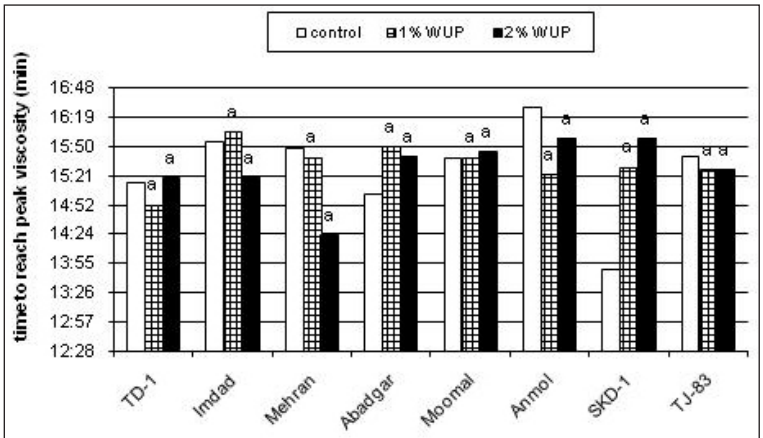

Fig. 22. The effect of water-unextractable pentosan (WUP) on the time to reach peak viscosity of flours from different hard white spring wheat cultivars. Bars labeled with a 'a' indicate an insignificant difference from their control at P<0.05.

Insignificant differences were found among the TPV of 1% and 2% flours (Fig. 23, p. 98). The time taken by 1% WUP-substituted flours to reach their peak viscosity was found in the narrow range of 14:53–16:05 min. Flour of cultivar TD-1 took the lowest time, whereas

more time was needed by the flour of cultivar Imdad to reach its maximum viscosity. With the exception of Mehran, 2% WUP-substituted flours of all cultivars took more than 15 min to reach their maximum viscosity ranging from 15:20 min to 15:58 min. Among the 2% WUP-substituted flours, the shortest time to reach maximum viscosity in all the cultivars was that of the flour of Mehran.

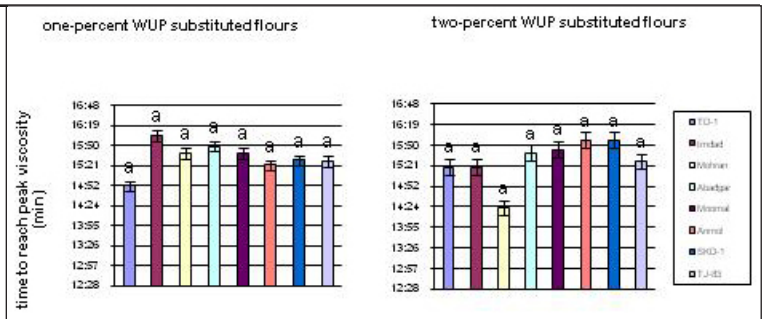

Fig. 23. Time to reach peak viscosity of water-unextractable pentosan (WUP)-substituted flours. Bars labeled with different letters are significantly different at P<0.05

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The effect of water-unextractable pentosans on pasting temperature of hard wheat flours.

Saqib Arif, Qurrat ul ain Afzal, Mubarik Ahmed, Abid Hasnain, Shazia Arif, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Pasting temperature (PT) is defined as the temperature when the first rise in viscosity is recorded by a viscoamylograph. The granules of starch undergo swelling and amylose leaching when the suspension is heated to more than the specific temperature in the presence of excess water.

The temperature needed to begin the pasting process usually depends on the cooking conditions and the type of starch present in the suspension. Added components also may shift the PT of wheat flour, but lacks a comprehensive study. This study will determine the influence of water-unextractable pentosans (WUP) on the PT of wheat flour by of different wheat cultivars.

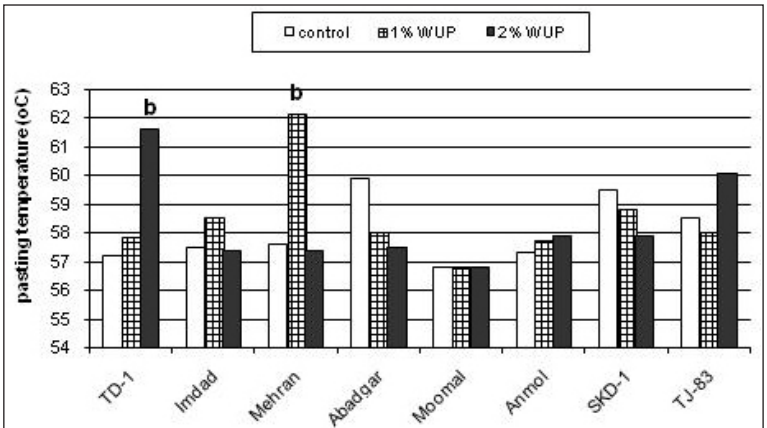

Fig. 24. The eEffect of water-unextractable pentosans (WUP) on the pasting temperature of different cultivar flours. The means of bars labeled with a 'b' differ significantly at P<0.05 from the control.

The effect of WUP on pasting temperature was found to be more inconsistent compared to the effect of water-extractabe pentosan (WEP) and varied from cultivar to cultivar (Fig. 24). WUP delayed the pasting temperatures of flours of cultivars TD-1, Anmol, and TJ-83, whereas the pasting temperature of flours of Imdad, Mehran, Abadgar, and SKD-1 were earlier in the presence of WUP. A lower PT means a faster swelling of granules. No change observed in the pasting temperature of the variety Moomal.

The pasting temperature of 15 1% and 2% WUP-substituted flours of all cultivars (except Mehran) ranged between 56.8 and 58.8°C (Fig. 25). Flour of Mehran exhibited an exceptionally higher (62.1°C) pasting temperature, which was found to be statistically different from all other cultivars except SKD-1. Higher

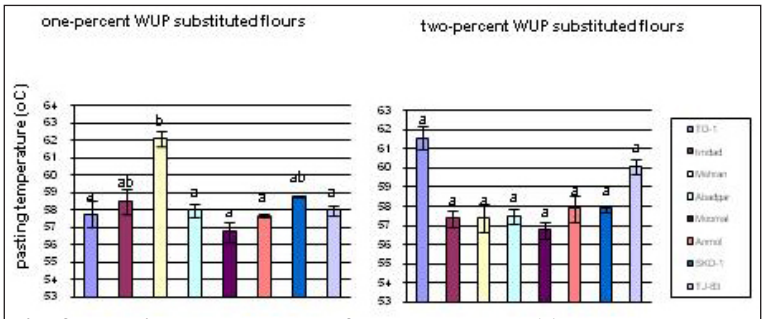

Fig. 25. Pasting temperatures of water-unextractable pentosans (WUP) added to the flours of hard white spring wheat cultivars. Bars labeled with different letters are significantly different at P<0.05.

temperature would be due to the presence of 1% WUP, which delays the swelling and amylose leaching of flour of in Mehran; a phenomenon that can not be generalized over all cultivars. For 2% WUP-substituted flours, the temperature needed to gelatinize starch granules was in a narrow range, 56.8–57.9°C for all cultivars except TJ-83 and TD-1. Flours of TJ-83 and TD-1 had PTs higher than that of all 25 WUP-substituted flours and was greater than 60°C. However, the differences in the 2% WUP-substituted flours were found to be statistically insignificant.

The effect of water-unextractable pentosans on hot paste and cold paste viscosities of hard wheat.

Saqib Arif, Qurrat ul ain Afzal, Mubarik Ahmed, Abid Hasnain, Shazia Arif, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Hot paste viscosity. Hot paste viscosity (HPV) is defined as the viscosity measured after a holding period of 10 mins at 95°C. A wheat starch slurry held at 95°C leads to a reduction in the pasting viscosity of the wheat flour slurry as it is subjected to mechanical stress. This isothermal phase eventually results in the rupture of swollen starch granules responsible for viscosity development. Our study shows the influence of water-unextractable pentosan (WUP) addition on the hot paste viscosity of flours of different hard white spring wheat cultivars (Fig. 26).

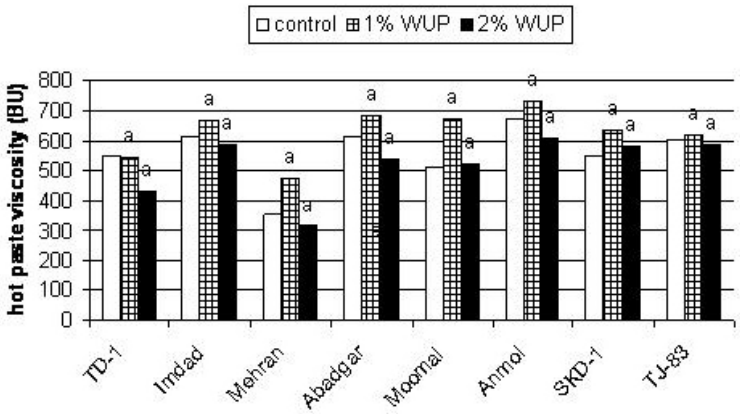

Fig 26. The effect of water-unextractable pentosans (WUP) on the hot paste viscosity (HPV) of flours from different hard white spring wheat cultivars. Bars labeled with an 'a' indicate an insignificant difference from control at $P<0.05$.

WUP did not induce significant changes in HPV. Moreover, the changes were inconsistent and varied with the cultivar and WUP concentration. The variable effect of WUP might be due to the insoluble nature of WUP even though WUP is expected to compete for water.

The hot paste viscosities of WUP substituted flours of different wheat varieties are shown (Fig. 27). Among the 1% flours, the least viscous hot paste was the flour of cultivar Mehran; the most viscous hot paste was that of Anmol. With the exception of Mehran and Anmol, the hot paste viscosities of flours of all cultivars ranged between 542 and 685 BU. Insignificant differences were found between the HPV of 2% WUP-substituted flours. The HPV of Mehran exhibited the lowest viscosity (315 BU) and the most viscous (606 BU) was the flour of Anmol. Hot paste viscosities of 2% WUP-substituted flours of all other cultivars (except Mehran and Anmol) varied between 431 and 587 BU.

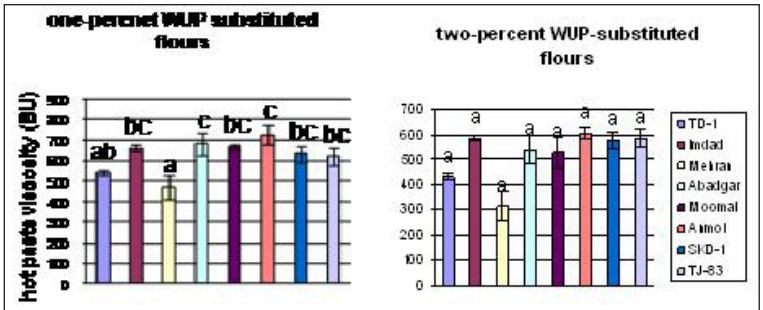

Fig 27. Hot paste viscosities of water-unextractable pentosans (WUP)-substituted flours of different hard white spring wheats.

Cold paste viscosity. Cold paste viscosity (CPV) is the viscosity measured after holding the slurry at 50°C for 10 min. The value of CPV serves as an indicator of paste stability after cooking. The paste- or gel-forming ability of starch after cooling corresponded well with CPV values (Shimelis et al. 2006). Moreover, CPV is a significant attribute in some food processing operations, such as canning (Beta et al. 2001), and predicts the starch property in the preparation of food items (such as instant soup, creams, and sauces) that require cold thickening capacity (Alves et al. 1999).

Duncan's test was used to determine the effect of WUP in the CPV of each cultivar (Fig. 28, p. 100). We found that within a cultivar, the addition of WUP to wheat flour did not make significant changes in CPV of all cultivar

flours. WUP also made irregular changes in CPV and varied from cultivar to cultivar. The insignificant impact of WUP may reflect the less interference of WUP in reassociation of amylase molecules upon cooling after cooking. The WUP added flours can be used in the formulation of instant soups, creams etc without significantly affecting their cold thickening capacity.

Cold paste viscosities of 1% WUP-substituted flours ranged between 982 and 126 1BU (Fig. 28). Flours of cultivars TD-1, Mehran, SKD-1, and TJ-83 exhibited cold paste viscosities less than 1,100 BU. Cold paste viscosities of flours of Imdad, Abadgar, Moomal, and Anmol were greater than 1,200 BU. Cold paste viscosities of 2% WUP-substituted flours of all other cultivars varied between 916 and 1,124 BU (Fig. 28). Similar to hot paste viscosity, the cold paste viscosity of Mehran was the lowest viscosity. The most viscous cold paste was found to be the flour of Imdad (Fig. 29).

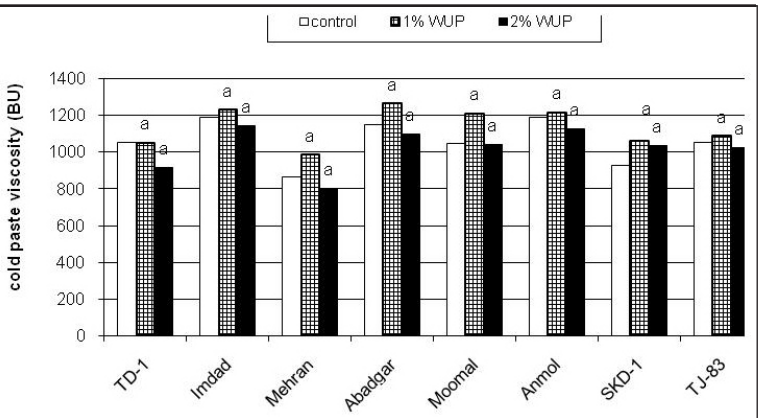

Fig. 28. The effect of water-unextractable pentosans (WUP) on the cold paste viscosity of flours from different hard white spring wheat cultivars. Bars labeled with an 'a' indicate a insignificant difference from the control at P<0.05.

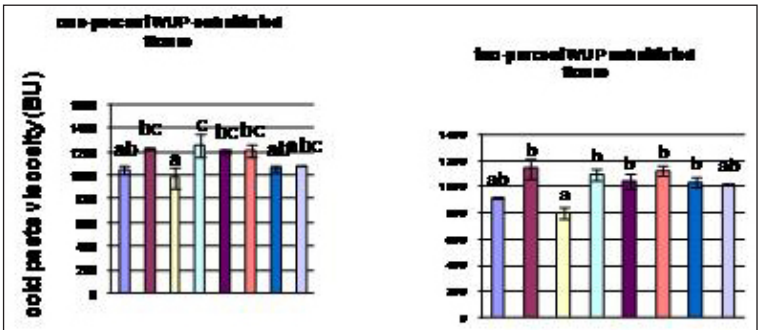

Fig 29. Cold paste viscosities of water-unextractable pentosan (WUP)-substituted hard white spring wheat flours. Bars labeled with the same letters are not significantly different at P<0.05.

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Effects of water-unextractable pentosans on breakdown and setback viscosities of hard wheat flours.

Saqib Arif, Qurrat ul ain Afzal, Mubarik Ahmed, Abid Hasnain, Shazia Arif, Ahmad Ali, Nosheen Shafqat, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

Breakdown viscosity. Breakdown viscosity (BD) is the measure of fragility of starch granules. Higher breakdown viscosity indicates less tendency of starch granules to resist shear. The present findings depict the influence of water-unextractable pentosan (WUP) addition on the BD of wheat flour (Fig. 30). The addition of WUP to wheat flour induced insignificant change in BD values of all cultivar flours, suggesting that WUP does not influence the

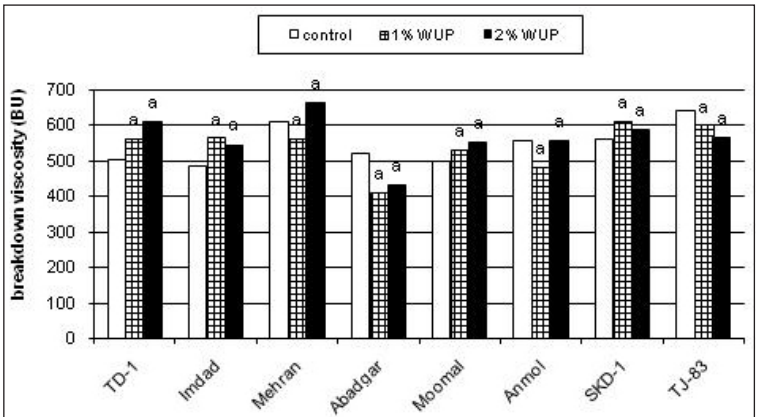

Fig. 30. Effect of water-unextractable pentosans (WUP) on the breakdown viscosity of flours from different hard wheat cultivars. Bars labeled with an 'a' indicate a significant difference from the control at P<0.05.

tendency of starch granules against shearing probably due to the insoluble nature of WUP. Moreover, the effect of WUP was irregular on BD values depending on the genotype of wheat flour and concentration of WUP.

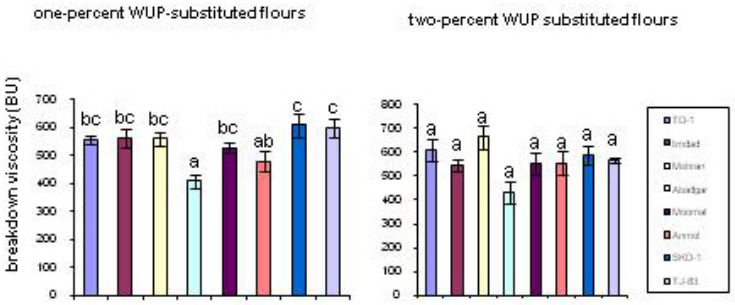

Fig. 31. Breakdown viscosities of water-unextractable pentosan (WUP)-substituted flours. Bars labeled with same letters are not significantly different at P<0.05.

Comparing the BD viscosities of 1% and 2% WUP-substituted flours. One-percent WUP, when added to the flour of cultivar Abadgar, was the most resistant to shearing as interpreted by the lowest BD values (Fig. 31). Pastes of all other cultivars exhibited BD viscosities ranging from 481 to 609 BU. The least tendency against shearing was found in the flour of variety SKD-1. Flour of the cultivar Mehran showed less tendency against shear. The most resistant flour was found to be of the cultivar Abadgar. The BD values of 2% WUP added flour of all cultivars varied insignificantly between 433 and 612BU.

Setback viscosity. Setback viscosity (SB) is the measure of retrogradation tendency of starch granules (Abd Karim et al. 2000). After gelatinization, the leached out linear amylose chains start reassociating on cooling, which subsequently results in increased the viscosity of flour pastes. Retrogradation of starch is a good indicator of bread staling. Other flour components such as gluten, lipids, and pentosans also may be involved in the process of staling (Martin et al. 1991; Shelton and D'Appolonia 1985).

Within cultivars, WUP insignificantly increased the setback viscosities of all cultivars except Imdad (Fig. 32). Only a 5% reduction in setback value was found at a 2% WUP substitution level in the cultivar Imdad. WUP possibly facilitated the reassociation of amylase chains upon cooling after gelatinization that subsequently caused slight increase in the viscosity of flour pastes. WUP itself might increase in viscosity upon cooling after cooking. The SB viscosity of wheat flour becomes greater when the nonstarch polysaccharides from rice and ragi are added up to the level of 0.5% (Rao et al. 2007).

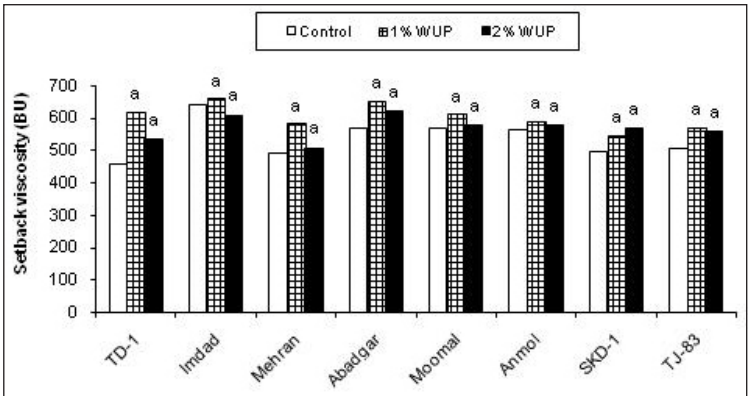

Fig. 32. The effect of water-unextractable pentosans (WUP) on the setback viscosity of flours from different wheat cultivars. Bars labeled with an 'a' indicate a significant difference from the control at P<0.05.

Setback viscosities of 15 WUP-substituted flours of all cultivars was between 543 and 657 BU (Fig. 33). The lowest retrograded valus were in the flour of SKD-1 and the highest was in Imdad. Setback viscosities of 2% WUP-substituted flours varied insignificantly, between 509 and 622 BU. The most retrograded flours was that of Abadgar and least in Mehran.

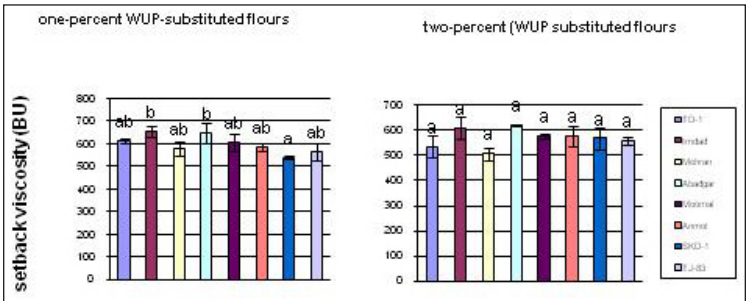

Fig. 33. Setback viscosities of water-unextractable pentosan (WUP)-substituted flours of different hard white spring wheat cultivars. Bars labeled with same letters are not significantly different at P<0.05

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The influence of water-unextractable pentosan concentration on paste viscosities of flours from different wheat cultivars.

Saqib Arif, Qurrat ul ain Afzal, Mubarik Ahmed, Abid Hasnain, Najmus Sahar, Shazia Arif, Sumaira Farrakh, Alvina Gul Kazi, and Abdul Mujeeb-Kazi.

The study was designed to determine the influence of water-unextractable pentosan (WUP) addition on the paste viscosities of flours of different wheat cultivars.

Peak viscosity. Changes were inconsistent at both levels of WUP supplementation and different types of effects (increasing or decreasing) were found on peak viscosity (PV) with increasing concentrations of WUP (Fig. 34). In addition to the type of WUP effect on PV, the degree of the effect varied widely among the cultivars. The maximum decrease in PV was in the cultivar TJ-83 (17.3% decrease) , followed by Abadgar (14.2% decrease). the maximum increase was in the PV of Moomal (18.9% increase), followed by SKD-1 (12.2% increase).

Hot paste viscosity (HPV). The effect of WUP was the opposite at 1% and 2% supplementation levels in all cultivars except TD-1, Moomal, and SKD-1 (Fig. 35). In general, WUP increased HPV at a 1% concentration, whereas HPV was reduced at a 2% concentration. The variability in magnitude of the WUP effect is reflected (Fig. 35). At a 15 concentration, the increase in HPV (3–35%) was found in all cultivars except TD-1 (only 1% decrease). The maximum increase was exhibited in Mehran. A reduction in HPV was found at a 2% concentration level in all cultivars except Moomal (3% increase) and SKD-1 (6% increase). The maximum reduction was found in TD-1 (22%). When the concentration of WUP increased, the amount of WUP component was enough to impart a similar impact as did the components of water-extractable pentosan. However, the mode of action might not be same. WUP possibly facilitates the process of rupturing of swollen starch granules without direct interaction with starch molecules.

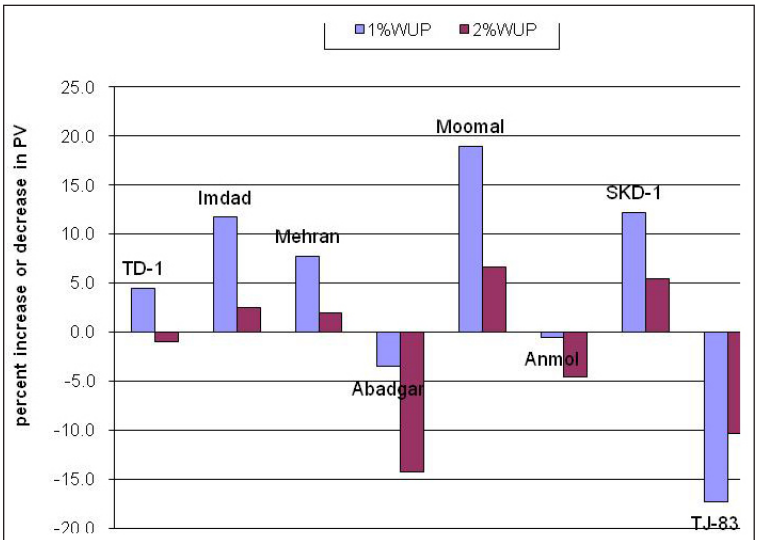

Fig. 34. The magnitude of increase or decrease (in %) in the peak viscosity (PV) of hard white spring wheat flours due to the addition of water-unextractable pentosan (WUP).

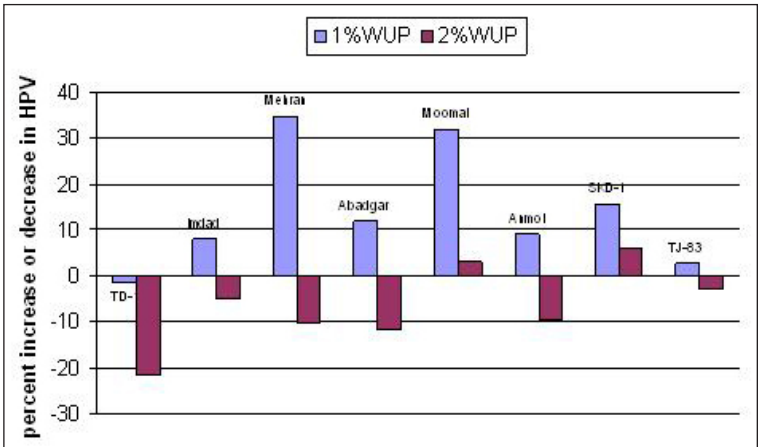

Fig. 35. The increase or decrease in hot paste viscosity (HPV) due to the addition of water-unextractable pentosan (WUP).

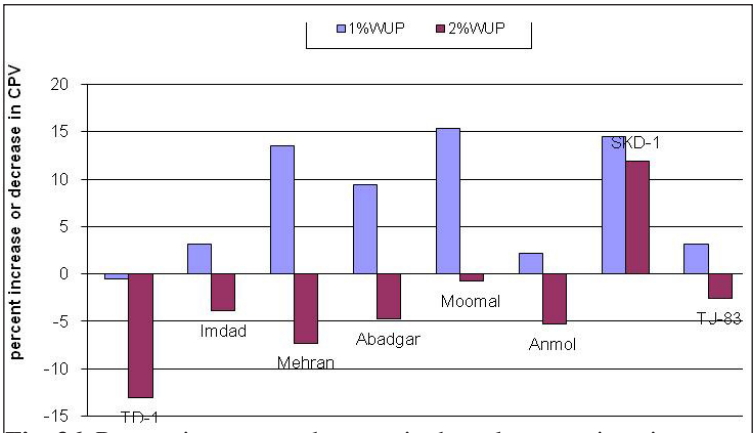

Fig. 36. Percent increase or decrease in the cole paste viscosity (CPV) with the addition of water-unextractable pentosan (WUP).

Cold paste viscosity (CPV). The effect of WUP was found variable on CPV of different varieties depending largely on the concentration of WUP (Fig. 36, p. 102). At the 1% and 2% concentration levels, the effect of WUP was the opposite on CPV of all cultivar flours except SKD-1 and TD-1. At a 1% concentration, the CPV of all cultivar flours except TD-1 increased (2–15%), whereas the CPV decreased between 1% and 13% with the addition of 2% WUP in all cultivar flours except SKD-1.

Breakdown viscosity (BD). The supplementation level of WUP did not induce similar changes in BD of all cultivar flours (Fig. 37). Within cultivar, the same type of influence (increase or decrease) at both concentration levels and a similar type of WUP influence (increase or decrease) were found for each cultivar except Mehran. However, the magnitude of effect was variable at different WUP concentrations.

Setback viscosity (SB). Setback value did not linearly increase with increasing concentration of WUP (Fig. 38). The increase in SB values was variable at both 1% and 2% concentrations depending on the genotype of wheat flour. In fact, higher increases were noticed at 1% substitution in SB values of almost all cultivars, ranging between 2% and 34%. At a 2% concentration, the increase in setback value reached up to 17%.

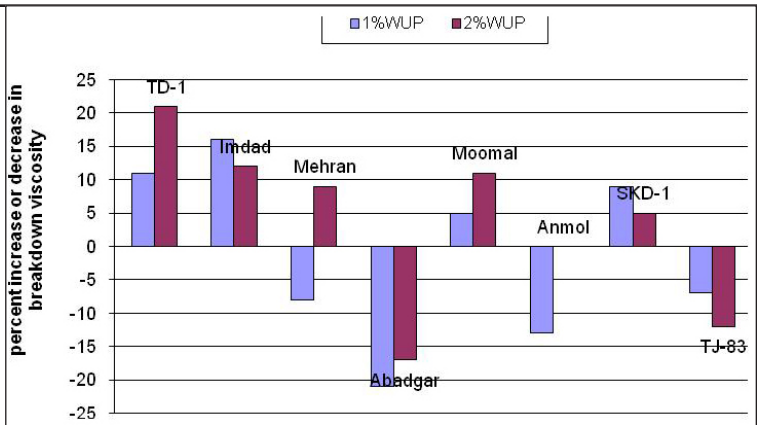

Fig. 37. Percent increase or decrease in the breakdown viscosity values of different hard white spring wheat cultivar flours.

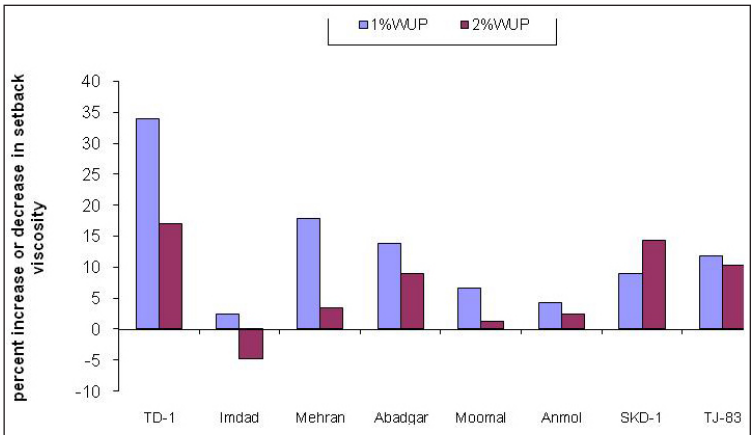

Fig 38. Percent decrease or increase in the setback viscosity of different hard white spring wheat cultivar flours due to the addition of water-unextractable pentosan (WUP) addition.

Influence of water unextractable pentosan concentration on the pasting temperature and the time required to reach the pasting temperature of wheat flours.

Saqib Arif, Qurrat ul Ain Afzal, Mubarik Ahmed, Abid Hasnain, Najmus Sahar, Shazia Arif, Alvina Gul Kazi, and Abdul Mujeeb-Kazi

Pasting temperature (PT) is one of the most important parameters of wheat flour pasting properties. Determining the time required to reach peak viscosity also is important. Many intrinsic and extrinsic factors are known to determine the PT of wheat flour. The addition of pentosan, i.e., water unextractable in nature, to the flour may influence the PT of wheat flour.Assuming this, our study added water-unextractable pentosan (WUP) at 1% and 2% concentrations to eight different wheat flours in order to examine the shifting of PT with at different concentrations.

The effect of WUP at 1% and 2% addition was variable on the PT of flours (Fig. 39). Few cultivars showed an increase in PT whereas

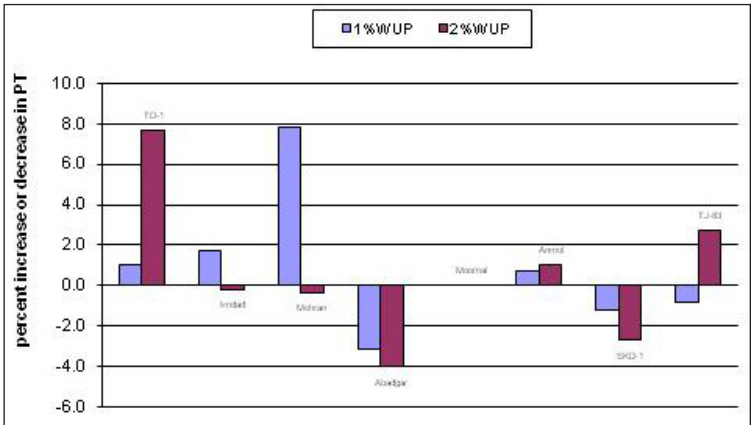

Fig. 39. The magnitude of increase or decrease (in %) in the pasting temperature (PT) of hard white spring wheat flour due to the addition of water-unextractable pentosan.

others had lower PT compared to the respective control flours. The largest reduction in PT was observed in the flour of Abadgar, followed by that of SKD-1 at both 1% and 2% levels of WUP. The reduction is valuable in bread-making because it implies an earlier beginning of starch gelatinization and, in turn, an increase in the availability of starch enzyme substrate during baking period (Rojas et al. 1999). The PT of flour of Moomal remained unchanged in the presence of WUP. The highest delay in PT was observed in the flour of cultivar Mehran at 1% WUP addition with a slight decrease when the WUP level increased to 2%. The PT of flour of TD-1 also was delayed at both levels of WUP, although to a higher degree at 2% WUP addition.

Fig. 40. Increase or decrease in time to reach peak viscosity due to the addition of water-unextractable pentosan (WUP).

Time to reach peak viscosity (TPV). Both levels of WUP supplementation imparted similar effects (whether increasing or decreasing) for each cultivar with the exception of TD-1 and Imdad (Fig. 40). However, the magnitude of the influence was different among all cultivars except TJ-83. The maximum delay was found in TPV of SKD-1 upon addition of both concentration levels, and the highest reduction (85 s) was in the flour of Mehran.

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The impact of a foliar application of manganese on various physiological and biochemical attributes of two contrasting wheat lines grown at different osmotic levels.

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Salinity is one of the major problems in arid and semi-arid regions of the world where annual precipitation is less than evapo-transpiration and is major impediment to crop production. Saline conditions result in high ion cytotoxicity and low osmotic potential, which in turn reduces the uptake of micronutrients. This reduction in micronutrients is another factor limiting plant growth under saline conditions. Along with other micronutrients, absorption of manganese also is reduced under the low osmotic potential of the medium because of excessive accumulation of soluble salts. Exogenous application of manganese is used to improve the growth of wheat, because manganese is component of photosystem II and, thus, involved in photosynthesis.

Two genotypes and three levels of salinity, 0 (control), 150, and 300 molm^{-3} NaC were used to detect the effect of each level of salinity on two wheat cultivars tested with and without Mn applied through the roots and shoots. Different physiological and biochemical parameters, such as ionic balance, osmotic potential, leaf chlorophyll content, photosynthetic rate, soluble proteins, proline, free amino acids, and soluble sugar, were estimated to understand the physiological and biochemical mechanism of salt tolerance in contrasting wheat cultivars.

Increased sodium concentration due to salt stress reduced the fresh and dry weight of shoot and root of both Pasban 90 and PBW 343; however the depression of biomass was ameliorated by Mn application through the roots (Table 28, p. 105). Salinity induced through a Mn deficiency might have been responsible for the retarded growth in all treatments, but the impact was more pronounced at salt stress with Mn deficiency. Foliar application increased the growth of salt-stressed plants by increasing the Mn availability but not to an extent that was accomplished when Mn was available through the soil medium. Potassium concentration in the plant tissue was reduced and that of Na increased as the Na salinity in the root medium increased. Manganese application, through the medium or by foliar application, enhanced the uptake of K and decreased that of Na in the shoots of the tolerant cultivar Pasban 90, which might have helped it maintain a low Na:K ratio; an essential feature for plants to survive under saline conditions. Increasing salt

Table 28. Mean shoot and root fresh and dry biomass (g/plant) of two wheat cultivars grown under different salinity levels with and with out manganese application.

Cultivar	Treatment	Shoot fresh weight (g)	Shoot dry weight (g)	Root fresh weight (g)	Root dry weight (g)
Pasban 90 (INIA-66/AG. DI.//INIA-66/3/ GENARO-81 [1388][2857])	T_0 (control)	47.7 A	10.15 B	21.6 A	4.69 B
	T_1 (-ive control)	33.6 B	7.87 C	16.6 B	2.86 C
	T_2 (NaCl 150 molm^{-3})	30.4 C	5.05 D	15.2 B	2.41 D
	T_3 (NaCl 300 molm^{-3})	24.2 D	3.57 E	7.8 EF	1.45 EFG
	T4 (-Mn +NaCl 150 molm^{-3})	17.2 F	3.96 E	5.0 HI	0.97 HI
	T_5 (-Mn + NaCl 300 molm^{-3})	7.6 I	1.15 H	4.6 I	0.84 I
	T_6 (-Mn + NaCl 150 molm^{-3} + Mn foliar spray	20.0 EI	3.58 E	9.6 DE	1.39 EFG
	T_7 (-Mn + NaCl 300 molm^{-3} + Mn foliar spray)	13.8 G	2.4 FG	6.6 FGH	1.22 FGH
PBW 343 (ND/VG9144// KAL/BB/3/ YACO/4/ VEE#5)	T_0 (control)	46.4 A	11.23 A	21.0 A	4.25 A
	T_1 (-ive control)	34.2 B	11.48 A	15.8 B	3.08 BC
	T_2 (NaCl 150 molm^{-3})	20.0 E	3.30 EF	12.0 C	1.69 E
	T_3 (NaCl 300 molm^{-3})	14.0 G	3.78 E	6.2 FGHI	1.4 EFG
	T4 (-Mn +NaCl 150 molm^{-3})	14.4 G	3.22 EF	5.8 GHI	1.18 GH
	T_5 (-Mn + NaCl 300 molm^{-3})	8.4 HI	1.87 GH	4.8 HI	1.13 GH
	T_6 (-Mn + NaCl 150 molm^{-3} + Mn foliar spray	14.6 G	3.54 E	10.6 CD	2.35 D
	T_7 (-Mn + NaCl 300 molm^{-3} + Mn foliar spray)	10.6 H	2.29 G	7.6 FG	1.64 EF
LSD Values	2.240	0.918	1.878	0.3601	

treatments consistently increased Cl and decreased the Mn content of shoots and roots of both wheat cultivars. The application of Mn significantly increased the shoot and root Mn content of both cultivars and application through roots had a significantly higher impact on Mn accumulation in shoots and roots of both cultivars. Manganese application, esspecially though the rooting medium, significantly enhanced the accumulation of organic osmotica such as soluble proteins, free amino acids, and proline. This, along with the uptake of inorganic osmotica might have helped to reduce the osmotic potential tolerant cultivar to produce turgidity. Increased salinity levels decreased chlorophyll a content and photosynthetic rate of both cultivars, and a foliar Mn application ameliorated this effect more effectively than a Mn application through roots.

Our results make it clear that Mn application ameliorated the negative consequences of salinity stress by enhancing the accumulation of inorganic and organic osmotica, chlorophyll content, and photosynthetic rate. In most cases, Mn application through the rooting medium showed more pronounced, positive impacts, except for chlorophyll content and photosynthetic rate, where Mn application through shoots enhanced the both parameters under salt stress. This study also shows that Mn application has partially ameliorated the hazardous effects of salinity stress by ionic balancing through reducing the uptake of Na and Cl and enhancing that of K and Mn; accumulating organic osmotica including soluble proteins, free amino acid, and proline to maintain better turgor; and increasing the chlorophyll content and photosynthetic rate. Manganese application through rooting medium had a more pronounced, positive effect on nearly all parameters except chlorophyll content and photosynthetic rate, whereas Mn application through shoots alleviated the negative consequences of salinity more effectively than Mn application through roots. To elucidate how this all happens, it will be necessary to work out the role of Mn on various enzymes involved in metabolism and photosystems.

Screening synthetic hexaploid wheat for drought tolerance at early growth stages.

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Drought is one of the main agricultural menaces limiting the successful exploitation of land potential and consequently reducing crop productivity. Screening of available germ plasm of a crop for drought tolerance is of considerable value for utilizating drought-affected soil. During the early phase of this study, response of 116 wheat lines from a diverse gene pool was assessed at germination stage using polyethylene glycol (PEG 8000) at 10% and 20% in full strength Hoagland's nutrient solution (Table 29, pp. 106-107). Seedling growth was determined in an independent study. The

Table 29. Description of seed material of the synthetic hexaploid (SH) wheats used in the study to screen for drought tolerance.

Genotype	Original #	Source/Origin
Dh Drought-1	1	CPI/GEDIZ/3/GOO//JO69/CRA/4/*Ae. tauschii* (208)/5/Opata
	58	
	59	
	70	
	102	
	120	
Dh Drought-2	3	YAV-3/SCO//JO69/CRA/3/YAV79/4/*Ae. tauschii* (498)/5/Opata
	29	
	64	
	36	
	43	
Dh Drought-3	49	D67.2/P66-270//*Ae. tauschii* (257)/3/Opata
	50	
	60	
	61	
	70	
Dh Drought-4	6	GAN/*Ae. tauschii* (897)//Opata
	22	
	51	
	61	
	70	
Dh Drought-5	5	DOY1/*Ae. tauschii* (458)//Opata
	17	
	35	
	46	
	53	
SH DD Drought BV/02	10	D67.2/P66.270//*Ae. tauschii* (217) DVERD_2/*Ae. tauschii* (221) D67.2/P66.270//*Ae. tauschii* (257) CETA/AE. CETA/*Ae. tauschii* (1026) CETA/*Ae. tauschii* (1031)
ONMKDISH	1	Altar 84/*Ae. tauschii*//Opata
	2	CROC_1/*Ae. tauschii* (224)//Opata
	3	CROC_1/*Ae. tauschii* (224)//Opata
	4	CHEN/*Ae. tauschii*//2*Opata
	5	Altar 84/*Ae. tauschii*/2*Opata
MX101-02		
M11SAWYT	1	CROC_1/*Ae. tauschii* (205)//KAUZ/3/Sasia
	2	Dharwar Dry/Nesser
	3	Dharwar Dry/Nesser
	4	SUJATA/SERI
	6	PASTOR/3/MUNIA//CHEN/Altar 84/5/CNDO/RI43//ENTE/MEXI_2/3/*Ae. tauschii*/4/Weaver
	7	FILIN/Irena/5/CNDO/RI43//ENTE/MEXI_2/3/*Ae. tauschii*/4/Weaver
	18	URES/PRL//AV32
	20	Pastor/VAV92
	21	Irena/Babax//Pastor
	22	Irena/Babax//Pastor
	23	CROC_1/*Ae. tauschii* (224)//Opata/3/Pastor
	25	BJY/COC//PRL/BOW/3/Attila

Table 29. Description of seed material of the synthetic hexaploid (SH) wheats used in the study to screen for drought tolerance.

Genotype	Original #	Source/Origin
M11SAWYT	26	Parus/Pastor
	27	FILIN/3/CROC_1/*Ae. tauschii* (205)//KAUZ/4/FILIN
	28	FILIN/3/CROC_1/*Ae. tauschii* (205)//KAUZ/4/FILIN
	29	GEN*2//BUC/FLK/3/2*Pastor
	32	VEE/MJI//2*TUI/3/2*Pastor
	33	Milan/KAUZ//Babax/3/Babax
	35	Pastor/3/Altar 84/*Ae. tauschii*//Opata
	37	URES/JUN//KAUZ/3/Babax
Darwar		
Sitta		
Nesser		
Opata		
Veebli		
Inqalab-91		
Kohistan-97		
Baraine 83		
C-591		
C-271		
Rohtas 90		

Richardselection-3	Richard selection-16	Richard selection-25
Richard selection-4	Richard selection-17	Richard selection-26
Richard selection-6	Richard selection-18	Richard selection-27
Richard selection-8	Richard selection-19	Richard selection-28
Richard selection-9	Richard selection-20	Richard selection-29
Richard selection-13	Richard selection-21	Richard selection-30
Richard selection-14	Richard selection-22	Richard selection-32
Richard selection-15	Richard selection-23	Richard selection-33
Richard selection-34	Richard selection-52	Richard selection-75
Richard selection-35	Richard selection-53	Richard selection-76
Richard selection-41	Richard selection-57	Richard selection-77
Richard selection-42	Richard selection-58	Richard selection-79
Richard selection-46	Richard selection-62	L. sequia-8
Richard selection-47	Richard selection-66	L. sequia-11
Richard selection-48	Richard selection-69	L. sequia-12
Richard selection-49	Richard selection-70	L. sequia-13
Richard selection-50	Richard selection-71	
Richard selection-51	Richard selection-74	

study was designed across two treatments each with three replicates having three plants/pot. T_0 was the control treatment and T_1 was the drought evaluation treatment.

The lines M11SAWYT 25, 36, 21, and 34 showed significantly higher total germination percent in the PEG solutions and C-271 and the Richard's selections 29, 33, 34, 23, and 41 showed lower germination than the other lines. The M11SAWYT 32 line and Richard's selections 3 and 79 showed relatively poor performance for all the growth parameters measured at the seedling stage. No consistent relationship between tolerance was observed at the germination and seedling stages, however, the presence of a great amount of variability for drought tolerance in wheat observed in this study

at each stage can be beneficial for improving drought tolerance through recurrent selection. Data for different growth parameters at the seedling stage showed that WEEBLI; Dh Drought 2/29, 3/60, 4/70, and 5/5; and ONMKDISH 1 produced significantly greater biomass in absolute terms, and these lines also were characterized as tolerant lines on the basis of their performance for number of tillers and relative water content under drought stress (Table 30). The good performance of these tolerant lines could be attributed to osmotic regulation, which might have enabled the maintenance of cell turgor that could assist plant survival under drought stress. High tiller numbers in these drought-tolerant lines could have played central role in their recovery from early season drought, and this character might have contributed to greater biomass production under water-deficient conditions. The prolific root system of these drought-tolerant lines could have improved

Table 30. Classification of 116 lines of wheat on the basis of their performance

Class	Range	Number	Genotypes
Root length (cm) after two month growth under drought stress			
I	36–40	1	Dh drought-4/6
II	31–35	9	Sh DD Drought; Dh Drought-3/60, -3/61, -3/77; ONMKDISH 1, 2, 3, Weebli, M11SAWYT 36
III	26–30	29	C-591; L. Sequia-8, -13; Kohistan-97, Opata, C-271, Dh Drought-2/29, -2/43, -3/49, -3/50, -4/22, -4/70, -5/5, -5/17; M11SAWYT 18v, 5, 6, 7, 20, 21, 33, 34, Richard selections-14, -48, -49, -51, -70, -75, -76
IV	21–25	70	ONMKDISH 5, Richard selections-3, -4, -6, -9, -13, -15, -16, -17, -18, -19, -20, -21, -22, -23, -25, -26, -27, -28, -29, -30, -32, -33, -34, -35, -41, -42 , -46, -47, -50, -52, -53, -57, -58, -62, -66, -68, -69, -71, -74, -77, -79; L. Sequia-11; Nesser Baranie-83; M11SAWYT 8, 10, 11, 22, 23, 25, 27, 28, 29; Dh drought-1/102, -1/1, -1/120, -1/58, -2/34, -2/36, -4/61, -4/51, -5/46, -5/35, -5/53; Rohtas-90, Inqalab-91; ONMKDISH 4, Darwar, L. Sequia-12
V	15–20	7	Dh drought 1/59, 1/70, 2/3; Sitta; M11SAWYT 26, 32; Richard selection-8
Relative water content after two month growth under drought stress			
I	91–100	7	Dh drought-1/59, -2/3, -2/43, -3/60; M11SAWYT 7; Weebli; Sitta
II	81–90	48	M11SAWYT 18v, 5, 6, 10, 11, 18v, 20, 21, 23, 25, 29, 32, 36; ONMKDISH 4; Nesser; M11SAWYT 6, 10, 25, 26, 32; C-271; ONMKDISH 1, 2, 3, 5; Richard selections-52, -58; Rohtas-90, Sh DD Drought, Inqalab-91; Darwar, Dh Drought-1/120, -1/1, -1/70, -1/58, -2/34, -2/29, -2/36, -3/50, -3/61, -3/77, -3/49, -4/51, -4/6, -4/70, -4/22, -4/61, -5/35, -5/53, -5/5, -5/17, -5/46, -5/5
III	71–80	42	C-591; M11SAWYT 8, 22, 27, 28, 33; Kohistan-97; L. Sequia-8, -11, -12; Baranie-83; Richard selections-3, -4, -6, -8, -9, -13, -14, -16, -18, -19, -22, -28, -29, -30, -32, -33, -35, -41, -46, -47, -48, -51, -53, -57, -69, -70, -71, -74, -76, -77, -79
IV	61–70	17	Opata; M11SAWYT 34; L. Sequia-13; Richard selections-15, -17, -20, -23, -25, -26, -27, -34, -42, -49, -50, -62, -66, -68
V	51–60	1	Richard selection-21
VI	40–50	1	Richard selection-75

the water uptake and this imperative character might have increased the above-ground, dry biomass. The tolerance of WEEBLI; Dh Drought-2/29, -3/60, -4/70, and -5/5; and Dh ONMKDISH 1 observed at the seedling stage was not conferred at the germination stage, because all these lines were ranked as moderately tolerant for germination percent.

The results presented here deal with the drought tolerance of 116 lines of wheats at two initial growth stages, germination and seedling. The tolerance observed in six lines (WEEBLI; Dh Drought -2/29, -3/60, -4/70, and -5/5; and Dh ONMKDISH 1) at the early growth stages may or may not be conferred at the adult stage. Nevertheless, the tolerance observed here is of great economic importance, because many workers have emphasized that the assessment of drought tolerance at every growth stage of a plant species is of considerable value in determining the magnitude of genetic diversity and the ultimate tolerance of the species.

Development of drought-tolerant wheat cultivars.

Abid Subhani, Attiq-ur-Rehman, M. Ashraf Mian, M. Sabar, M. Ihsan, M. Tariq, and Abdul Mujeeb-Kazi.

Wheat is a staple diet of major population in Pakistan (Ahmad et al. 200; Ajmal et al. 2009) contributing 13.1% of the value added in agriculture and 2.7% to GDP (Anonymous 2011). Wheat supplies 72% of the calories and proteins in the average diet (Azam et al. 2007). Areas receiving rainfall above 350 mm annually are able to grow a wheat crop under rainfed conditions. The Barani areas have a great significance in wheat production, covering about 12.3% of the area under wheat and contributing about 7% of the wheat production in Punjab and 7.65% in Pakistan (Anonymous 2009). Rain fall distribution is, of course, very crucial for crop production. With considerable year to year variation in precipitation, uneven rainfall and crop yields are more frequent in the Rabi season than in Kharif (Fig. 41).

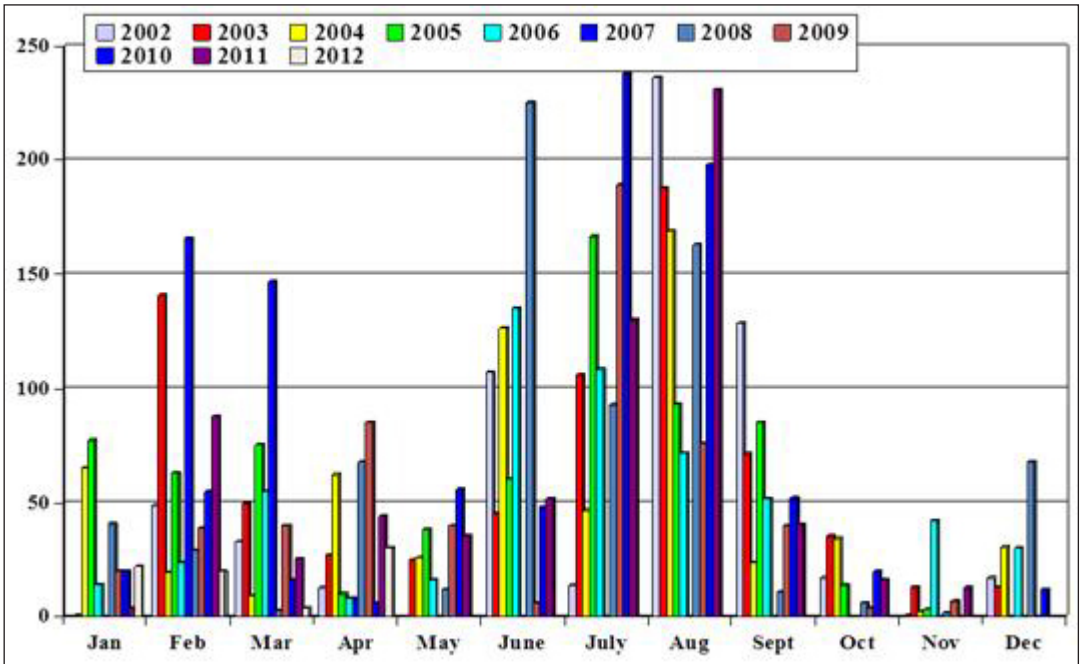

Fig. 41. Rainfall at Chakwal, Pakistan, 2002-2012.

Both biotic and abiotic stresses are major threats to the crop productivity. In Pakistan, brown and yellow rusts are among the serious diseases of wheat and, in most cases, wheat cultivars are replaced with newer cultivars due to susceptibility (Rattu et al. 2007). Wheat cultivars with narrow, erect to semi erect leaves and deep root systems that tolerate drought stress and leads to higher biomass and grain production are the most suitable. Presently, Chakwal-50, GA-2002, and Inqilab-91 are commonly grown in the rain-fed areas of the Punjab. Because these cultivar have become susceptible to rusts, it is necessary to develop new, disease-tolerant wheat cultivars suitable for the rainfed areas.

A team of devoted, highly qualified research Scientists is engaged at Barani Agricultural Research Institute, Chakwal, for this purpose. The wheat research team is working day and night to develop high-yielding cultivarss and production technology for wheat in the Barani area with the main objectives to

collect and maintain germ plasm of wheat for utilization in the hybridization program,
morphologically and physiologically characterize germ plasm,
develop high-yielding, drought-tolerant and disease resistant wheat cultivars suitable for rainfed areas,
collaborate with national and international organizations through exchange and evaluation of wheat material, and
maintain genetic purity of wheat cultivars through BNS, pre-basic and basic seeds.

The Barani Agricutural Research Institute, Chakwal, received a set of 36 wheat genotypes from Dr. Abdul Mujeeb-Kazi during the 2011–12 season. This material was planted at the Barani Agricultural Research Institute in six 5-m rows and evaluated under rainfed conditions. Fertilizer was applied at 90:60:30 N:P:K. Morphological and disease data were recorded and nine high-yielding genotypes were selected based on this data. These nine genotypes are now included in crossing block and gene pool of Barani Agricultural Research Institute. These selected genotypes are included in a preliminary wheat yield trial in 2012–13 for further evaluation (Table 31, p. 110).

Table 31. Phenological parameters evaluated in a set of 36 wheat genotypes in the 2011–12 crop cycle at the Barani Agricutural Research Institute, Chakwal, Pakistan.

Genotype	Germination %	Plant height (cm)	Spike length (cm)	Tillers/m	Grains/spike	Yield (kg/ha)
K1	90	116	10.8	130	56.7	6,170
K2	87	116	11.7	141	57.0	5,740
K3	82	112	12.1	133	43.7	4,540
K9	82	117	10.5	142	51.3	2,140
K11	80	113	11.4	128	44.0	3,250
MK2	82	95	10.2	168	43.7	4,057
MK3	80	100	11.0	120	47.0	3,511
MK4	82	98	11.6	132	46.7	4,591
MK5	78	97	10.4	160	40.0	3,102
MK6	75	101	12.4	137	36.7	3,115
MK7	82	104	11.3	174	53.7	4,582
MK8	87	113	12.6	160	61.0	5,742
MK9	84	103	10.7	123	71.7	4,048
MK10	82	104	11.1	164	76.7	3,844
MK11	76	105	11.7	132	63.7	3,218
MK12	82	110	12.0	158	75.0	4,262
MK13	72	103	12.1	130	69.3	5,506
MK14	73	99	10.5	168	50.0	3,266
MK15	77	104	12.3	180	66.7	4,622
MK17	80	104	11.2	188	58.3	4,364
MK18	75	104	11.4	135	41.0	4,164
MK19	72	107	10.6	152	70.3	4,368
MK20	74	97	10.4	191	44.7	3,471
MK21	70	88	11.2	139	62.3	3,840
MK22	76	90	11.9	135	57.3	3,142
MK23	72	101	11.4	181	50.3	3,386
MK24	74	101	11.8	193	64.0	3,599
MK25	65	101	11.0	207	53.7	2,738
MK27	75	105	12.5	122	67.0	4,284
MK28	70	103	12.1	105	41.0	4,613
MK29	72	107	12.2	118	55.7	3,689
MK30	70	93	12.0	190	49.0	3,129
MK31	62	92	11.9	153	46.0	2,040
MK32	65	92	12.1	184	53.3	2,866
MK33	75	99	11.1	168	53.3	2,998
MK34	78	105	13.4	197	57.0	1,652
Selected wheat material						
K1	90	116	10.8	130	56.7	6,170
K2	87	116	11.7	141	57.0	5,740
MK8	87	113	12.6	160	61.0	5,742
MK13	72	103	12.1	130	69.3	5,506
MK15	77	104	12.3	180	66.7	4,622
MK28	70	103	12.1	105	41.0	4,613
K3	82	112	12.1	133	43.7	4,540
MK4	82	98	11.6	132	46.7	4,591
MK7	82	104	11.3	174	53.7	4,582

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Breeding of wheat genotypes for the semidwarf character and high grain yield.

Wheat is the basic staple food for most of the population and largest grain source of the country. The importance of wheat is always recognized when formulating agricultural policies. Wheat contributes 12.5% to the value added in agriculture and 2.6% to GDP. Wheat was cultivated in an area of 8.666 x 10^6 ha in 2011–12 (Table 1), a decrease of 2.6% over last year when the area of 8.901 x 10^6 ha. A production of 23.5 x 10^6 is estimated in 2011–12. The yield/ha posted a negative growth of 4.2% compared to 11% last year, which is due to the fact that sowing was delayed because of standing water and other climatic factors.

Table 1. Area, production, and yield of wheat grown in Pakistan between the 2007–08 and 2011–12 crop years (Source: Pakistan Economic Survey (2011–12).

Year	Area		Production		Yield	
	x 10^6 ha	% change	x 10^6 ha	% change	x 10^6 ha	% change
2007–08	8.550	–0.3	20.959	–10.0	2.451	–9.8
2008–09	9.046	5.8	24.033	14.7	2.657	8.4
2009–10	9.132	1.0	23.311	–3.0	2.553	–3.9
2010–11	8.901	–2.5	25.214	8.2	2.833	11.0
2011–12	8.666	–2.6	23.517	6.7	2.714	–4.2

Progress of cultivar NIA-Sunhari. A total of 5,000 kg of NIA-Sunhari pre-basic seed was produced from three acres during 2011 out of which 2,500 kg seed sold to growers and seed companies during 2011–12.

Candidate varietal material. Two candidate cultivars (NIA-22-03 and NIA-54-03) are being maintained for sending as entries to National Uniform Wheat Yield Trials (NUWYT).

Zonal Trial studies. Two genotypes (NIA-6-12 and NIA-CIM-04-10) were tested in zonal/regional trials in the Sindh province. The performance of NIA-CIM-04-10 was comparatively better for grain yield (kg/plot).

Trial I. This trial was conducted with 14 advanced lines with two check cultivars, NIA-Sunhari and Kiran. The trial was sown on 2 November, 2010. The trial had six rows of each genotype with a 4-m row length in three replicates. The results indicated that line (01) had the highest grain yield/plot (2.52 kg). Possible reasons could be that the line also had a

higher number of spikelets/spike and increased main spike grain yield. Subsequent lines that had higher grain yield were 03 (2.37 kg), 08 (2.4 kg), Kiran (2.32 kg), 12 (2.23 kg), 11 (2.20 kg), and 02 (2.18 kg). Line 10 had the lowest grain yield (0.983 kg), possiblally because ths line possesses the lowest number of grains/main spike.

Trial II. This trial consisted of 16 genotypes including the two check cultivars NIA-Sunhari and Kiran. The trial had six rows of each genotype with a 4-m row length in three replicates. Sowing of the trial was completed on 2 November, 2010. The results showed that line 09 had the highest grain yield/plot (2.75 kg). Subsequent lines that had higher grain yield were Kiran (2.70 kg) and line 02 (2.65 kg). The possible reasons for higher grain yield in Kiran and line 02 could be due to increased main spike grain yield. Line 05 (1.65 kg) had the lowest grain yield per plot compared with other lines and cultivars.

Trial III (Isoline material). Thirty-six genotypes of isogenic material for Norin-10 (*Rht1*, *Rht2*, *rht*) were sown on 3, November, 2010. The trial had six rows of each genotype with a 4-m row length of four meters in three replicates. In this comparison, line 03 (2.433 kg) had the highest grain yield/plot. Subsequent lines with a higher grain yield were 17 (2.400 kg), 28 (2.383 kg), 26 (2.333 kg), 19 (2.283 kg), 30 (2.250 kg), 18 (2.233 kg), and 2 (2.217 kg). Line 08 (0.750 kg) had the tallest plant height with the lowest grain yield due to lodging.

Mutation breeding for drought tolerance. The material for drought consisted of 72 M_4 genotypes planted with two 4-m rows in three replicates. Lines 62 and 63 had produced the highest grain yield (388 g/plot) under zero/no irrigation. Line 64 had produced the highest grain yield (542 g/plot) under two irrigations. Line 58 had produced the highest grain yield (542 g/plot) under full irrigation conditions. Line 64 had also produced the highest grain yield (461 g/plot) average performance over three irrigation treatments.

Salinity tolerance. The salinity of soil ranged from 3.1 to 11.4 EC (1:2.5) ds/m. The same 72 M_4 progenies used for the drought studies also were grown in a saline soil. Line 9 (330 g/plot), from mutated progenies of the cultivar Bhittai, had the highest grain yield/plot than that of all other genotypes and check cultivars. Subsequent lines derived from Bhittai that had a higher grain yield were 17 (245 g), 15 (233 g), 30 (230 g), 10 (225 g), and 22 (215 g). The mutated line 10 (270 g) derived from the cultiar Kiran, had the highest grain yield/plot. Subsequent lines with higher grain yield/plot from the cultivar Kiran were 5 (237 g) and 23 (215 g). The Kiran check had 202 g grain yield/plot. Thirty mutant progenies were selected for next M_5 generation.

Mutation studies under normal conditions. The 72 mutated M_4 progenies used for drought studies also were grown in normal soil conditions. The lines that produced highest grain yield/plot were 14 (428 g), 15 (425 g), 21 (440 g), and 27 (437 g) from the cultivar Bhittai. However, the maternal Bhittai parent had a plot grain yield of 328 g. Lines that had the highest grain yield/plot were 11 (588 g), 15 (563 g), and 17 (588 g) from the cultivar Kiran. However, the maternal Kiran parent had a comparatively lower grain yield/ plot (430 g).

I**ntroduction of *Rht8*.** The semidwarfing gene *Rht8* was transferred from the Italian cultivar Mara. The material is in the F_5 generation.

Breeding material. The breeding material consisted of three crosses F_1 generation, four crosses from the F_4 generation, two crosses from the F_7, one cross from the F_8.

Germ plasm material. We are maintaining 140 lines with different characteristics for use in the crossing program.

Sanction of a new international project. A new IAEA/RCA/RAS/05/56 project entitled 'Supporting mutation breeding approaches to develop new crop varieties adaptable to climate change' has been sanctioned for the 2012 and 2015.

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ITEMS FROM POLAND

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Genomes of wheat and other grasses during the cell cycle and apoptosis.

R. Kosina and P. Tomaszewska.

In nature, some plants of hybrid origin cannot easily be distinguished from their known or supposed parental species, mother or father, in terms of their gross morphology. This means that both parental genomes act unequally, being either expressed or suppressed. In the literature, this is known as uniparental dominance. This phenomenon has been well exemplified for sets of microstructural characters in the hybrid progeny of wheat tetraploids and within the tribe Triticeae for amphiploids (Kosina 1995, 1996). For instance, a set of caryopsis traits of *T. turgidum* subsp. *polonicum* dominates over similar ones in *T. turgidum* subsp. *carthlicum*. Another important process that defines genome relationships in plant hybrids is apoptosis (Tomaszewska and Kosina 2013). Apoptosis is a programmed phenomenon eliminating bad or useless nuclei, cells, or tissues. A question arises: Is the apoptotic role of parental genomes equal or unequal in a hybrid body? Many examples of apoptosis can be observed on the embryonic path of a plant. In antipodals, in differentiating xylem vessels, or in an aging root cap, nuclei are condensed (Fig. 1) and next a chromatin and DNA are cleaved by nucleases.

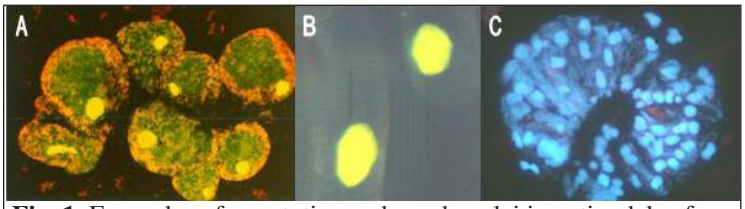

Fig. 1. Examples of apoptotic condensed nuclei in antipodals of *Leymus glaucus* (A), in young xylem vessels of an amphiploid *T. turgidum* subsp. *carthlicum* / *Ae. tauschii* (B), and in a root cap of *Brachypodium distachyon* (C).

Two main arrangements of parental genomes are recognized in plant hybrids: a sectorial one (Fig. 2A) or side-by-side, where both types of genomes can be expressed – hybrids are, most often, intermediate to parents, a concentric one (Fig. 2B), where the genome of one parent is enclosed by that of the second parent, and the outer genome is expressed – hybrids present uniparental dominance. Surprisingly, a change in the genome arrangement is noted in nuclei entering apoptosis (Fig. 2B).

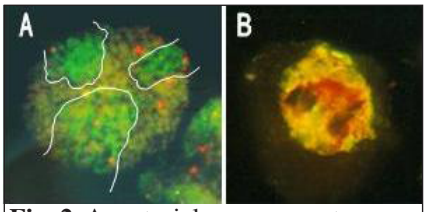

Fig. 2. A sectorial arrangement of parental genomes (green and brown) in a root prophase nucleus of an amphiploid *T. turgidum* subsp. *durum* / *Thinopyrum distichum* / *Lophopyrum elongatum* and a concentric one (yellow and red) in an apoptotic nucleus of the same amphiploid.

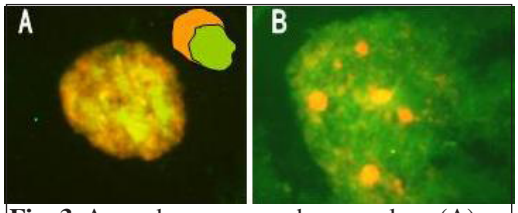

Fig. 3. An endosperm prophase nucleus (A) in an amphiploid *Avena barbata* (AABB) / *A. nuda* (AA) with probed A genomes (green) and B genomes (red) – genomes are arranged side-by-side, and an early apoptotic endosperm nucleus (B) of the same amphiploid – the B genome of *A. barbata* is highly condensed and situated within a green sphere of A genomes of both parental species.

Two parental genomes in an *Avena* amphiploid were identified by GISH and they are arranged side-by-side in a living endosperm nucleus (Fig. 3A). When the nucleus approaches apoptosis, one selected genome, here B, is condensed and not expressed within green A genomes belonging to both parents (Fig. 3B).

These results prove that the roles of parental genomes in a hybrid plant are different during a normal cell cycle and during apoptosis. In apoptosis, the choice of a functional genome is not species, but genomically, specific.

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Cytogenetic events in a 'Triticum timopheevii subsp. timopheevii / Aegilops umbellulata' amphiploid with native and demethylated genomes.

R. Kosina, A. Koźlik, and K. Markowska.

In the above amphiploid, the expected number of chromosomes is 42. The number observed most often is 41, in both, native and demethylated. Telocentrics also were noted for both types of genomes (Fig. 4A). Divided telocentrics were noted as laggards during telophase and, as a consequence, two micronuclei were eliminated. The most important phenomenon in the amphiploid with native genomes was the appearance of fully decondensed, anaphase chromosomes. In an amphiploid with demethylated genomes, we observed both elongated metaphases in the form of 'multi-Rabl' (Fig. 4B) and increased export of RNA from nucleoli into cytoplasm. Cytomixis is common here (Fig. 4C), and this is not for the native state. Cytomixis results in an unequal distribution of chromatin (genes) in sister nuclei and asymmetric condensation of chromatin is linked with this. Telocentrics also were observed in other amphiploids of the Triticeae, and this resulted in a higher variation of the chromosome number (Kosina and Heslop-Harrison 1996). Enhanced RNA export is correlated with the synthesis of abundant RNA after demethyhlation of genomes, as previously reported (Kosina and Markowska 2010). The frequency of the most common number of nucleoli in the main and lateral roots also is changed after demethylation (Kosina and Markowska 2011).

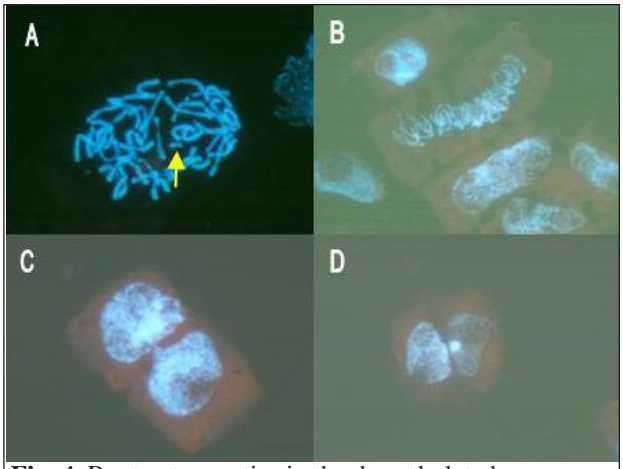

Fig. 4. Root cytogenetics in the demethylated amphiploid. A – metaphase with telocentric chromosome (arrow), B – an elongated Rabl configuration in prophase–metaphase, C – cytomictic nuclei, and D – asymmetric chromatin condensation in cytomictic nuclei. All pictures in DAPI fluorescence.

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On interrelations between a placental xylem and a nucellar projection in a 'Triticum timopheevii subsp. timopheevii / Aegilops umbellulata' amphiploid

R. Kosina, A. Koźlik, and K. Markowska.

A placental vascular bundle, composed of xylem and phloem, develops on the abaxial side of the caryopsis. The phloem is most often divided into two separate strands and is hardly seen in a ripe caryopsis. Xylem vessels (proto and meta) are

recognized clearly by means of a polarizing or fluorescence microscope. Cellulosic vessels are either optically active in polarizing light or express light blue auto-fluorescence. In *T. timopheevii* subsp. *timopheevii*, 7 to 20 (minimum–maximum) vessels were identified, and they were close to the pigment strand. Smaller caryopses of *Ae. umbellulata* have narrower bundles with 5–9 (minimum–maximum) vessels. In the amphiploid with native genomes, we noted 4–14 (minimum–maximum) vessels, and with demethylated genomes the range was almost the same, i.e., 4–11 (minimum–maximum). The amphiploid xylem is an intermediate between its parents (Fig. 5A,B). Among wheat tetraploids, *T. timopheevii* subsp. *timopheevii* has the richest placental bundle (Kosina 1988). Wang et al. (2008) provided data showing that xylem and phloem apoptosis in such a bundle is different. In the last stages of caryopsis development, both elements of the vascular bundle are dead.

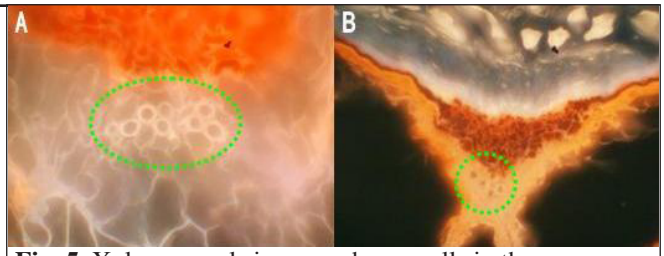

Fig. 5. Xylem vessels in parenchyma cells in the demethylated amphiploid (A), and vessels (outlined) in an acellular, obliterated tissue below a brown pigment strand in the native amphiploid (B). A and B – different magnifications.

Chalazal tissues, pigment strand and nucellar projection, are paths for assimilates, which flow into endosperm. The pigment strand, even when it is suberized, has active plasmodesmata and is open to assimilates (Oparka and Gates 1982). The gross morphology of nucellar projection in both parental species is different, and this kind of variability is illustrated (Fig. 6A and B) as two extreme variants occurring in the amphiploid. In the cross-section, the nucellar projection can be either narrow and high (Fig. 6A) or broad and low (Fig. 6B). Most often, a high nucellar projection is formed by elongated cells of the chalazal region. High correlations between the size of caryopsis and the number of xylem vessels were found; large caryopses developing in lower parts of the spikelets have richer vascular bundles. Another important correlation ($r = 0.77$; significant at $\alpha = 0.001$) exists between the number of xylem vessels and the height of nucellar projection. This means that an abundant placental vasculature positively affects both the growth of nucellar projection as transfer tissue and the caryopsis as a whole. As a reverse example, we noted a nucellar projection in *Ae. umbellulata* with a degraded cellular structure, which strongly influenced the development of caryopsis.

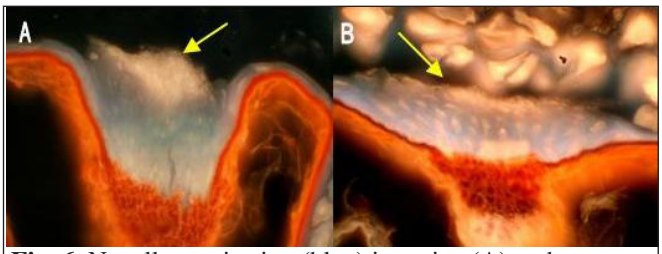

Fig. 6. Nucellar projection (blue) in native (A) and demethylated (B) types of amphiploid. Light apoptotic remnants of nucellar tissue shown by arrows

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Variability of pigment strand in a 'Triticum timopheevii subsp. timopheevii / Aegilops umbellulata' amphiploid and related species.

R. Kosina, A. Koźlik, and K. Markowska.

The pigment strand is a caryopsis structure which develops between the placental vascular bundle and nucellar projection, i.e., a changed area of chalaza. The pigment strand is connected with a pigment layer surrounding the caryopsis and is a component of the testa. Koźlik (2013) offered the first description of the pigment strand structure in a *Triticum*/*Aegilops* amphidiploid with native and demethylated genomes. Kosina (1995) proved that the gross morphology of the

pigment strand is genotypically significantly differentiated in wheat tetraploids. Also, its shape greatly varies in wheats and goat grasses. In grasses, cell walls and protoplasts of the pigment strand are often changed by suberin, phytomelans, carotenoids and phenols, which determine, among other factors, seed dormancy. Despite the suberinization of cell walls, the pigment strand has long active plasmodesmata and its role as transductory tissue continues (Oparka and Gates 1982). In the *Triticum*/*Aegilops* amphiploid, cell walls of the strand show light fluorescence of cellulose and a brown, suberized layer inside. This is recognized as an initial stage of suberinization, without synthesis of the subsequent layers of cellulose and suberin. Kosina and Tomaszewska (2011) discovered that, in interspecific *Avena* amphiploids, a transfer complex (pigment strand + nucellar projection) is highly diversified. The variability of the pigment strand in the studied *Triticum*/*Aegilops* amphiploid is presented (Fig. 7). The pigment strand can either be very narrow and composed of a few cells (Fig. 7A) or can be broad and multi-celled (Fig. 7B). Its cells are like puzzles (Fig. 7C), and such a structure distinctly increases the plasmolemma surface, the number of plasmodesmata and the transductory potential of this tissue. On the other hand, its cells can be round and their conductive role is then weaker (Fig. 7D). Suberinization of the strand occurs unequally within this structure and a kind of mosaic with lighter and darker parts is documented (Fig. 7E,F). Koźlik (2013) found that only the width of the pigment strand is positively correlated with the size of caryopsis and this proves its role as a conductive gate for developing endosperm.

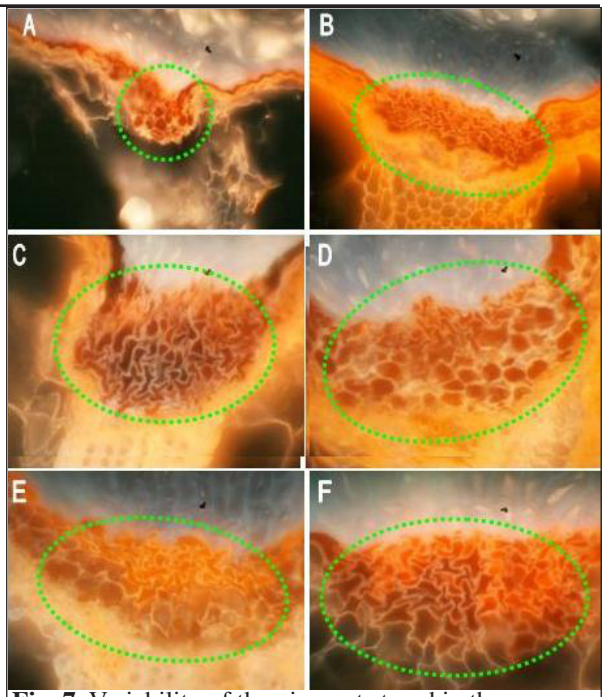

Fig. 7. Variability of the pigment strand in the amphiploid with native (A-E) and demethylated (F) genomes. The strand is outlined.

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Structural characteristics of grass hybrid endosperm development.

R. Kosina, M.K. Bureś, M. Florek, A. Grabińska, P. Kawa, B. Kłyk, Ł. Kochmański, A. Koźlik, A. Kurek, J. Skowrońska, P. Tomaszewska, and D. Zając.

The idea on hybrid endosperm development presented below is drawn from many studies of the grass genera, *Triticum*, *Avena*, *Bromus*, *Lolium*, and *Brachypodium* and intergeneric hybrids of the tribe Triticeae. Ivanovskaja (1983) presented data on the differentiation of isolated nuclei and cells in a young hybrid grass embryo sac or nuclear endosperm. A cytogenetic approach identified these nuclei and cells as subsyncytial units (Kosina 1996). Kosina (1992, 2012) proved that grass endosperm is composed of developmentally distinct units, i.e., cell clones. One of the most intriguing development issues is the relationship between the development of nucellar chalaza and endosperm. The apoptotically changed chalaza is preserved in the form of a long and narrow border running along the axis of the caryopsis and reaching the dorsal aleurone layer (Fig. 8A, p. 117). Potentially, it forms two large endosperm domains which cover the two halves of the caryopsis. These two halves can develop independently and can differ structurally. However, their persistence over time can have its beginning in the nuclear endosperm and the present status can be a result of earlier nuclear differentiation, including subsequent intrusive growth of chalazal tissue. We have observed the same growth behavior, as in chalaza, in the nucellar epidermis or in the aleurone layer (Fig. 8B, p. 117). Cells growing intrusively always separate endospermal

areas, i.e. domains. The subsyncytial nature of nuclear endosperm and its later domainal development are two basic processes creating a mosaic endosperm. It is especially frequent in grass hybrids (Kosina 2007, Koźlik 2013). Very large domains (Fig. 10A) or smaller cell clones (Fig. 11A) can be distinguished in both endospermal tissues, aleurone and starchy. Each characteristic of endosperm relating to metabolism, structure or function can be mutationally changed; for instance: synthesis of cell wall components (Fig. 9B), expression of starch phenotype in the aleurone layer (Fig. 9A,10B), polyploidization (Fig. 11A), synchronized cell cycle (Fig. 11B), and protein, starch or globoids synthesis. This extremely variable nature of endosperm and its simple tissue composition, aleurone plus starchy cells, creates the best research model subject in the area of applied plant biology.

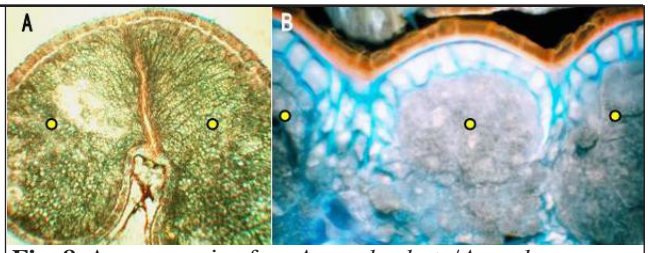

Fig. 8. A - caryopsis of an *Avena barbata*/*A. nuda* amphiploid with a light line of chalazal remnants penetrating deeply into the starchy endosperm; two possible domains are marked by yellow dots. B - caryopsis of an *Elymus canadensis*/*Pseudoroegneria libanotica* amphiploid with an aleurone layer (blue) growing deeply into starchy endosperm; three possible domains are marked by yellow dots.

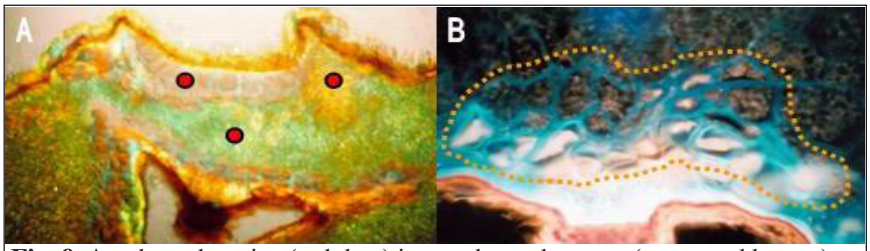

Fig. 9. A – three domains (red dots) in starchy endosperm (green and brown) and in an aleurone layer (pink) in a *T. turgidum* subsp. *dicoccum*/*Ae. tauschii* amphiploid. B – a thick-walled domain (outlined) of the callus-like aleurone layer in a *T. timopheevii* subsp. *timopheevii*/*Ae. umbellulata* amphiploid.

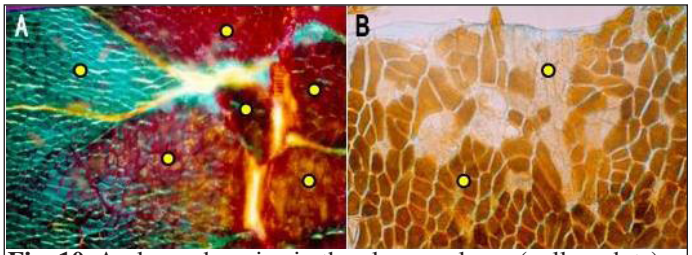

Fig. 10. A - large domains in the aleurone layer (yellow dots) with distinct developmentally disturbed borders between them in a *T. turgidum* subsp. *orientale*/*Ae. tauschi*i amphiploid- B – large starch domains (lighter) in the aleurone layer (brown) in an *Elymus canadensis*/*Pseudoroegneria libanotica* amphiploid.

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Endospermal domains in a 'Triticum timopheevii subsp. timopheevii / Aegilops umbellulata' amphiploid.

R. Kosina, A. Koźlik, and K. Markowska.

The following information is part of an MSc thesis written by Koźlik (2013).

The clonal substructure of grass endosperm has been documented already (Kosina 1992, 2012), and can be realized in different ways, e.g., by changes of metabolism or growth. In the above amphiploid, an intrusive growth of aleurone cells into starchy endosperm divides

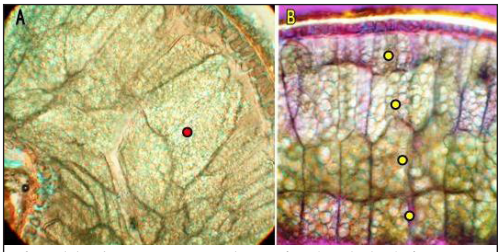

Fig. 11. A – a mega-domain (red dot) in starchy endosperm composed of highly polyploidized cells in an *Avena barbata*/*A. nuda* amphiploid. B – horizontal starchy domains created by simultaneous periclinal cytokineses in *Lolium temulentum* (yellow dots).

this tissue into several domains (Fig. 12A). These domains are composed either of aleurone and starchy cells, or, within the aleurone layer, only a starchy phenotype is expressed (Fig. 12B). In intrusively growing aleurone cells, all the assimilates support synthesis of the components of cell walls, cellulose and hemicelluloses, and cells do not store any protein (Fig. 12B). Such aleurone cells also can be treated as a domain presenting changed metabolism and growth. The domainal bordering role of aleurone cells also is exemplified (Fig. 12C), where a group of starchy cells is separated by aleurone cells surrounding them. Two special domains have been recognized in *Ae. umbellulata*, a paternal species of the amphiploid (Fig. 12D). The first change concerns the expression of a starchy phenotype instead of the aleurone one. The second change relates to starch metabolism; there are two starch grain phenotypes in adjacent cells, large and small. If we accept a sister origin for both types of cells, then we can conclude that this change is caused by somatic (mitotic) crossing-over. These two types of cells can also be of different origin, they will then be recognized as two various clones. The process of somatic crossing-over has often been documented in grasses (Kosina 2007).

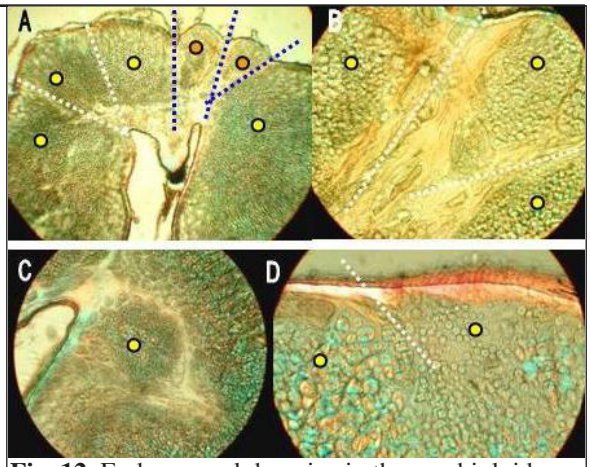

Fig. 12. Endospermal domains in the amphiploid (A,B,C) and its paternal species, *Ae. umbellulata* (D). Dotted lines and dots separate and mark the domains. A – two domains (brown dots) presenting the expression of a starch phenotype in the aleurone layer.

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Koźlik A. 2013. Zmienność mikrostrukturalna owocu amfiploida *Triticum timopheevii* x *Aegilops umbellulata*. MSc Thesis, Institute of Experimental Biology, University of Wroclaw, Wroclaw (In Polish).

Variability in the endosperm cavity in a 'Triticum timopheevii subsp. timopheevii / Aegilops umbellulata' amphiploid.

R. Kosina, A. Koźlik, and K. Markowska.

Kosina (1984) distingushed two main types of endospermal cavity in fossil and contemporary wheats, similar to the letters, T or I. Most often, the cavities are of small volume, but sometimes a large body filled by nucellar apoptotic remnants is formed. Deformed and collapsed cells are often identified just above the nucellar projection. An excess of starch grain is located in the cavity, and it is varietal specific (Kosina et al. 2012). The cavity develops just above the nucellar projection as a symmetric or asymmetric structure, and penetrates the starchy endosperm. In *T. timopheevii* subsp. *timopheevii*, the cavity is narrow and long as the letter I (Fig. 13A); in *Ae. umbellulata*, it is like the letter T or letter I (Fig. 13B). For the amphiploid with native or demethylated genomes, different cavities were documented, and they developed as large symmetric or asymmetric letters T (Fig. 14, p. 119). The asymmetry was sometimes coupled with developmental abnormalities of the nucellar epidermis (Fig. 14C, p. 119) or aleurone and starchy endosperm (Fig. 14D, p. 119). Nucellar epidermis anomalies appeared as its polyploidization and the location of assimilates in its very thick walls (hemicelluloses). The morphology of cavities is uniform in parental species and its variation is greatly increased in the amphiploid.

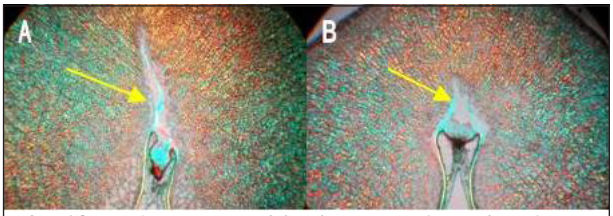

Fig. 13. Endosperm cavities in parental species: A – *T. timopheevii* subsp. *timopheevii*; B – *Ae. umbellulata*.

References.
Kosina R. 1984. Morphology of the crease of wheat caryopsis and its usability for identification of some species – a numerical approach. *In:* Plants and Ancient Man (van Zeist W and Casparie WA, Eds). A.A. Balkema, Rotterdam, pp 177-191.
Kosina R, Tomaszewska P, and Kamińska K. 2012. On caryopsis crease and endosperm cavity in wheat and *Brachypodium distachyon*. Ann Wheat Newslet 58:196-197.
Koźlik A. 2013. Zmienność mikrostrukturalna owocu amfiploida *Triticum timopheevii* x *Aegilops umbellulata*. MSc Thesis, Institute of Experimental Biology, University of Wroclaw, Wroclaw (In Polish).

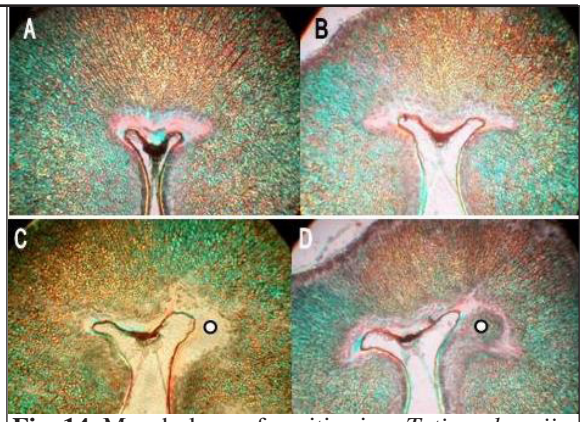

Fig. 14. Morphology of cavities in a *T. timopheevii* subsp. *timopheevii*/*Ae. umbellulata*. amphiploid. In C and D, white dots mark asymmetric cavities filled by nucellar remnants and by a starch-aleurone body, respectively.

Asymmetry of aleurone layer development in a 'Triticum timopheevii subsp. timopheevii / Aegilops umbellulata' amphiploid.

R. Kosina, A. Koźlik, and K. Markowska.

The final periclinal cell divisions in the outer layer of young starchy endosperm provide a new aleurone cell phenotype. In wheat and rye, this layer is one-celled, but in barley and, e.g. in *Thinpyrum distichum*, it is 2-4-celled (Kosina 2012a) and in *Brachypodium distachyon* it is 1-4-celled. Around nearly all the caryopsis the aleurone layer is uniform, but in the area of the crease, the cells have a different morphology. Aleurone cells covering an embryo are of a different structure and function. In the amphiploid, the aleurone layer is structurally more variable than in parental species. The layer in the dorsal part of the caryopsis has cells of different size and shape, sometimes intrusively elongated into starchy endosperm. Periclinal divisions locally increase the number of cells within the layer. Callus-like assemblages of aleurone cells are created by multi-directional cytokineses. In the amphiploid, we documented rare developmental events in the layer. In two halves of the same caryopsis, the following developmental patterns of aleurone cells occurred on both sides of the crease:

- cells grow tangentially, without expected cytokineses and do not grow radially; when looking at the surface of the layer, we can see a set of large cells (Fig. 15A).
- cells frequently divide anticlinally and their tangential growth is not observed. A group of small cells is visible when looking at the surface (Fig. 15B).
- development of the aleurone layer is similar to that in Fig. 15A (Fig. 15C)
- development of the aleurone layer is similar to that in Fig. 15B, but with radial growth of aleurone cells towards the starchy endosperm (Fig. 15D).

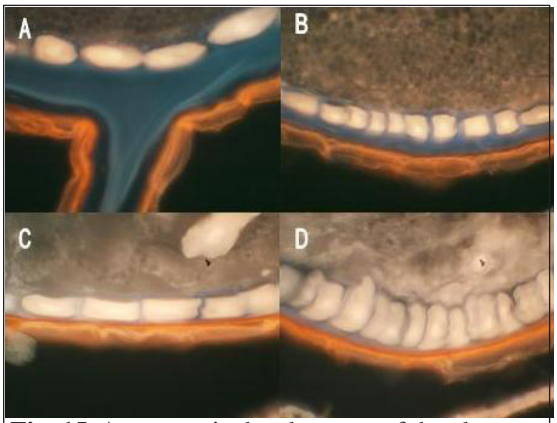

Fig. 15. Asymmetric development of the aleurone layer in two (A, B and C, D) caryopses.

These examples provide evidence that, within the aleurone layer, selected groups of cells develop independently, having specific cell cycles and types of growth. They appear in the form of a mosaic. The larger the segment of cells, the older it is and vice versa, and this depends on the clonal behaviour of endosperm (Kosina 2012a). Such phenomena prove the domainal structure of this tissue, especially in hybrid forms (Kosina 2007, Kosina 2012b, Koźlik 2013).

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A high protein subaleurone layer in a 'Triticum timopheevii subsp. timopheevii / Aegilops umbellulata' amphiploid.

R. Kosina, A. Koźlik, and K. Markowska.

Development of a high-protein (HP) subaleurone layer has been documented in many grasses, including cereals; the variation in wheats was studied by Kosina (1988). Kosina and Tomaszewska (2012) reported on the variability of the layer in some 'wheat/*Thinopyrum distichum*' amphiploids. Koźlik (2013) documented such a layer in a '*Triticum*/*Aegilops*' amphiploid. In *T. timopheevii* subsp. *timopheevii*, the HP layer is poorly developed, but in *Ae. umbellulata*, it is created by 1-2 rows of cells with a large amount of protein and with few starch grains. Despite the highly diploidized nature of the amphiploid and the expected small variation, its HP layer is very variable. The layer can be poor, expressed in the form of scattered small cells (Fig. 16A) or rich and uniform with a large amount of protein (Fig. 16B). The layer is often composed of several rows of cells or isolated callus-like groups of cells. The HP layer is especially thick in the lateral sides of caryopsis, adjacent to the crease. In the study, we also noted a mega domain of the HP layer covering half of the caryopsis. The development of the subaleurone layer may be further complicated by the creation of highly polyploidized cell clones assisted by apoptosis of adjacent starch tissue, such as can be noted in *Brachypodium distachyon* (Kosina et al. 2012).

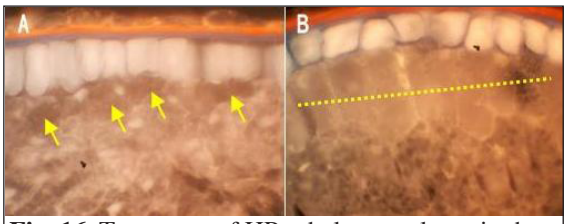

Fig. 16. Two types of HP subaleurone layer in the amphiploid with native genomes.

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Kosina R, Tomaszewska P, and Kamińska K. 2012. On caryopsis developmental events in wheat and *Brachypodium distachyon*. Ann Wheat Newslet 58:197-198.

Koźlik A. 2013. Zmienność mikrostrukturalna owocu amfiploida *Triticum timopheevi*i x *Aegilops umbellulata*. MSc Thesis, Institute of Experimental Biology, University of Wroclaw, Wroclaw (In Polish).

Numerical taxonomy of a 'Triticum timopheevii subsp. timopheevii / Aegilops umbellulata' amphiploid and its parents.

R. Kosina, A. Koźlik, and K. Markowska.

Plants of the amphiploid and its parents were cultivated in two environments, marked b and d in the diagram (Fig. 17). The amphiploid was studied in two forms, with native and demethylated genomes (Koźlik 2013). Caryopses of the amphiploid and its parental species were evaluated by twelve characteristics. The mean taxonomic distance was calculated between each pair of OTUs (operational taxonomic units). The matrix of taxonomic distances was used in a non-metric multidimensional scaling to put OTUs within an ordination space (axes x, y, and z). The group of amphiploid forms is strictly intermediate between parents and their distance in this minimum spanning tree is 0.93. The intraspecific

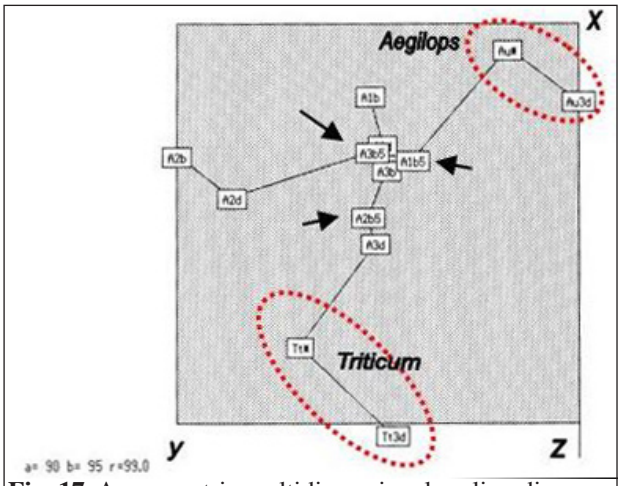

Fig. 17. A non-metric multidimensional scaling diagram with *Triticum*, *Aegilops*, and an amphiploid OTUs described by twelve characteristics of caryopsis.

variation of two *Aegilops* accessions is larger (a distance of 1.36) than that of two accessions of *Triticum*, which have a distance of 0.87. All the amphiploid forms are characterized by a range of distances from 0.07 to 0.91. The demethylated forms are situated in the centre of the diagram (arrows). No significant distance is noted for both types of amphiploid. A study of character covariation proved that demethylation shows not significant positive interaction with the development of starch cells instead of the protein phenotype in the aleurone layer, and with the thickness of the aleurone layer in the dorsal part of caryopsis. This study adds new data on the interparental variation of hybrid progeny and its expression in changing genomes (Kosina 1996, Tomaszewska and Kosina 2013).

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Notes on Brachypodium distachyon cytogenetics.

R. Kosina, K. Kamińska, and P. Tomaszewska.

The cytogenetic status of *B. distachyo*n is not simple. A unique variation exists in the species regarding the number of chromosomes in the intra- and interpopulational space (Jaroszewicz et al. 2012). This variation is caused by multiple cytogenetic anomalies in the form of bridges, rings and laggards (Kosina and Kłyk 2011a). Bridge behavior determines increased nucleolar variability (Kosina and Kłyk 2011b).

Metaphases (Fig. 18A, B, and C) show that the chromosome numbers in the species are from 2n = 10 to 2n = 60, from diploid (2x) to dodecaploid (12x). Also, some aneuploid numbers are common. In the literature, some basic numbers are reported: 10, 20 and 30. If we look at the chasmogamic breeding system found in all studied accessions (Kosina and Tomaszewska 2012), the species is open to hybridization, and a new cytogenetic variation can be created. Dodecaploids (Fig. 18B) are formed after an endomitotic process. Bimodality related to chromosome length is confirmed here (Fig. 18A). The side-by-side separation of short and long chromosomes, frequently observed in metaphases (Fig. 18A), proves that both chromosomal sets can be of different species origin, and their expression can be possible without suppression of any loci.

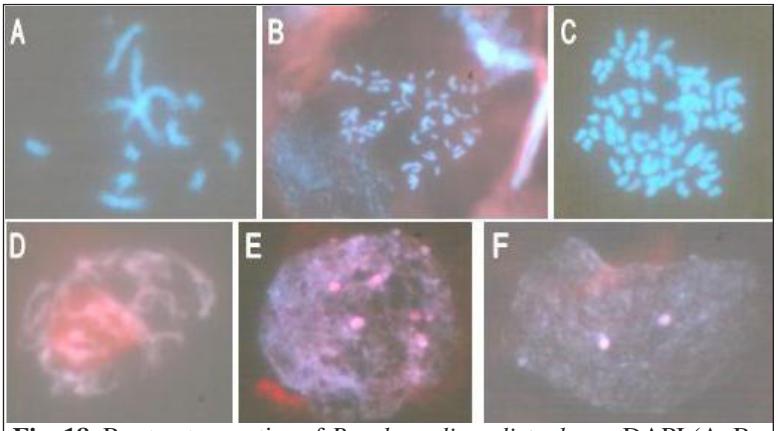

Fig. 18. Root cytogenetics of *Brachypodium distachyon*. DAPI (A, B, and C) and DAPI-PI (D, E, and F) staining. A, 2n = 12; B, 2n = ~60; and C, 2n = 53. D–F, red staining of RNA.

The activity of rDNA loci can be measured by identification of associated proteins using the Ag-NOR method (Kosina and Kłyk 2011b) or by localization of RNA in NORs or outside these loci, e.g. 5S rDNA, in the DAPI – propidium iodide (PI) technique. The latter method involves DAPI staining first and then PI (Fig. 18D, E, and F). The red RNA is discriminated as one large body associated with a nucleolus in a late prophase (Fig. 18D) and such a picture dominates in the studied material – one large nucleolus is common in the DAPI stained chromatin. In earlier prophases two 'RNA loci' on one pair of SAT-chromosomes (Fig. 18F) are identified. The RNA loci are also seen on several pairs of chromosomes (Fig. 18E). The DAPI-PI method also showed the location of RNA loci on bridges.

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Intraspecific variation of caryopsis microstructure in Brachypodium distachyon.

R. Kosina and K. Kamińska.

In *Brachypodium distachyon*, a large variation of gross morphology has been documented (Jaroszewicz et al. 2012). Many of its traits can be considered as weedy (Kosina et al. 2011). The caryopsis structure is related to that in wheat (Kosina and Tomaszewska 2011, Kosina et al. 2012a,b).

Caryopses of the species (22 accessions) were evaluated by 15 microstructural characters. Plants were cultivated for three years (2004, 2006, and 2009), and OTUs in the diagram are marked with one, two, or three dots, respectively. The accessions are species units from a broad geographical range. The analyzed characters were as follows: width and height of caryopsis on cross-section, cell number between ventral and dorsal parts of caryopsis, number of single periclinal divisions in a 10-celled aleurone dorsal segment, number of double periclinal divisions in a 10-celled aleurone dorsal segment, a high protein subaleurone layer (+, –), grain filling by starch cells (+, –), thickness of nucellar epidermis, width and height of pigment strand, height of nucellar projection, thickness of dorsal aleurone layer, thickness of anticlinal cell walls in starchy endosperm, starch cells adjacent to nucellar projection (+, –), amd cylindrical cells in the middle part of caryopsis (+, –).

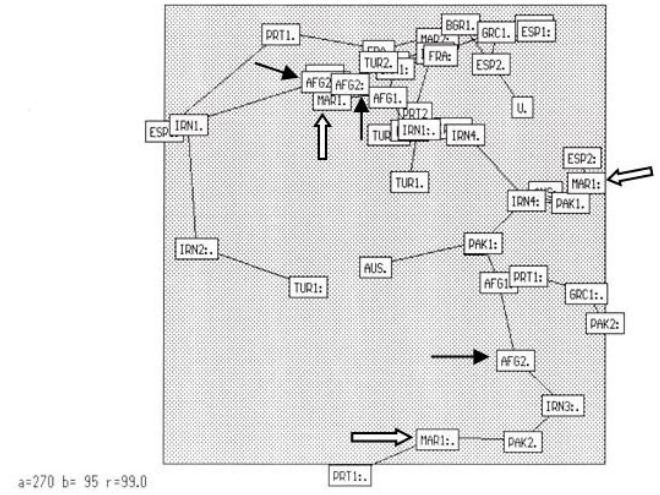

Fig. 19. An nmMDS and MST (non-metric multidimensional scaling and minimum spanning tree) diagram for different accessions of *Brachypodium distachyon* from a geographical range between Portugal (PRT) and Afghanistan (AFG).

Numerical taxonomy of multivariate units (OTUs) proved that intraspecific relationships between them vary in different years. We noted only a partial geographical relationship for some accessions. The individual accessions show different seasonal variation. Two examples, MAR1 from Morocco and AFG2 from Afghanistan, are provided (Fig. 19). Three Moroccan units are more distant from each other than is the case in Afghanistan. These data prove that an ample variation exists in the species and interactions within it are worthy of further research.

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Nucellar projection types in Brachypodium distachyon.

R. Kosina and K. Kamińska.

In a crease of wheat caryopsis, we recognized various tissues, such as parenchyma of the pericarp, phloem-xylem bundles, zig-zag puzzle cells of the pigment strand, transfer cells of the nucellar projection, an apoptotic body of the endosperm cavity, and specific cells of the aleurone layer. Association and interaction of these cells and tissues are based on the broad variation of microstructures (Kosina 1984, 1995). These crease structures determine the nutritional value of the caryopsis and the starch/protein ratio in it (Kosina 1988). The crease structure is highly variable in *Avena* amphiploids (Kosina and Tomaszewska 2011a). In the set of tissues, the nucellar projection plays a very important role. Koźlik (2013) discovered in a '*Triticum*/*Aegilops*' amphiploid that this projection has an intermediate morphology to parents, and is even transgressive to paternal species. In *B. distachyon*, some data related to microstructure of the crease region have been presented by Kosina and Tomaszewska (2011b) and Kosina et al. (2012a,b).

Analyses of the nucellar projection were made under epifluorescence and polarizing microscopes. A blue cellulose autofluorescence was stronger in thick, transfer walls in cells adjacent to the pigment strand. A part of the nucellar projection close to starch endosperm presents patterns of correlated development of the projection and starch tissue, open like a fan or collapsed. Our structural data prove that the size and shape of the nucellar projection depend on nucellar tissue preserved between the chalaza and enlarging embryo sac. In *B. distachyon*, we determined three basic types of nucellar projections (Fig. 20)

Fig. 20. Basic types of nucellar projection morphology in caryopses of *Brachypodium distachyon*.

- a high projection, elongated between the pigment strand and starchy endosperm (This structure is composed of elongated cells. Such a structure promotes the transport of assimilates through relatively few cell walls and plasma membranes (Fig. 20A)),
- an intermediate structure with shorter and multi-directionally oriented cells (Fig. 20B), and
- a low projection formed by many small cells, such as bricks in the wall (Fig. 20C, Here, the assimilates must flow through more cell walls and plasma membranes).

The two extreme types of nucellar projection differ in the number of cell divisions and cell growth. Anticlinal cytokineses and elongated growth dominate in the first type, and periclinal divisions and tangential growth dominate in the third type.

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Kosina R, Tomaszewska P, and Kamińska K. 2012a. On caryopsis crease and endosperm cavity in wheat and *Brachypodium distachyon*. Ann Wheat Newslet 58:196-197.

Kosina R, Tomaszewska P, and Kamińska K. 2012b. On caryopsis developmental events in wheat and *Brachypodium distachyon*. Ann Wheat Newslet 58:197-198.

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The role of nucellar epidermis during the germination of Brachypodium distachyon.

R. Kosina and K. Kamińska.

In cereal grains, the nucellar epidermis is preserved as a very thin, light layer. It is rather acellular, cell lumina can be rarely seen. In wheat octoploids, remnants of this epidermis create a thicker layer (Kosina and Bureś 2011). In wild grasses, the nucellar epidermis is thick, but simultaneously their starchy endosperm is poorer than in cereals. One can recognize two developmental paths and two directions of assimilate storage by

starchy and aleurone endosperm develops via metabolism of assimilates and nucellar apoptotic products and

assimilates and nucellar apoptotic products, the latter only in part, excluding the nucellar epidermis, are used for endosperm and nucellar epidermis development and differentiation.

The second path was recognized in *B. distachyon*. In this species, the nucellar epidermis is very thick (Kosina and Tomaszewska 2011), but also interaccessional variation is noted (Kosina and Jaroszewicz 2007; Jaroszewicz et al. 2012). The same interaccessional (interpopulational) variation is exhibited by a perennial species, *B. sylvaticum*. In *B. distachyon*, there is a balance between storage of assimilates in endosperm as starch and protein, and in nucellar tissue as hemicelluloses. The hemicelluloses are mainly stored in tangential walls of epidermis cells (Fig. 21A).

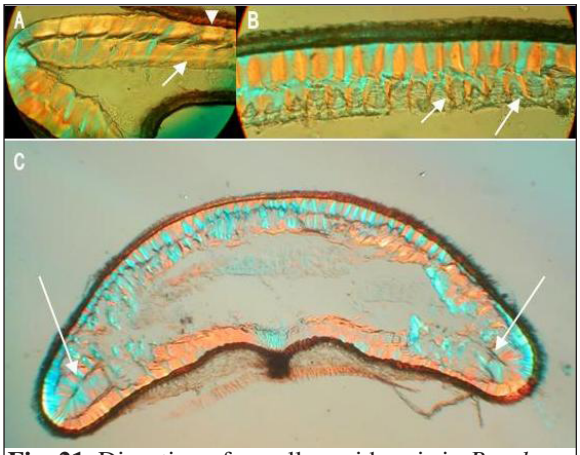

Fig. 21. Digestion of nucellar epidermis in *Brachypodium distachyon* under germination (polarized microscope). A – arrows show thick tangential, hemicellulosic walls in epidermis cells, B – arrows show anticlinal, not digested more cellulosic walls, C – arrows show undigested closed up lateral parts, when starchy endosperm is not fully developed.

Germination of unripe and ripe caryopses of *B. distachyon* was studied over three periods, after 3, 7, and 14 days. Starchy endosperm and aleurone protein are digested first; however, the integrity of the aleurone cells is long maintained. The nucellar epidermis is digested first in the dorsal part (Fig. 21B,C) and this relates to internal tangential walls. Anticlinal walls have more cellulose and they are more persistent (Fig. 21B). In the lateral parts of caryopsis, which can be closed up (see Fig. 21B), when there is some scarcity of starchy endosperm, hemicelluloses are digested with more difficulty. Probably, due to the lower activity of the aleurone layer in the crease area, the nucellar epidermis there is slowly digested. The data show that, in grasses, there are two strategies for the storage of assimilates, germination of diaspores, and seedling growth.

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Jaroszewicz A, Kosina R, and Stankiewicz P. 2012. RAPD, karyology and selected morphological variation in a model grass, *Brachypodium distachyon* (L.) Beauv. Weed Res 52:204-216.

Structural types of endosperm in Brachypodium distachyon.

R. Kosina and K. Kamińska.

The endosperm starts to develop from nuclei scattered inside an embryo sac, contiguous to its wall. Alveolar units are pushed to the center by successive periclinal cytokineses. Finally, these units fill the endospermal space in the form of clonal pyramids. This has been well exemplified for *Thinopyrum distichum* (Kosina 2012). In *B. distachyon*, regular columns of starch cells develop between dorsal and ventral parts of caryopsis (Kosina and Tomaszewska 2011). Such a structure is determined by cell divisions and their growth. Here, one can recognize three types of cytokineses: periclinal,

Fig. 22. Architecture of starchy endosperm in *Brachypodium distachyon*. A – cylindrical endosperm, B – isodiametric endosperm, C – poorly developed endosperm with a changed ratio between starch and protein levels.

anticlinal and diagonal, and three types of cell growth: tangential, radial and intrusive. These divisions and growth create two main patterns of endosperm in cereals: cylindrical and isodiametric. The first type is characteristic for the central part of caryopsis, the second is in lateral parts. However, we found an intraspecific variability of endosperm architecture in B. distachyon. The cylindrical or isodiametric cell arrangement was observed in the whole volume of endosperm. In the cylindrical endosperm (Fig. 22A), a few periclinal cytokineses and radial cell growth dominate. Sometimes, an intrusive growth formed very long and narrow starch cells. In the isodiametric endosperm (Fig. 22B), multiple periclinal and diagonal cytokineses are associated with poor or a weak radial cell growth. Isodiametric endosperm is also formed in plants with poor assimilation, and then starch synthesis in endosperm is suppressed. These two processes, assimilation and starch synthesis, can be independent. Such anomalous endosperm is composed of irregular cells with a small amount of starch and increased protein levels (see the pink fluorescence of protein) (Fig. 22C). The data show that an unknown, new type of endosperm variability is detected in *B. distachyon*.

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Variability of caryopsis micromorphology in Brachypodium distachyon.

R. Kosina and K. Kamińska.

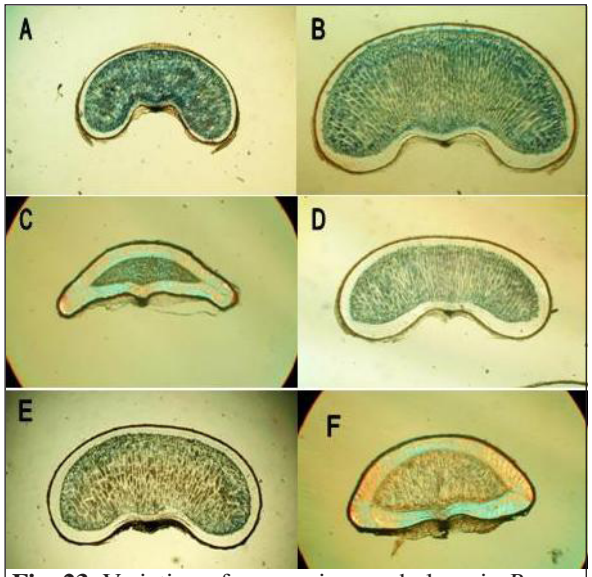

Fig. 23. Variation of caryopsis morphology in *B. distachyon*. A and B – small vs large, C and D – unfilled vs. filled by starch, E and F – variation of a starch/hemicelluloses ratio.

Many microstructural characteristics of caryopsis are linked with ploidy levels, and this has been well exemplified in wheat by Kosina (1984, 1995, 1999). Ample cytogenetic variation in *B. distachyon* (Jaroszewicz et al. 2012) is the basis for ploidal differentiation. So, one can expect a similar variability of micromorphology as in wheats. This study was conducted on caryopsis cross-sections for 22 accessions of *B. distachyon*. Small and large caryopses (Fig. 23A and B) are also differentiated by ploidy levels. The volume of endosperm also shows the difference between wild and cultivated grasses. This difference can also be discussed within the scope of the weediness of the species (Kosina et al. 2011). The difference in starch synthesis appeared for unfilled and filled caryopses (Fig. 23C and D). As an annual and wild species, *B. distachyon* locates assimilates into nucellar tissue (hemicelluloses in epidermis) and in endosperm (starch and protein). These syntheses are not simultaneous. The possible sequence of syntheses could be as follows: hemicelluloses in nucellar cell walls – starch – aleurone protein. A difference in a ratio between hemicelluloses and starch synthesis is shown (Fig. 23E and F). These 'hemicellulosic' or 'starchy' caryopses add new components to intraspecific variability. The cross-section types of caryopses show that this variation creates the possibility of the adaptation of the species to different climates and disturbed habitats, which could be effected by differentiation of the competitive ability of seedlings using caryopsis energy resources.

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Jaroszewicz A, Kosina R, and Stankiewicz P. 2012. RAPD, karyology and selected morphological variation in a model grass, *Brachypodium distachyon* (L.) Beauv. Weed Res 52:204–216.

Aleurone starch domains in Brachypodium distachyon.

R. Kosina and K. Kamińska.

Kosina (2007) presented a review of mosaic structures in grasses. Mosaics can be expressed in various plant organs and structures, and also in caryopsis. There are two main causes of this phenomenon: the activity of transposons and somatic crossing-over. Within the aleurone layer, two phenotypes are expressed, mainly aleurone cells in the outer layer of endosperm and, rarely, starch cells situated between the aleurone ones. Examples of such a phenomenon have been found in the Triticeae tribe (Kosina and Tomaszewska 2010, Kosina and Zając 2010), in the genus *Avena* (Kosina and Tomaszewska 2011) and in *B. distachyon* (Kosina et al. 2012). Most often, this change of phenotype occurs in plants of hybrid origin. In the 22 accessions of *B. distachyon* studied, we found such a change to be too frequent to be random. Mosaics rarely occurred in the dorsal part of caryopsis, in the form of single starch cells (Fig. 24D) and frequently in the area adjacent to the nucellar projection. Many scattered starch cells were noted, sometimes with thick walls (Fig. 24A), and often there were large starch segments composed of many cells (Fig. 24B and C), sometimes with increased levels of protein (Fig. 24C). This original development is probably dependent on several factors; spatial relationships between the chalazal part of the nucellus and the enlarging embryo sac, conductive ability of transfer tissues in the crease, and the total time available for supply of assimilates.

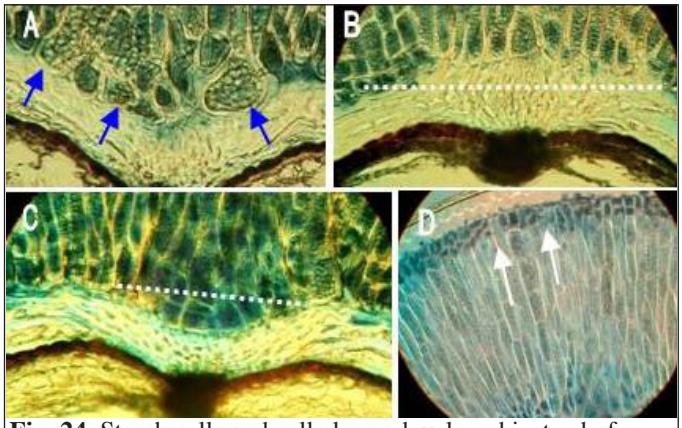

Fig. 24. Starch cells and cell clones developed instead of an aleurone layer in a crease region in *B. distachyon*. A – a group of scattered cells contiguous to the nucellar projection (blue arrows), B – a large segment of starch cells close to the nucellar projection (blue dotted line), C - a large segment of starch cells close to the nucellar projection (dotted line) with increased levels of protein, D – two starch cells in the dorsal aleurone layer (arrows).

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Expression of aleurone starch phenotypes in cereal endosperm.

R. Kosina and A. Koźlik.

Commonly, the development of endosperm is finished by periclinal cytokineses separating the cells of the aleurone layer. The layer is uniform, especially in the dorsal part of caryopsis (Kosina 1992, 2012). The layer is a one- or 2-4-celled structure. A mosaic of aleurone – starch cells has been described in the layer (Kosina 2007). Depending on tissue spatial relationships in endosperm and on its clonal nature, the aleurone cells can develop according to a callus pattern (Fig. 25A). An amphiploid Triticum timopheevii / Aegilops umbellulata will serve here to show various paths of expression of the starch phenotype within the aleurone layer (Koźlik 2013). In Fig. 25B,C,D, the disappearing aleurone cells are presented (see arrows). Such a phenomenon relates to the amphiploid with native genomes (Fig. 25B,C) as well as that with demethylated genomes (Fig. 25D). These pictures prove that the aleurone cells can be apoptotically eliminated during endosperm development – first by digestion of their aleurone protein (Fig. 25B,D) and secondly by digestion of their walls (Fig. 25C). The sites released by them are occupied by cells of starchy endosperm which are additionally developmentally enhanced by apoptotic metabolites. In the second path of starch cell occurrence in the aleurone layer, there is a change of phenotype expression after the last periclinal cytokinesis. Therefore, mistakes in the interpretation of the nature of the starch-aleurone cells are possible. The variability presented here probably exists in other grasses.

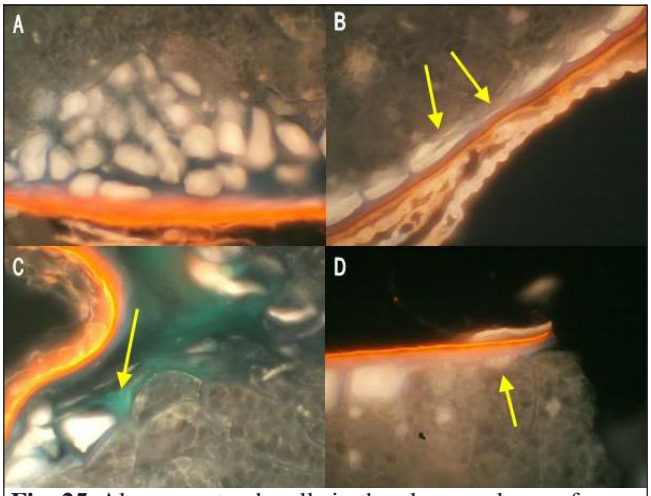

Fig. 25. Aleurone starch cells in the aleurone layer of a *Triticum timopheevii/Aegilops umbellulata* amphiploid. Natural fluorescence with the use of a triple-band filter under an epifluorescence microscope. A–C, the amphiploid with native genomes; D, with demethylated genomes. A, a callus-like aleurone layer; B, apoptotically changed flat aleurone cells; C, wall remnants of an aleurone cell contiguous to a starch cell, on the right; and D, remnants of an aleurone protein in the digested part of the aleurone layer.

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ITEMS FROM ROMANIA

AGRICULTURAL RESEARCH & DEVELOPMENT STATION — S.C.D.A.
401100, Turda, Agriculturii street 27, Jud. Cluj, Romania.

'ANDRADA' - a new winter wheat cultivar.

V. Moldovan and Rozalia Kadar.

Andrada, a hard red winter wheat cultivar (*Triticum aestivum* L. subsp. *aestivum* var. *Ferrugineum*) developed by the Agricultural Research & Development Station of Turda, was released in 2012 because of its high yield associated with enhanced bread-making quality. Andrada was selected from the cross 'Dropia/T57-90' using a pedigree-selection method. The single cross between Dropia and T57-90 was made in 2001. The main aim of this cross was to obtain a combination between the superior bread-making quality of the cultivar Dropia with the high-yielding capacity from our breeding line T57-90. Following our breeding procedure that was presented previously (Ann Wheat Newslet 48:113-115), individual selection started in the F_2 generation and, after subsequent reselection, we obtained the line T150-03 that was advanced to the Official Yield Trials at the State Institute for Variety Testing and Registration (ISTIS) in the autumn of 2008. After 3 years of evaluation (2009–11) in eight locations, T150-03 was registered under the name 'Andrada' and released to growers due to its good yield performance and broad adaptation to Transilvania environments as well as improved bread-making quality.

Andrada is an awned, red-spiked, semidwarf wheat. Juvenile growth is semierect. Foliage is green at the boot stage. Plant height (82 cm) is similar to that of Dropia, and 5-10 cm shorter than that of T57-90. Spikes are awned and lax with red glumes. Kernels are red, ovate, with a medium-sized germ; the kernel crease is medium wide and medium deep, with rounded cheeks. The kernel size is quite large; 1,000-kernel weight averagex 47 g and has a quiet good test weight (volume weight) at 77 kg/hectoliter.

Andrada is medium-early in maturity (267 days), similar to that of Dropia. The winterhardiness of Andrada is adequate for most Transilvania growing conditions. Andrada has excellent straw strength, which confers good lodging resistance.

For disease resistance, Andrada is moderately resistant to yellow rust and powdery mildew, but is moderately susceptible to leaf rust. Andrada also showed moderate resistance to Fusarium head blight.

Andrada has shown good yield performance in most of official test sites (ISTIS). Averaged across three years (2009–11) and eight locations (24 location-years), Andrada did not differ widely in grain yield from that of the highest yielding entry in the trials. The average grain yield of Andrada in 2011 was 7,011 kg/ha, which is 8% higher than check cultivar Delabrad 2. The maximum grain yield of Andrada was 8,016 kg/ha, obtained at Center for Testing Varieties (CTS), Targu-Secuiesc.

Andrada meets domestic quality criteria for high-quality bread flour production. The quality characteristics of Andrada are reflected by a grain protein content up to 12.73% associated with an excellent glutenic index (76–100) and loaf volume (520 cm^3). Andrada can be classified in the B1–A2 quality wheat. Breeder and foundation seed of Andrada will be maintained by the Agricultural Research & Development Station Turda.

ITEMS FROM THE RUSSIAN FEDERATION

AGRICULTURAL RESEARCH INSTITUTE FOR THE SOUTH-EAST REGIONS
Department of Genetics, Laboratory of Genetics and Cytology, 7 Toulaikov St., Saratov, 410010, Russian Federation.

Evaluating spring bread wheat introgression lines of the Genetics and Cytology Laboratory at ARISER for resistance to stem rust race Ug99 + Sr24 (TTKST).

S.N. Sibikeev and A.E. Druzhin.

In 2013, a set of introgression lines was evaluated for resistance to stem rust race Ug99 + *Sr24* (TTKST) in the experimental fields at the Kenyan Agricultural Research Insititute, Njoro, Kenya, under natural epidemic conditions of the pathogens. One part of this material has been repeatedly screened; the other part has passed an initial screening. Among the 20 samples from repeated screening, 12 had a resistant type of reaction. These resistant lines were NILs with the translocation combinations *Lr19*/*Sr25*+*Lr24*/*Sr24* and *Lr19*/*Sr25*+*Lr26*/*Sr31*, and also lines with translocations from durum wheat and *Ae. speltoides*. Among the lines from the initial screening, except for lines with the above-stated gene combinations, resistant lines with 6Agi (6D) substitutions and 6Agi (6D) + *Lr26*/*Sr31* and *Lr19*/*Sr25* + *Lr37*/*Sr38* combinations were detected.

Destruction of winter wheat cultivarst by viral diseases in the Lower Volga Region.

T.S. Markelova and E.A. Baukenova.

Lately, the spread of viral diseased to wheat is increasing in the Lower Volga Region, which is characterized by unusual injuries, especially in epidemics. The most common viral disease is winter wheat mosaic virus (WWMV), which damages both spring and winter wheat, rye, barley, panic grass, oats, and many other wild cereals. The symptoms of disease become visible in the autumn. Chlorotic spots appear on the leaves of damaged plants, which become yellow after time and then merge into lengthwise stripes. Winter wheat is bushing out redundantly with rosette. The most vivid symptoms are seen at the end of May and the beginning of June at spike formation stage as different degrees of bushiness, lagging growth, and some plant death.

The virus is vectored by the striped (*Psammotettix striatus* Fall.) and six-point (*Macrosfeles laevis* L.) leafhoppers. Both types overwinter in the egg stage on newly sown winter wheat. In autumn, especially in dry weather, the of young, growing wheat leaves are inhabited by many types of leafhoppers; 40–50% of the general quantity. The probability of plant infection increases during a long, warm autumn, because species containing the virus often change place while feeding. In such cases, the loss of harvest from WWMV can reach 15–20%.

In 2010–12, we assessed the spread of winter wheat in the Lower Volga Region damaged by viral diseases. The results showed that highly resistant cultivars were not damaged by viral disease. The degree of damage depended on infection background strength. Saratovskaya 8, Saratovskaya 90, Levoberezhnaya 1, and Donskaya Awnless had a high percent damage (25–30% and more) due to viral disease spread; Lutescense 230, Guberniya, Mironovskaya 808, and Pearl of Volga Region were stable in 2010 and 2011 with moderate damage. In those years, damage did not exceed 5–10%. Victoria 95 and Smuglyanka were resistant during three years of research; only 1–5% was damaged. These conclusions suggest optimism in the direction of selecting cultivars resistant to viral disease.

Identifying wheat–Aegilops columnaris lines with addition, substitution, and translocation chromosomes.

E.D. Badaeva (Institute of General Genetics, Gubkina St. 3, Moscow), A.S. Rouban (Russian State Agrarian University, Moscow Timiryazev Agricultural Academy, Timiryazevskaya St. 49, Moscow), S.N. Sibikeev, and A.E. Druzhin.

During 2007–12, *Ae. columnaris* (UX, 2n=28) was crossed with the spring bread wheat cultivars Saratovskaya 68, L503, and Dobrynya, and backcrossed 2–4 times with the cultivar recipients. Among the sets of wheat–*Ae. columnaris* lines, we observed lines with club ears, hairy ears, light green plants, reduced plant height, and resistance to leaf rust and powdery mildew. The C-banding pattern of these lines showed the addition lines with 2X and 6X chromosomes; substitution lines with 3U (3D), ? (5D) + ? (6D); monosomic 6X (6D); translocation T4BS-4BL-?L plus two additional 6X chromosomes; and the translocation T1DS-1DL-?L. In the future, studying these lines for agronomic performance and analyzing C-banding patterns will continue.

Effect of drought conditions on the change in wheat loose smut populations.

A.E. Druzhin, S.N. Sibikeev, T.D. Golubeva, and T.V. Kalintseva.

We began to identify wheat loose smut races in the Laboratory of Genetics and Cytology in 2000. For differential fungus races, we used a Canadian test-cultivar set. In 2001, we found that race 18 was the predominant race in the local wheat loose smut populations. Further studies have shown that the local population of loose smut includes six races (Table 1). These races could not be identified from a key of known races. We found that the more virulent races of the pathogen in a population usually appear after dry years. Races I-S36 and I-S66 were identified after 2007, which was characterized as dry, and race XX was identified after the drought of 2010. Analysis of the data indicates that the dry conditions are more favorable for formation of new, more virulent races in the loose smut population.

Table 1. Reaction of different cultivars to loose smut of wheat (S = susceptible).

Cultivar diffrentiator	Year						
	2001	2005	2008	2008	2009	2009	2011
	Race 18	Race I-S60	Race I-S36	Race I-S66	Race I-SZ	Race I-UV2	Race XX
TD-1							
TD-2							
TD-3					S		
TD-4	S	S	S	S	S	S	S
TD-5	S	S	S	S		S	S
TD-6							
TD-7	S	S	S	S	S	S	S
TD-8	S	S	S	S	S	S	S
TD-9	S	S	S	S			S
TD-10							
TD-11							
TD-12				S			S
TD-13	S	S	S	S	S	S	S
TD-14							
TD-15				S		S	S
TD-16			S	S			
TD-17							
TD-18							
TD-19							

VAVILOV SARATOV STATE AGRARIAN UNIVERSITY
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Morphological-anatomical changes in somatic wheat calli in vitro under the effect of bacterial lipopolysaccharide.

O.V. Tkachenko and Yu.V. Lobachev; N.V. Evseeva, L.Yu. Matora, G.L. Burygin, K.I. Minlikayeva, and S.Yu. Shchyogolev (Institute of Biochemistry and Physiology of Plants and Microorganisms, Russian Academy of Sciences, 13 Prospekt Entuziastov, Saratov 410049, Russian Federation); and V.A. Spivak (Chernyshevsky Saratov State University, 83 Ul. Astrakhanskaya, Saratov 410012, Russian Federation).

We examined the effect of the lipopolysaccharide (LPS) of the associative, plant-growth-promoting bacterium *Azospirillum brasilense* Sp245 on the morphogenetic parameters of somatic tissues of spring wheat *in vitro*. For this purpose, we used a genetic model including two near-isogenic lines of wheat cultivar Saratovskaya 29 that differed in the *RhtB1c* gene and had contrasting embryogenic capacities. The work was conducted in accordance with the classic scenario of cellular transformations in a culture *in vitro* of immature (14-day-old) wheat embryos by way of indirect somatic embryogenesis. The control was a Linsmaier–Skoog medium containing 2 mg/L of 2,4-dichlorophenoxyacetic acid (2,4-D). In the experimental treatments, the standard medium, after being autoclaved, received 10 μg/mL of LPS isolated from the bacterial outer membrane. The resultant calli and regenerated plants were analyzed by morphometric and morphological-anatomical analysis. On day 30 of cultivation, analysis of the calli showed that in all experimental treatments, there was active callusing of somatic cells of the immature embryos. The callusing effectiveness was high (close to 100%) and did not depend on the genotype or the presence of LPS in the nutrient medium.

Yet in both lines, the formation of morphogenic calli in the control was less than 20% of the number of explants used. On the whole, the addition of LPS to the medium increased the activity of morphogenetic processes. Particularly noticeable was an increase in the yield of calli with meristematic activity loci in the tall line in the presence of 10 μg/mL of LPS in the nutrient medium. Consequently, the morphogenic potential of the calli of the lines under study leveled off. The yield of regenerated plants was influenced profoundly by the explant genotype. The regeneration capacity of the dwarf line was greater than that of the sister line, a fact evident in the control plants. In the experimental treatments, the yield of regenerated plants increased in the tall line under the effect of LPS. Morphological-anatomical analysis confirmed the positive effect of the *RhtB1c* gene at all stages of tissue culturing *in vitro*. LPS was found to promote the secondary differentiation of callus cells and the formation of morphogenesis loci as early as day 7 of embryo culturing, regardless of the genotype. Meristematic activity was observed mostly in the nodal plate, vascular bundles of the scutellum, and coleoptile of the embryos.

On the basis of this data, we established that the LPS of the associative bacterium *A. brasilense* Sp245 promotes the secondary differentiation and the regeneration capacity of wheat callus cells, thereby improving the effectiveness of culturing of genotypes with low embryogenic potential. A nutrient-medium concentration of LPS of 10 μg/mL has the greatest physiological activity toward callus cells. These results make possible a deeper understanding of the mechanisms of morphogenesis in tissue culture and can be used to increase the embryogenic potential of plant objects, specifically wheat, in an *in vitro* culture.

ITEMS FROM UKRAINE

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The content of free amino acids and water-soluble carbohydrate in leaves of isogenic by VRN gene lines of wheat.

O.A. Avksentyeva, Han Bing, and V.V. Zhmurko.

Abstract. In a three-year experiment, we observed the effects of *VRN* genes in soft wheat on the development rate (shoot–booting period), content, and distribution of free amino acids and water-soluble carbohydrates in isogenic lines of Mironovskaya 808 and Olviya. Slowly developing isolines are characterized by a lesser amount of free amino acids and monosaccharides in the leaves. The correlation index C/N clearly correlates with the rates of the isoline development. We propose that *VRN* genes are involved in synthesis control, composition, and distribution of the basic photoassimilates of soft wheat.

Introduction. The transition of wheat plants from vegetative to generative is a major stage in the ontogeny, determining many economically valuable traits. The key genes in this process are the *VRN* genes, which define the reaction of wheat to vernalization and, thus, determine the rate of development. The reaction to vernalization in wheat is controlled by at least five genes, three of which, *Vrn-A1a, Vrn-B1a,* and *Vrn-D1a*, are localized on chromosomes 5A, 5B, and 5D, respectively. Winter-type plant development is manifested only when all three genes are recessive. If at least one of these genes is dominant, then the plants are spring type. *VRN* genes have been actively researched at the molecular-genetic level (Loukoianov A et al. 2005; Oliver SN et al. 2009) and have been cloned and several allelic variants for wheat described (Preston JC et al. 2009). Genes *Vrn-A1a* and *Vrn-B1a* are transcription factors (Distelfeld A et al. 2009; Trevaskis B 2010). These studies have deepened the notion of genetic and molecular-biological control of wheat development.

The primary products of photosynthesis are soluble carbohydrates and free amino acids. Their content, correlation, and distribution between the main organs, in many ways, define the rate of plant development, productivity, and protein content in grain. At present, nitrogen and carbonic metabolism of cereals is intensively studied, including the distribution of metabolites in plant organs and aspects of their reutilization of photoassimilates during grain formation (Amane M 2011), the connection between photosynthesis and general productivity (Champigny M-L 1995), and the increase of valuable proteins in grain (Marte P et al. 2003). Coruzzi and Bush (2001) have shown that low-molecular-weight nitrogen and carbohydrate compounds in plants in nature are not just metabolites, but signal molecules that activate and/or repress many genes.

Perhaps, the *VRN* genetic system, which determines the rate of plant development, reveals the mediated effects on the content of basic metabolites (photoassimilates) of the whole plant, their distribution to the organs of the main shoot, and the correlation of carbohydrate and nitrogenous metabolites. Our investigation researched the content and distribution, by plant organ, of the carbohydrates and amino acids in connection with the rate of wheat development.

Materials and methods. A proper study of the physical-biochemical process is possible only using near isogenic lines (NILs), which have minimal differences in all the genes besides the marked specific feature. We used NILs that were monogenic dominant for the *VRN* genes of soft wheat, which differed in their rate of development. These lines were created in the background of winter wheat cultivars Mironovskaya 808 (tall) and Olviya (short). Field experiments were made during 2010–12 at experimental plots of the Department of Physiology and Biochemistry, V. N. Karazin Kharkov National University. Observations were made on the duration of the shoot-to-boot period and biochemical analyses of the content of free amino acids in the different plants organs and the fractional composition of reduced saccharides in the leaves during the of booting–flowering period.

Results and discussion. The distribution of free amino acids in the organs of the main shoot in the NILs is at a maximum in the leaves, the main photosynthetic organ. Sufficiently high amine nitrogen content was revealed in forming spikes, which become the major sink during flowering and accept both nitrogenous and carbohydrate photoassimilates. The least amount of free amino acids was in the stem, perhaps because it is mainly for transport does not take part in metabolic processes. Such a distribution of free amino acids by the organs of main spike is typical for tall (Mironivskaya 808) and short (Olviya) NILs. However, we noted that Olviya NILs had a higher content of free amino acids in the forming spikes, which may be explained by its faster development compared to that of the taller Mironovskaya 808.

A clear influence of NIL genotype on the content of free amino acids is revealed only in the leaves. In both cultivars, slow-developing NILs, with *VRN-B1a*, were characterized by the minimum content and quick-developing NILs, with *VRN-A1a* and *VRN-D1a*, were characterized by the maximum.

The level of amino acids in the spikes of slowly developing, *VRN-B1a* of Olviya was higher than that of the spikes of the quicker developing, *VRN-A1a* and *VRN-D1a* Olviya lines (Table 1), which may be connected with a slower amino acid synthesis of protein in the forming grain in slowly developing line compared with rapidly developing lines.

Table 1. The distribution of free amino acids in the organs of the main shoot of *VRN* (dominant gene) NILs of wheat.

Cultivar	Isoline genotype	Amino acid content (mg/g dry mass (mean ± σ)		
		Leaf	Stem	Spike
Mironovskaya 808	*VRN-A1a*	1.39 ± 0.06	0.74 ± 0.06	0.99 ± 0.02
	VRN-B1a	1.21 ± 0.03	0.57 ± 0.02	0.53 ± 0.01
	VRN-D1a	1.32 ± 0.05	0.55 ± 0.07	0.77 ± 0.02
Olviya	*VRN-A1a*	1.37 ± 0.04	0.66 ± 0.07	0.46 ± 0.01
	VRN-B1a	1.08 ± 0.02	0.65 ± 0.04	1.95 ± 0.02
	VRN-D1a	1.34 ± 0.05	0.63 ± 0.02	1.33 ± 0.05

Water-soluble carbohydrates, the main products of photosynthesis, are represented in plants mainly by glucose, fructose, and the disaccharide saccharose. The fractional composition of water-soluble carbohydrates in leaves of isogenic by genes *VRN* lines of wheat (Table 2) showed that monosaccharides predominate and make up 60–80% of the synthesized unstructured carbohydrates in leaves. The content of the different fractions (mono-, oligo-, and total) of water-soluble carbohydrates in leaves of both the tall Mironovskaya 808 and short Olviya were virtually the same. The level of monosaccharides in the leaves correlates with the rate of isoline development. The slow-developing *VRN-B1a* isoline is characterized by a minimal content, and rapidly developing *VRN-A1a* and *VRN-D1a* lines by the maximum. We also found the same results while investigating free amino acids in the leaves (Table 1). The amount of oligosaccharide that is present as the main transport carbohydrate in plants, saccharose, is the minimum in isolines that are the first the flower among isolines of Mironovskaya 808 with *VRN-D1a* and among isolines of Olviya with *VRN-A1a* (Table 2). Maybe, the spikes, which are greater sinks, have faster rates of photoassimilates for synthesis of reserve carbohydrates and proteins during grain formation.

Table 2. The content of water-soluble carbohydrates in leaves isogenic by *VRN* gene in NILs (dominant gene). of wheat.

Cultivar	Isoline genotype	Content of soluble carbohydrates, mg/g dry mass (mean ± σ)		
		Mono-	Oligo-	Total
Mironovskaya 808	*VRN-A1a*	37.6 ± 0.06	15.6 ± 0.06	53.2 ± 0.02
	VRN-B1a	36.5 ± 0.03	15.2 ± 0.02	51.7 ± 0.02
	VRN-D1a	45.3 ± 0.05	8.6 ± 0.07	53.9 ± 0.02
Olviya	*VRN-A1a*	38.0 ± 0.04	13.9 ± 0.07	51.9 ± 0.02
	VRN-B1a	36.2 ± 0.02	19.0 ± 0.04	55.2 ± 0.02
	VRN-D1a	38.7 ± 0.05	16.3 ± 0.02	55.0 ± 0.02

Flowering is the determiner for forming the future yield. Photosynthetic activity and nitrogenous status influence the dry mass and content of protein in grain (Champigny M-L 1995; Lawlor DW 2002). The duration of the shoot-to-flowering period showed that the NILs are clearly divided into slowly and rapidly developing groups (Table 3). Independent from the general duration

Table 3. The duration of the shoot-to-flowering period and the correlation of C/N in leaves of isogenic *VRN* lines of wheat (mean ± σ).

Cultivar	Isoline genotype	Shoot-to-flowering (days)	C/N
Mironovskaya 808	*VRN-A1aB1bD1b*	57 ± 1	38.27 ± 1.2
	VRN-A1bB1aD1b	65 ± 2	42.73 ± 1,8
	VRN-A1bB1bD1a	52 ± 1	40.83 ± 1.6
Olviya	*VRN-A1aB1bD1b*	47 ± 1	37.88 ± 0.9
	VRN-A1bB1aD1b	61 ± 2	51.11 ± 2.1
	VRN-A1bB1bD1a	50 ± 1	41.04 ± 1.5

of the transition time to the generative stage that is defined by genotype, individual *VRN* genes in both cultivars slow down or speed up the isoline development. Slowly developing isolines have the *VRN-AlbBlaDlb* genotype and quickly developing isolines are *VRN-AlaBlbDlb* and *VRN-AlbBlbDla* genotypes. In the field conditions of natural photoperiod at latitude 50° Kharkov, the Mironovskaya 808 NIL with *VRN-AlbBlbDla* was the first to flower followed by the Olviya NIL with *VRN-AlaBlbDld*.

The correlation of carbohydrate and nitrogen levels serves as indirect balance index of the carbohydrate and nitrogen metabolism of plants. We calculated the correlation of carbohydrate and free amino acid content. The correlation index, C/N, in leaves had a maximm value in NILs that were characterized by the longest period of days-to-heading, i.e., the slowly developing NILs, which can be evidence for dependence of carbohydrate and nitrogen metabolism on genotypes of *VRN* genes.

We found out that individual *VNR* genes, and maybe genetic systems in general, which regulate the rate of plant development in wheat, indirectly determine the amount, composition, and distribution between the organs of the main photoassimilates, water-soluble carbohydrates and free amino acids. Slowly developing NILs are characterized by a lower amino acid and monosaccharide content in the leaves during flowering, which possibly can be connected with less productivity of photosynthetic processes in these isolines. The C/N correlation index clearly agrees with the rate of development in the NILs. These effects do not depend on the genotype of the cultivar, but appear in NILs irrespective of their cultivar peculiarities.

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Chemical protection of winter wheat shoots using a presowing seed treatment.

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In the chemical protection of winter wheat, especially at the first stages of organogenesis, a presowing seed treatment is ecologically safe for the environment, technologically easy, and economically profitable. Lately, among the assortment of insecticide treatments, neonycotynoids are obtaining wide application.

Our investigation studied the phytosanitary role of chemical treatment of winter wheat seed with systemic insecticides, especially from the neonycotynoid, group for reducing pest injury, and, to increase grain yield.

All studies were conducted at the Laboratory for Plant \Pproduction and Cultivar Investigations of Plant Production Institute nd. a. V.Ya. Yuriev (Eastern Forest-Steppe of Ukraine) during 2008–12. The soil was a typical medium-humus black earth soil on loess with humus with a plowing layer to 5.4%. Seed of winter bread wheat were pretreated with a tank preparation mixture of insect-fungicid. The active agent, imidaclopryd, was 0.25 kg/t and 0.35 kg/t of seed. Winter wheat was sown at optimal dates with the following rates: 4.0 x 10^6 viable seed/ha on black fallow and 5.0 x 10^6 viable seeds/ha after dried peas. Winter wheat was sown in two blocks, without fertilizer and with and application of organic-mineral fertilizer (humus, 6.7 t/ha of crop rotation area and (NPK) 30–60). The agronomic techniques employed are widely used in the investigated area. Counts for plant damage by the larvae of flies were according to conventional methods.

Results. Insecticide treatments, on the basis of imidaclopryd, was highly effective against intrastalk pests, namely fly larvae (Table 1). Averaged over two years, the effectiveness of a pretreating winter wheat seed with imidaclopryd prior to sowing was 78.3%, including *Oscinella* (Diptera: Chloropidae), 85.2%; *Mayetiola destructor* (Diptera: Cecidomyiidae), 66.7%; and *Phorbia securis* (Diptera: Anthomyiidae), 62.5%.

Table 1. Carbon isotope ratio and carbon concentration in leaves of six cover crops grown in Manhattan, KS, and harvested in 2011 (mean of eight values + SE).

Cover crop	$\delta^{13}C$ (‰)	C (%)
Winter wheat	–27.31±0.15	34.41±2.71
Triticale	–29.02±0.29	29.73±4.95
Oat	–29.07±0.10	31.92±2.05
Austrian winter pea	–29.52±0.65	39.70±0.87
Red clover	–30.48±0.16	41.93±4.64
Alfalfa	–31.35±0.51	39.36±0.74

During the autumn tillering in 2010, solitary damage to plants by flies was observed, so observations were not made at this time. In autumn 2011, a drought resulted in a majority of seed sprouting 30–35 days after sowing. The plants finished the autumn vegetation at the two–three leaf stage. Again, counts for plant damage by pests during this period were not carried out. In the winter wheat fields during autumn tillering in 2012, the dominant flies were *Oscinella spp*. These larvae caused 25.1% shoot damage in the control and 8.8% shoot damage in the (fungicide+insecticide). Chemical treatment of winter wheat seeds with imidaclopryd (0.35 kg/t seeds) has shown to be effective against *Oscinella* larvae (36.4%). The lowest level of damage to the shoots with *P. securis* and *M. destructor* larvae were 0.9% and 0.2%, respectively. According to the 2012 data, imidaclopryd was highly effective against soil pests. The technical effectiveness of the preparation against larvae (Coleoptera: Elateridae) with a dose 0.25 kg active agent/t of seed was 87.5% and 100% at 0.35 kg/t.

Pretreating winter wheat seed prior to sowing with a tank mixture of fungicides and insecticides (on the basis of imidaclopryd) is economical profitable, especially in fertilized blocks (Table 2). Averaged over 2009–12, the increase in the grain yield from applying tank mixtures of fungicides and insecticides (imidaclopryd) was 0.24 t/ha, ranging from 5.25 t/ha (control, without chemical protection) to 5.49 t/ha. Seed pretreatment with this mixture in the block with organic-mineral fertilizers contributed to the increase in grain yield by 33.9% (from 3.63 t/ha, control, without chemical protection and fertilizers, to 5.49 t/ha).

Table 2. Economical effectiveness of pre-sowing seed treatment of winter bread wheat with insecticide–fungicide compositions during autumn tillering in 2008–09.

Variant	Yield capacity (t/ha)				Yield increase (t/ha) from	
	2009	2011	2012	2009–12 average	chemical protection	chemical protection + fertilizers
Control (without fertilizers)	3.18	4.80	2.90	3.63	—	—
Control (organic - mineral block)	5.67	6.75	3.34	5.25	—	—
Fungicide + insecticide with active agent imidaclopryd	6.04	6.90	3.53	5.49	0.24	1.86

ITEMS FROM THE UNITED STATES OF AMERICA

KANSAS

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Water use efficiency of six cover crops.

Oliver W. Freeman and M.B. Kirkham.

Cover crops are crops grown to protect soil from erosion and loss of nutrients by leaching. Cover crops are desirable because they not only hold the soil in place but also are efficient in using water. Little information exists concerning the water use efficiency of different cover crops. One way to determine water use efficiency is to analyze the carbon isotope ratio of the leaves. The number, called the $\delta^{13}C$, represents the difference between the ratio of ^{13}C–^{12}C found in a given sample and the ratio that exists in a standard. The ratio is expressed as a per mill ($^{o}/_{oo}$) deviation from the standard. In plants with the C_3 photosynthetic system, an inverse relationship exists between the carbon isotope ratio and water use efficiency (Kirkham 2011). That is, plants with the least negative value of $\delta^{13}C$ have the highest water use efficiency.

Because no one had determined the water use efficiency of cover crops in Kansas by measuring the carbon isotope ratio, we grew six cover crops and determined their ratios. The crops, all with C_3 photosynthesis, were planted in the fall of 2010 in Manhattan, Kansas. The six crops were winter wheat, triticale, oat, Austrian winter pea (*Pisum sativum* var. *arvense* Poir.), red clover (*Trifolium pratense* L.), and alfalfa (*Medicago sativa* L.). In the spring of 2011, an area 1 m^2 from each plot was harvested. The leaves were ground, and a sample was taken and placed in 120-cm^3 plastic container and submitted to the Stable Isotope Mass Spectrometry Laboratory in the Division of Biology at Kansas State University. The laboratory determined the $\delta^{13}C$ of the different crops as well as the carbon concentration in the leaves (Table 1).

Table 1. The effectiveness of pretreatment of winter bread wheat seed with insecticide (imidaclopryd (0.35kg/t))–fungicide compositions at tillering, average for 2008–09.

Species of fly	Damaged tillers		Technical effectiveness (%)
	Control (fungicide)	Fungicide + insecticide	
Mayetiola destructor	1.5	0.5	66.7
Oscinella spp.	5.4	0.8	85.2
Phorbia securis	0.8	0.3	62.5
Others	0.6	0.2	66.7
Total	8.3	1.8	78.3

The three grain crops (wheat, triticale, and oat) had the least negative $\delta^{13}C$ and the lowest carbon concentration in their leaves. Wheat had the least negative $\delta^{13}C$ of all cover crops, and its value differed from the next value (triticale) by almost 2%. The three legumes (Austrian winter pea, clover, and alfalfa) had the most negative $\delta^{13}C$ and highest carbon concentration in the leaves. Because wheat had the least negative $\delta^{13}C$, the results indicated that it had the highest water use efficiency of the six cover crops studied. Therefore, in addition to being a hardy cover crop to grow during the cold winters in Kansas, winter wheat also appears to have the benefit of having high water use efficiency.

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News.

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Notice of release of KS14WGRC61 Fusarium head blight-resistant wheat germ plasm.

Bernd Friebe, William Bockus, P.D. Chen, L.L. Qi, Joey Cainong, Duane L. Wilson, W. John Raupp, Jesse Poland, Robert L. Bowden, Allan K. Fritz, and Bikram S. Gill.

The Agricultural Research Service, U.S. Department of Agriculture and the Kansas Agricultural Experiment Station announce the release of **KS14WGRC61** hard red winter wheat germ plasm with resistance to Fusarium head blight (FHB) for breeding and experimental purposes. KS14WGGRC61 is derived from the cross TA5655/TA3809*2//TA9121*2 F_3, where TA5655 is a disomic wheat-*Elymus tsukushiense* Honda Robertsonian translocation TW·1E^{ts}#1S, TA3809 is a Chinese Spring stock homozygous for the *ph1b* mutant allele, and TA9121 is the hard red winter wheat cultivar Everest. KS14WGRC61 is homozygous for a distal wheat–*E. tsukushiense* recombinant chromosome TWL·WS-1E^{ts}#1S, consisting of the complete long arm and most of the short arm of a wheat chromosome and a distal segment derived from 1E^{ts}#1S. The E^{ts}#1S segment in this translocation has a gene that confers type-2 resistance to FHB. The TWL·WS-1E^{ts}#1S stock is a novel source of FHB resistance and may be useful in wheat improvement. Small quantities (3 grams) of seed of KS14WGGRC61 are available upon written request. We request that the appropriate source be given when this germ plasm contributes to research or development of new cultivars. Seed stocks are maintained by the Wheat Genetics Resource Center, Throckmorton Plant Sciences Center, Kansas State University, Manhattan, KS 66506.

Evaluating a core collection for stress tolerance in the field.

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A core set of *Aegilops*, *Triticum*, and *Dasypyrum* accessions was evaluated at the Rocky Ford Research Area, Manhattan, KS, for field resistance to leaf rust, barley yellow dwarf virus, and powdery mildew (Table 1, pp. 138-147). The lines also were evaluated for heading date. Leaf rust reaction was recorded on three dates and barley yellow dwarf and powdery mildew on two. Virus infection was rated as symptoms on visible as chlorosis, necrosis of the leaf tips and leaves, or purpling of the leaves. One accession of *Ae. columnaris*, *Ae. peregrina*, *Ae. sharonensis*, *Ae. umbellulata*, *T. aestivum*, and *T. zhukovskyi*; two accessions of *Ae. longissima*, *T. turgidum* subsps. *carthlicum* and dicoccum; and six accessions of *T. turgidum* subsp. *polonicum* were winterkilled.

Table 1. Data from the core set of *Triticum* and *Aegilops* species evaluated for disease severity in the field, Manhattan, KS, for field resistance to leaf rust, barley yellow dwarf virus, and powdery mildew. Heading date also was recorded. Leaf rust was evaluated on the Cobb scale, where a number indicating the percent of leaf area affected is followed by a letter designation, R = resistant flecks or very small pustules, MR = moderately resistant small pustules, M = moderate small to medium size pustules, MS = moderately susceptible medium to large pustules, and S = susceptible with large pustules. Rating of the leaves with virus symptoms was 0 = no visible signs of infection, L = low infection with 10% or less of the leaf area with visible symptoms, M = moderate infection with up to 40% of the leaf area with visible symptoms, and H = high infection with over 40% of the leaf area showing symptoms. Powdery mildew present on leaves rated as 0 = no mildew seen, L = low infection with 10% or less of leaf area with mildew spores, M = moderate infection with up to 40% of the leaf area having spores, and H = high infection with over 40% of the leaf area having spores present. — = no test.

TA	Genus	species	Country of origin	Leaf rust			Barley yellow dwarf virus		Powdery mildew		Heading date
				5/22	6/9	6/13	5/22	6/6	5/22	6/6	
2909	*Triticum*	*aestivum* 'Jagger'	United States	10R	20MS	40MS	M	H	H	M	17-May
				20MR	70S	70S	M	H	H	M	18-May
2922	*Triticum*	*aestivum* 'Heines IV'	Germany	10R	20MR	40S	L	M	M	L	11-Jun
				10R	30MS	30MS	M	M	M	M	12-Jun
2951	*Triticum*	*aestivum* 'TAM 107'	United States	10R	70MS	80S	L	M	0	L	17-May
				1M	80S	—	0	M	H	H	17-May
3009	*Triticum*	*aestivum* 'Wichita'	United States	10R	50MS	70MS	M	H	M	L	27-May
				30MR	50MS	60MS	M	H	M	M	27-May
3014	*Triticum*	*aestivum* 'Courtot'	France	0	20MS	60MS	M	H	0	L	26-May
				10R	40MS	75S	M	H	L	M	26-May
10374	*Triticum*	*aestivum* landrace	Uzbekistan	0	10MS	40MS	M	H	L	L	4-Jun
				10R	20MS	40MS	M	H	L	L	8-Jun
10380	*Triticum*	*aestivum* landrace	Kyrgyzstan	10R	10M	15MS	M	H	L	L	22-May
				25MR	25MS	40MS	L	M	L	L	24-May
10395	*Triticum*	*aestivum* landrace	Tajikistan	0	45S	60S	M	M	H	H	8-Jun
10428	*Triticum*	*aestivum* 'Kirik'	Turkey	10R	40MS	—	H	H	H	H	27-May
				15R	40S	50S	H	H	H	M	28-May
10431	*Triticum*	*aestivum* landrace	Iran	1R	20MR	—	M	H	M	L	3-Jun
				1R	5MR	20MR	M	H	L	L	11-Jun
2601	*Triticum*	*aestivum* subsp. *compactum*	Turkey	10R	50MS	50M	H	H	H	M	28-May
				10R	40S	40S	H	H	H	H	5-Jun
10430	*Triticum*	*aestivum* subsp. *compactum*	Pakistan	10R	30MS	50MS	M	H	M	L	27-May
				5MS	70MS	70S	M	M	H	H	7-Jun
10861	*Triticum*	*aestivum* subsp. *compactum*	Kazakhstan	10R	40S	—	L	M	H	H	18-Jun
				15MR	30S	—	H	H	M	M	9-Jun
10862	*Triticum*	*aestivum* subsp. *macha*	Iran	10MR	60S	80S	L	H	H	H	12-Jun
				15R	80S	80S	L	M	H	H	10-Jun
10863	*Triticum*	*aestivum* subsp. *macha*	Georgia	10R	60S	60S	M	H	M	L	12-Jun
				10R	10MS	30MS	M	H	H	H	10-Jun
2603	*Triticum*	*aestivum* subsp. *spelta*	Switzerland	10R	20MS	20S	M	H	M	L	10-Jun
				10R	30S	40S	L	H	M	M	12-Jun
10424	*Triticum*	*aestivum* subsp. *spelta*	Iran	10R	50S	70S	H	H	H	H	12-Jun
				25MR	70S	80S	M	H	H	H	29-May
2605	*Triticum*	*aestivum* subsp. *sphaerococcum*	Greece	15R	50S	80S	L	M	H	L	26-May
				15R	60MS	70S	M	M	M	M	20-May
10864	*Triticum*	*aestivum* subsp. *sphaerococcum*	India	10R	15MS	30S	M	H	H	M	20-May
183	*Triticum*	*monococcum* subsp. *aegilopoides*	Iran	10R	15MR	—	L	H	0	0	27-May
				20MR	25MS	—	M	H	0	L	26-May
236	*Triticum*	*monococcum* subsp. *aegilopoides*	Iraq	0	10MR	20MR	0	M	0	0	22-May
				10R	25M	40M	L	H	L	L	24-May
352	*Triticum*	*monococcum* subsp. *aegilopoides*	Iraq	1R	5M	—	L	H	L	L	18-May
				0	10MR	—	L	H	0	L	18-May
463	*Triticum*	*monococcum* subsp. *aegilopoides*	Iraq	1R	5M	—	M	H	0	L	26-May
				10R	20MR	30M	L	H	0	L	23-May
547	*Triticum*	*monococcum* subsp. *aegilopoides*	Lebanon	5R	5MR	—	L	M	0	0	27-May
				10R	15MR	20MR	L	H	0	L	28-May

Table 1. Data from the core set of *Triticum* and *Aegilops* species evaluated for disease severity in the field, Manhattan, KS, for field resistance to leaf rust, barley yellow dwarf virus, and powdery mildew. Heading date also was recorded. Leaf rust was evaluated on the Cobb scale, where a number indicating the percent of leaf area affected is followed by a letter designation, R = resistant flecks or very small pustules, MR = moderately resistant small pustules, M = moderate small to medium size pustules, MS = moderately susceptible medium to large pustules, and S = susceptible with large pustules. Rating of the leaves with virus symptoms was 0 = no visible signs of infection, L = low infection with 10% or less of the leaf area with visible symptoms, M = moderate infection with up to 40% of the leaf area with visible symptoms, and H = high infection with over 40% of the leaf area showing symptoms. Powdery mildew present on leaves rated as 0 = no mildew seen, L = low infection with 10% or less of leaf area with mildew spores, M = moderate infection with up to 40% of the leaf area having spores, and H = high infection with over 40% of the leaf area having spores present. — = no test.

TA	Genus	species	Cou—ry of origin	Leaf rust			Barley yellow dwarf virus		Powdery mildew		Heading date
				5/22	6/9	6/13	5/22	6/6	5/22	6/6	
582	*Triticum*	*monococcum* subsp. *aegilopoides*	Armenia	0	10MR	15MR	0	H	0	0	6-Jun
				0	30MS	—	0	H	0	H	28-May
570	*Triticum*	*monococcum* subsp. *aegilopoides*	Azerbaijan	1R	15M	—	L	H	0	L	3-Jun
				5R	10MR	—	L	H	0	L	28-May
641	*Triticum*	*monococcum* subsp. *aegilopoides*	Turkey	5R	5M	—	L	H	0	L	22-May
				5R	5MR	10MR	0	M	L	L	23-May
2005	*Triticum*	*monococcum* subsp. *aegilopoides*	Turkey	0	1R	15MR	L	M	0	0	8-Jun
				0	1R	20MR	L	M	0	0	11-Jun
2010	*Triticum*	*monococcum* subsp. *aegilopoides*	Turkey	1R	15M	30M	L	H	0	0	9-Jun
				5R	10MR	20MR	M	M	0	L	29-May
137	*Triticum*	*monococcum* subsp. *monococcum*	Turkey	10R	15M	20M	M	M	0	0	9-Jun
				10R	15MR	15MR	L	H	L	0	12-Jun
2025	*Triticum*	*monococcum* subsp. *monococcum*	Turkey	10R	10R	15R	L	M	0	0	8-Jun
				30MR	30MR	35MR	M	M	0	0	29-May
2033	*Triticum*	*monococcum* subsp. *monococcum*	Portugal	0	5R	5R	M	M	L	L	7-Jun
				0	1R	5R	M	M	0	0	28-May
2034	*Triticum*	*monococcum* subsp. *monococcum*	Bosnia–Herzegovina	0	1R	5MR	M	M	0	0	8-Jun
				5R	5R	10MR	M	H	0	0	28-May
2039	*Triticum*	*monococcum* subsp. *monococcum*	Albania	10R	15MR	20MR	M	H	0	0	11-Jun
				10R	10R	30MR	L	M	0	0	3-Jun
10594	*Triticum*	*monococcum* subsp. *aegilopoides*	Turkey	5R	15MR	15M	0	H	0	M	11-Jun
				0	1R	30M	L	M	0	0	8-Jun
10604	*Triticum*	*monococcum* subsp. *monococcum*	Turkey	1R	1MR	5MR	M	M	H	0	8-Jun
				1R	1R	10MR	M	M	M	M	12-Jun
10634	*Triticum*	*monococcum* subsp. *monococcum*	Italy	10R	10MR	15MR	M	M	0	L	14-Jun
				5R	5R	20R	L	L	0	0	14-Jun
10635	*Triticum*	*monococcum* subsp. *monococcum*	Georgia	10R	10MR	10MR	L	M	H	H	12-Jun
				15R	15R	15MR	L	M	L	H	12-Jun
732	*Triticum*	*urartu*	Turkey	5R	—	—	L	—	M	—	23-May
				0	10M	—	L	H	L	L	22-May
768	*Triticum*	*urartu*	Lebanon	5R	5M	—	M	H	0	0	24-May
				10R	15MR	—	L	H	0	0	26-May
828	*Triticum*	*urartu*	Armenia	0	5M	—	L	H	H	H	10-Jun
				0	10MR	15M	M	M	H	H	28-May
831	*Triticum*	*urartu*	Iran	5R	10M	—	0	H	M	L	24-May
				1R	10M	—	0	H	H	H	21-May
856	*Triticum*	*urartu*	Iraq	5R	5M	—	M	H	H	L	23-May
				15R	30MS	—	L	H	H	H	20-May
4	*Triticum*	*timopheevii subsp. armeniacum*	Iran	1R	10M	30M	M	H	0	0	24-May
				1R	20M	—	M	H	0	M	23-May
6	*Triticum*	*timopheevii subsp. armeniacum*	Turkey	0	20MS	20MS	L	H	0	L	27-May
				5R	15M	20MS	M	M	0	L	29-May
7	*Triticum*	*timopheevii subsp. armeniacum*	Iraq	5R	1MR	—	M	H	0	0	26-May
				5R	10MR	—	L	H	0	L	28-May
36	*Triticum*	*timopheevii subsp. armeniacum*	Iraq	1R	10M	—	M	H	0	L	24-May
				1R	25M	—	L	H	H	M	27-May

Table 1. Data from the core set of *Triticum* and *Aegilops* species evaluated for disease severity in the field, Manhattan, KS, for field resistance to leaf rust, barley yellow dwarf virus, and powdery mildew. Heading date also was recorded. Leaf rust was evaluated on the Cobb scale, where a number indicating the percent of leaf area affected is followed by a letter designation, R = resistant flecks or very small pustules, MR = moderately resistant small pustules, M = moderate small to medium size pustules, MS = moderately susceptible medium to large pustules, and S = susceptible with large pustules. Rating of the leaves with virus symptoms was 0 = no visible signs of infection, L = low infection with 10% or less of the leaf area with visible symptoms, M = moderate infection with up to 40% of the leaf area with visible symptoms, and H = high infection with over 40% of the leaf area showing symptoms. Powdery mildew present on leaves rated as 0 = no mildew seen, L = low infection with 10% or less of leaf area with mildew spores, M = moderate infection with up to 40% of the leaf area having spores, and H = high infection with over 40% of the leaf area having spores present. — = no test.

TA	Genus	species	Cou—ry of origin	Leaf rust			Barley yellow dwarf virus		Powdery mildew		Heading date
				5/22	6/9	6/13	5/22	6/6	5/22	6/6	
49	*Triticum*	*timopheevii subsp. armeniacum*	Azerbaijan	0	1MR	10MR	L	H	0	0	6-Jun
				1R	20MR	30MR	L	H	0	M	29-May
896	*Triticum*	*timopheevii subsp. armeniacum*	Iraq	0	10MR	—	0	H	0	0	26-May
				1R	5MR	—	L	M	0	L	28-May
1900	*Triticum*	*timopheevii subsp. armeniacum*	Turkey	10R	20MR	30MR	H	H	0	0	8-Jun
				15R	15MR	30MR	H	H	0	L	11-Jun
2893	*Triticum*	*timopheevii subsp. armeniacum*	Armenia	1R	5MR	30M	L	M	0	0	10-Jun
				5R	30MR	30M	L	H	0	M	2-Jun
103	*Triticum*	*timopheevii subsp. timopheevii*	Serbia	5R	5MR	5MR	0	M	L	0	10-Jun
				10R	15R	20MR	L	M	L	L	12-Jun
10473	*Triticum*	*turgidum subsp. carthlicum*	Turkey	1R	30MS	40MS	M	H	L	M	27-May
				20MR	50MS	60MS	H	H	0	L	28-May
10477	*Triticum*	*turgidum subsp. carthlicum*	Georgia	10R	60MS	60MS	L	H	M	L	26-May
				25MR	60MS	60MS	H	H	H	H	28-May
60	*Triticum*	*turgidum subsp. dicoccoides*	Israel	10R	5M	—	M	H	0	L	24-May
				15MR	15M	—	H	H	0	M	27-May
107	*Triticum*	*turgidum subsp. dicoccoides*	Syrian Arab Republic (Golan Heights)	1R	5M	—	H	H	0	—	23-May
				10M	20M	—	M	H	H	M	23-May
1060	*Triticum*	*turgidum subsp. dicoccoides*	Lebanon	1M	50MS	—	L	H	0	L	26-May
				20MR	30MS	—	M	H	0	L	27-May
1082	*Triticum*	*turgidum subsp. dicoccoides*	Turkey	5R	30MS	—	H	H	H	L	6-Jun
				10R	50S	—	M	H	H	M	26-May
1181	*Triticum*	*turgidum subsp. dicoccoides*	Palestine (West Bank)	1R	10MS	—	H	H	0	—	26-May
				5M	20M	—	H	H	0	M	23-May
1385	*Triticum*	*turgidum subsp. dicoccoides*	Iraq	1R	20MS	—	M	H	H	L	22-May
				15MR	30MS	—	M	H	H	H	27-May
1454	*Triticum*	*turgidum subsp. dicoccoides*	Palestine (West Bank)	5R	25MS	—	L	H	0	L	26-May
				15R	25M	—	M	H	0	L	27-May
10479	*Triticum*	*turgidum subsp. dicoccum*	Turkey	5R	20MS	—	M	H	L	L	7-Jun
				15R	60MS	60MS	M	M	L	L	10-Jun
10480	*Triticum*	*turgidum subsp. dicoccum*	Turkey	10R	20MR	30S	L	H	M	L	3-Jun
				25MR	25M	30MS	M	M	0	L	26-May
10484	*Triticum*	*turgidum subsp. dicoccum*	Oman	10R	60MS	—	M	H	H	H	27-May
				15R	60S	80S	H	H	H	H	29-May
10504	*Triticum*	*turgidum subsp. dicoccum*	Iran	15MR	15MR	20MR	L	M	H	H	14-Jun
10517	*Triticum*	*turgidum subsp. dicoccum*	Armenia	5R	20M	40M	L	M	0	0	12-Jun
				15R	15MR	30MR	M	M	0	0	14-Jun
2807	*Triticum*	*turgidum subsp. paleocolchicum*	Georgia	5R	20M	30M	M	H	0	M	6-Jun
				15MR	15MR	60M	M	H	0	M	29-May
10858	*Triticum*	*turgidum subsp. polonicum*	Pakistan	10R	10MR	20M	M	M	M	L	4-Jun
				20R	20MR	20MR	M	H	0	L	4-Jun
10859	*Triticum*	*turgidum subsp. polonicum*	India	15MR	40MS	50S	L	H	H	L	28-May
10534	*Triticum*	*turgidum subsp. turanicum*	Turkey	10R	15M	15S	M	H	H	H	28-May
				15R	15MR	15M	L	L	0	L	6-Jun
10537	*Triticum*	*turgidum subsp. turanicum*	Iran	1R	25S	30S	M	M	M	L	11-Jun
				15R	30M	40S	M	M	M	M	4-Jun

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				5/22	6/9	6/13	5/22	6/6	5/22	6/6	
10541	*Triticum*	*turgidum subsp. turanicum*	Syrian Arab Republic	15R	35MS	40M	M	H	H	L	27-May
				10R	25MS	50MS	M	H	L	M	28-May
1945	*Aegilops*	*bicornis*	Egypt	0	30MS	—	L	H	0	L	22-May
				1R	25M	—	L	H	0	L	19-May
1951	*Aegilops*	*bicornis*	Israel	1M	—	—	L	—	0	—	18-May
				1R	20MS	—	M	H	0	L	17-May
2349	*Aegilops*	*biuncialis*	Turkey	1R	0	—	0	M	0	0	25-May
				1R	1R	10R	0	M	0	L	24-May
2663	*Aegilops*	*biuncialis*	Syrian Arab Republic	0	5M	—	M	H	0	L	18-May
				0	5MR	—	L	H	0	L	17-May
2783	*Aegilops*	*biuncialis*	Bosnia–Herze-govina	0	1MR	—	L	H	0	0	2-Jun
				0	5R	5MR	L	H	0	L	27-May
10057	*Aegilops*	*biuncialis*	Libya	0	5MR	—	0	H	0	L	20-May
				1R	5MR	—	L	H	0	L	20-May
10058	*Aegilops*	*biuncialis*	Azerbaijan	10R	15MR	—	M	H	0	L	24-May
				0	5MR	—	M	H	0	L	22-May
10060	*Aegilops*	*biuncialis*	Greece	5R	10M	—	L	H	0	L	22-May
				20MR	20MR	—	L	H	L	L	21-May
2106	*Aegilops*	*columnaris*	Turkey	0	5MR	15MR	0	H	0	L	28-May
				15MR	15MR	20MR	M	H	0	L	26-May
2182	*Aegilops*	*columnaris*	Turkey	0	1R	5MR	M	H	0	0	28-May
2656	*Aegilops*	*columnaris*	Syrian Arab Republic	5R	10M	—	M	H	0	L	20-May
				20R	20M	—	0	H	0	L	22-May
10049	*Aegilops*	*columnaris*	Azerbaijan	10R	15M	—	L	H	0	L	24-May
				0	10MR	—	L	H	0	L	18-May
2102	*Aegilops*	*comosa var. comosa*	Turkey	0	5MR	—	0	M	0	0	28-May
				5R	5R	—	L	M	0	0	26-May
2757	*Aegilops*	*comosa var. comosa*	Greece	5R	5M	—	L	H	0	0	25-May
				10R	15MR	15MR	L	H	0	0	27-May
2734	*Aegilops*	*comosa var. subventricosa*	Greece	0	10M	—	L	H	0	—	25-May
				0	5MR	—	0	H	0	L	22-May
1873	*Aegilops*	*crassa* (4x)	Iran	5R	25M	—	M	H	0	M	20-May
				5R	40MS	—	L	H	0	M	21-May
1876	*Aegilops*	*crassa* (4x)	Iran	0	20MS	—	L	H	0	L	24-May
				0	80S	—	0	H	0	L	26-May
1881	*Aegilops*	*crassa* (4x)	Afghanistan	1M	20M	—	L	H	0	L	20-May
				0	25MS	—	L	H	0	L	18-May
2319	*Aegilops*	*crassa* (4x)	Turkey	0	25MS	—	L	H	0	L	22-May
				0	30MS	—	L	H	0	L	20-May
10340	*Aegilops*	*crassa*	Tajikistan	1R	70MS	—	L	H	0	0	25-May
				0	80MS	—	L	H	0	L	26-May
1858	*Aegilops*	*cylindrica*	Turkey	0	10M	H	L	H	0	L	24-May
				1R	20M	—	L	H	0	L	26-May
2203	*Aegilops*	*cylindrica*	Iran	1MR	60MS	—	M	H	0	L	26-May
				0	60MS	—	L	H	0	L	26-May
2204	*Aegilops*	*cylindrica*	Armenia	1R	50MS	—	L	H	0	L	27-May
				0	60MS	—	L	H	0	L	28-May

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TA	Genus	species	Cou—ry of origin	Leaf rust			Barley yellow dwarf virus		Powdery mildew		Heading date
				5/22	6/9	6/13	5/22	6/6	5/22	6/6	
2331	*Aegilops*	*cylindrica*	Turkey	0	60MS	—	L	M	0	0	26-May
				0	60MS	—	0	H	0	L	28-May
10148	*Aegilops*	*cylindrica*	Kazakhstan	5M	30M	—	M	H	0	L	25-May
				0	15MS	—	M	H	H	M	20-May
10346	*Aegilops*	*cylindrica*	Tajikistan	0	30M	—	L	H	0	L	26-May
				1R	40MS	—	M	H	0	L	24-May
10356	*Aegilops*	*cylindrica*	Kyrgyzstan	0	—	—	M	H	0	—	26-May
				1R	20M	—	M	H	0	L	24-May
1801	*Aegilops*	*geniculata*	Turkey	0	5MR	—	L	H	0	L	22-May
				1R	5MR	—	L	H	0	L	24-May
1805	*Aegilops*	*geniculata*	Turkey	0	5MR	—	0	H	0	L	22-May
				1R	5MR	—	L	H	0	L	24-May
1879	*Aegilops*	*geniculata*	Jordan	5R	20MR	—	L	H	H	—	17-May
				1R	10MR	—	0	H	0	L	19-May
2041	*Aegilops*	*geniculata*	Morocco	1R	10MR	15MR	L	H	0	L	26-May
				10R	15R	—	0	M	0	L	26-May
2227	*Aegilops*	*geniculata*	Morocco	0	5M	—	L	H	0	L	22-May
				5R	5MR	—	L	H	0	L	20-May
2239	*Aegilops*	*geniculata*	Morocco	10R	15M	—	L	H	0	L	22-May
				5R	5MR	—	M	H	0	L	20-May
2649	*Aegilops*	*geniculata*	Turkey	1R	10M	—	0	H	0	—	18-May
				10R	10M	—	0	H	0	L	16-May
2650	*Aegilops*	*geniculata*	Syrian Arab Republic	0	1MR	—	0	M	0	L	20-May
				10R	20M	—	0	M	0	L	19-May
2786	*Aegilops*	*geniculata*	Bosnia–Herze-govina	1R	15MR	—	L	M	0	0	24-May
				5R	5R	10R	0	M	0	L	23-May
2787	*Aegilops*	*geniculata*	Croatia	0	1MR	—	L	H	0	0	24-May
				0	5R	5MR	L	H	0	L	23-May
2899	*Aegilops*	*geniculata*	Israel	15R	15MR	30MR	0	L	0	0	23-May
				5R	10MR	30MR	0	M	0	L	20-May
10029	*Aegilops*	*geniculata*	Morocco	1R	5MR	—	0	H	0	0	24-May
				1R	5MR	—	0	H	0	L	22-May
2346	*Aegilops*	*juvenalis*	Iran	0	20M	—	L	H	0	L	22-May
				1R	40MS	—	M	H	0	L	20-May
2347	*Aegilops*	*juvenalis*	Iraq	1M	25M	—	M	H	0	H	17-May
				5M	10M	—	M	H	L	L	22-May
1980	*Aegilops*	*kotschyi*	Israel	1R	10M	—	H	H	0	L	17-May
				5R	—	—	H	—	0	—	16-May
1981	*Aegilops*	*kotschyi*	Egypt	1R	10M	—	M	H	0	L	18-May
				0	5MR	—	H	H	L	M	17-May
1984	*Aegilops*	*kotschyi*	Egypt	0	5R	5MR	L	H	0	L	19-May
				0	10M	—	M	H	0	L	17-May
2207	*Aegilops*	*kotschyi*	Uzbekistan	10R	—	—	L	—	0	—	16-May
				30MR	30MR	—	L	H	0	L	16-May
2667	*Aegilops*	*kotschyi*	Jordan	1R	10M	—	M	H	0	—	18-May
				1R	10M	—	M	H	0	L	16-May

Table 1. Data from the core set of *Triticum* and *Aegilops* species evaluated for disease severity in the field, Manhattan, KS, for field resistance to leaf rust, barley yellow dwarf virus, and powdery mildew. Heading date also was recorded. Leaf rust was evaluated on the Cobb scale, where a number indicating the percent of leaf area affected is followed by a letter designation, R = resistant flecks or very small pustules, MR = moderately resistant small pustules, M = moderate small to medium size pustules, MS = moderately susceptible medium to large pustules, and S = susceptible with large pustules. Rating of the leaves with virus symptoms was 0 = no visible signs of infection, L = low infection with 10% or less of the leaf area with visible symptoms, M = moderate infection with up to 40% of the leaf area with visible symptoms, and H = high infection with over 40% of the leaf area showing symptoms. Powdery mildew present on leaves rated as 0 = no mildew seen, L = low infection with 10% or less of leaf area with mildew spores, M = moderate infection with up to 40% of the leaf area having spores, and H = high infection with over 40% of the leaf area having spores present. — = no test.

TA	Genus	species	Cou—ry of origin	Leaf rust			Barley yellow dwarf virus		Powdery mildew		Heading date
				5/22	6/9	6/13	5/22	6/6	5/22	6/6	
1910	*Aegilops*	*longissima*	Israel	1R	5MR	—	M	H	0	0	22-May
				0	5MR	20MR	M	H	0	0	22-May
1921	*Aegilops*	*longissima*	Jordan	1R	5M	—	H	H	0	0	20-May
				10MS	30MS	—	L	H	0	L	20-May
1906	*Aegilops*	*markgrafii*	Turkey	0	30M	—	L	H	0	L	20-May
				5R	5MR	—	M	H	0	L	22-May
1908	*Aegilops*	*markgrafii*	Unknown	10R	15MR	—	L	H	0	0	26-May
				5R	5MR	—	M	H	0	0	27-May
2087	*Aegilops*	*markgrafii*	Turkey	0	1R	5MR	L	H	0	0	8-Jun
				1R	5MR	10MR	L	H	0	L	10-Jun
2090	*Aegilops*	*markgrafii*	Turkey	1R	1R	—	L	H	0	0	26-May
				0	1MR	5MR	L	H	0	L	28-May
2096	*Aegilops*	*markgrafii*	Turkey	0	5MR	5MR	L	H	L	L	28-May
1961	*Aegilops*	*neglecta*	Iraq	1R	10MR	10MR	L	M	0	L	22-May
				15MR	15MR	—	L	H	0	L	23-May
2153	*Aegilops*	*neglecta*	Turkey	0	1R	5MR	L	M	0	L	28-May
				10R	10MR	20MR	0	M	L	L	27-May
2156	*Aegilops*	*neglecta*	Turkey	0	5MR	10MR	M	M	0	L	28-May
				0	10MR	20MR	L	M	0	M	28-May
2341	*Aegilops*	*neglecta*	Turkey	0	1M	—	M	H	0	0	23-May
				1R	5MR	—	L	H	0	L	25-May
2790	*Aegilops*	*neglecta*	Bosnia–Herze-govina	1R	5M	—	L	H	0	0	26-May
				1R	10M	—	L	H	0	L	26-May
2793	*Aegilops*	*neglecta*	Croatia	0	5MR	—	L	H	0	0	27-May
				10R	20MR	20M	L	H	0	L	27-May
10062	*Aegilops*	*neglecta*	Spain	0	1R	5MR	L	H	0	L	3-Jun
				0	5R	10MR	L	H	0	0	3-Jun
10064	*Aegilops*	*neglecta*	Greece	5R	5R	5MR	L	M	0	0	10-Jun
				0	1R	5MR	L	M	0	0	28-May
10065	*Aegilops*	*neglecta*	Ukraine	0	5MR	—	0	H	0	0	2-Jun
				0	1R	—	L	M	0	0	28-May
10066	*Aegilops*	*neglecta*	France	1R	5M	5M	L	H	0	0	24-May
				5R	5MR	1OM	L	H	0	L	24-May
1886	*Aegilops*	*peregrina*	Syrian Arab Republic	10R	10M	—	L	H	0	L	18-May
				20MR	20MR	—	M	H	0	L	18-May
1889	*Aegilops*	*peregrina*	Israel	10R	15MR	15MR	0	M	0	L	24-May
				5R	5MR	20MR	0	M	0	0	26-May
1897	*Aegilops*	*peregrina*	Turkey	10R	15MR	—	L	H	0	L	21-May
				10MR	15MR	—	L	H	0	M	19-May
1898	*Aegilops*	*peregrina*	Lebanon	0	20M	—	L	H	0	0	23-May
				0	10M	—	L	M	0	L	22-May
1918	*Aegilops*	*peregrina*	Turkey	0	5M	25M	L	M	0	L	26-May
				0	20MS	—	0	H	0	L	26-May
2775	*Aegilops*	*peregrina*	Israel	1R	1R	15R	L	M	0	0	24-May
				10R	15MR	15MR	L	H	0	L	23-May
1837	*Aegilops*	*searsii*	Palestine	5R	5M	—	M	H	H	H	26-May
				10R	25MS	—	L	H	H	L	26-May

Table 1. Data from the core set of *Triticum* and *Aegilops* species evaluated for disease severity in the field, Manhattan, KS, for field resistance to leaf rust, barley yellow dwarf virus, and powdery mildew. Heading date also was recorded. Leaf rust was evaluated on the Cobb scale, where a number indicating the percent of leaf area affected is followed by a letter designation, R = resistant flecks or very small pustules, MR = moderately resistant small pustules, M = moderate small to medium size pustules, MS = moderately susceptible medium to large pustules, and S = susceptible with large pustules. Rating of the leaves with virus symptoms was 0 = no visible signs of infection, L = low infection with 10% or less of the leaf area with visible symptoms, M = moderate infection with up to 40% of the leaf area with visible symptoms, and H = high infection with over 40% of the leaf area showing symptoms. Powdery mildew present on leaves rated as 0 = no mildew seen, L = low infection with 10% or less of leaf area with mildew spores, M = moderate infection with up to 40% of the leaf area having spores, and H = high infection with over 40% of the leaf area having spores present. — = no test.

TA	Genus	species	Cou—ry of origin	Leaf rust			Barley yellow dwarf virus		Powdery mildew		Heading date
				5/22	6/9	6/13	5/22	6/6	5/22	6/6	
2344	*Aegilops*	*searsii*	Syrian Arab Republic	1R	10M	—	L	H	H	—	24-May
				5R	—	—	H	—	H	—	24-May
2355	*Aegilops*	*searsii*	Palestine	1R	—	—	M	—	0	—	24-May
				1R	5MR	—	L	H	L	L	24-May
2669	*Aegilops*	*searsii*	Jordan	1R	10M	—	M	H	0	—	24-May
				5R	5MR	—	L	H	0	L	25-May
1996	*Aegilops*	*sharonensis*	Israel	5R	—	—	H	—	0	—	22-May
				5MR	20S	—	L	H	0	L	18-May
1998	*Aegilops*	*sharonensis*	Israel	1R	5MR	—	M	H	H	H	22-May
				0	5R	25MR	0	H	0	L	21-May
10434	*Aegilops*	*sharonensis*	Israel	1R	5MR	—	L	H	0	L	22-May
1772	*Aegilops*	*speltoides* var. *ligustica*	Turkey	10R	10MR	10MR	L	H	0	0	26-May
				5R	5R	5MR	L	M	0	0	27-May
1778	*Aegilops*	*speltoides* var. *ligustica*	Turkey	1R	1R	1R	L	M	0	0	26-May
				0	1R	1R	L	M	0	0	27-May
1789	*Aegilops*	*speltoides* var. *ligustica*	Iraq	1R	1R	1R	L	M	0	0	22-May
				1R	1R	1R	L	H	0	0	26-May
2646	*Aegilops*	*speltoides* var. *ligustica*	Turkey	0	1MR	1MR	L	H	0	0	8-Jun
				0	1R	1R	L	H	0	0	7-Jun
2781	*Aegilops*	*speltoides* var. *ligustica*	Israel	0	0	10R	L	M	0	0	3-Jun
				0	1R	1R	0	H	0	0	28-May
1795	*Aegilops*	*speltoides* var. *speltoides*	Iraq	0	0	1R	L	H	0	0	24-May
				5R	5R	5MR	L	H	0	0	26-May
1971	*Aegilops*	*speltoides* var. *speltoides*	Turkey	1R	0	1R	L	M	0	0	24-May
				1R	1R	5MR	L	H	0	0	25-May
2342	*Aegilops*	*speltoides* var. *speltoides*	Israel	0	0	1R	L	H	0	0	6-Jun
				0	1R	5MR	L	H	0	0	24-May
2356	*Aegilops*	*speltoides* var. *speltoides*	Turkey	0	1R	10R	0	L	0	0	2-Jun
				0	1R	1R	L	M	0	0	10-Jun
10545	*Aegilops*	*speltoides* var. *speltoides*	Syrian Arab Republic	0	5M	—	L	H	0	L	26-May
				10MR	10MR	15MR	L	H	0	L	24-May
2780	*Aegilops*	*speltoides* var. *speltoides*/*ligustica* mix	Israel	0	0	5R	L	M	0	0	2-Jun
				1R	1R	1R	L	H	0	0	27-May
1588	*Aegilops*	*tauschii*	Turkey	0	10M	40MS	L	M	0	L	4-Jun
				15R	30M	—	M	H	H	L	2-Jun
1626	*Aegilops*	*tauschii* f. *strangulata*	Turkmenistan	0	10M	30M	0	H	L	L	26-May
				5R	5MR	15M	M	M	0	L	27-May
1642	*Aegilops*	*tauschii* f. *strangulata*	Iran	10R	10MR	15MR	0	M	L	L	26-May
				5R	10MR	—	L	H	0	L	26-May
1659	*Aegilops*	*tauschii* f. *strangulata*	Azerbaijan	5R	10M	25M	L	H	0	L	24-May
				15MR	15M	—	L	H	0	L	26-May
1662	*Aegilops*	*tauschii*	Azerbaijan	1R	5M	—	M	H	0	0	26-May
				10R	15M	20MS	0	M	0	L	26-May
1668	*Aegilops*	*tauschii* f. *strangulata*	Azerbaijan	1R	1MR	—	0	M	0	0	26-May
				1R	5MR	10MR	L	M	L	L	27-May
1673	*Aegilops*	*tauschii*	Azerbaijan	0	5M	—	L	M	0	L	26-May
				5R	25M	—	L	M	0	L	24-May

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TA	Genus	species	Cou—ry of origin	Leaf rust			Barley yellow dwarf virus		Powdery mildew		Heading date
				5/22	6/9	6/13	5/22	6/6	5/22	6/6	
1717	*Aegilops*	*tauschii*	Iran	5R	5MR	—	L	H	M	M	22-May
				5R	10MR	—	L	H	L	L	21-May
2374	*Aegilops*	*tauschii*	Pakistan	1R	15M	—	M	H	M	L	17-May
				1R	50MS	—	M	H	H	L	16-May
2378	*Aegilops*	*tauschii*	Iran	5R	10MR	—	L	H	L	H	20-May
				5R	10MR	—	L	H	H	H	20-May
2388	*Aegilops*	*tauschii*	Afghanistan	1MR	20MS	—	M	H	H	H	16-May
				1MR	60S	—	H	H	H	H	16-May
2398	*Aegilops*	*tauschii*	Afghanistan	1R	30MS	—	L	H	H	H	19-May
				1MR	40MS	—	L	H	H	H	21-May
2422	*Aegilops*	*tauschii*	Afghanistan	1R	10M	—	M	H	0	L	20-May
				1MR	10MS	—	M	H	0	L	18-May
2489	*Aegilops*	*tauschii*	Iran	5R	20MS	—	M	H	H	H	8-Jun
				5R	40MS	—	M	H	H	H	28-May
2521	*Aegilops*	*tauschii*	Iran	5R	15M	—	M	H	H	H	22-May
				1R	60S	—	L	H	H	H	24-May
2536	*Aegilops*	*tauschii*	Afghanistan	10R	10M	—	L	H	0	L	17-May
				15R	30M	—	M	H	0	M	18-May
2574	*Aegilops*	*tauschii*	Armenia	0	50MS	—	M	H	H	H	27-May
				1M	40S	—	L	H	H	H	27-May
2586	*Aegilops*	*tauschii*	Georgia	1R	20M	—	M	H	0	L	26-May
				10R	20MS	—	M	H	0	L	4-Jun
10077	*Aegilops*	*tauschii*	Pakistan	1R	10M	—	L	H	0	L	20-May
				5M	40MS	—	M	H	0	M	16-May
10106	*Aegilops*	*tauschii*	Kyrgyzstan	1R	5M	—	H	H	H	H	4-Jun
				0	20MS	—	M	H	H	H	8-Jun
10134	*Aegilops*	*tauschii*	PR China	0	5M	30MS	M	H	0	0	11-Jun
				15MR	40MS	—	M	M	H	H	29-May
10142	*Aegilops*	*tauschii*	Syrian Arab Republic	1R	5MR	—	L	H	0	L	22-May
				1R	20MR	—	L	H	M	M	24-May
10155	*Aegilops*	*tauschii*	Tajikistan	10R	20M	—	M	H	L	L	20-May
				10MR	25M	—	M	H	0	L	18-May
10166	*Aegilops*	*tauschii*	Turkmenistan	10R	20MR	—	L	H	H	H	20-May
				25MR	25MR	—	L	H	H	H	18-May
10189	*Aegilops*	*tauschii*	Uzbekistan	1R	20M	—	H	H	H	H	22-May
				15R	30MS	—	H	H	H	H	19-May
10193	*Aegilops*	*tauschii*	Uzbekistan	5R	20M	—	L	H	L	L	25-May
				10R	40MS	—	L	H	H	L	22-May
1725	*Aegilops*	*triuncialis*	Turkey	20R	25MR	—	H	H	0	0	23-May
				15R	15MR	—	M	H	0	L	21-May
1740	*Aegilops*	*triuncialis*	Turkey	1R	1MR	—	L	H	0	0	25-May
				0	10MR	—	0	M	0	L	21-May
1748	*Aegilops*	*triuncialis*	Afghanistan	1R	10MR	—	M	H	0	0	20-May
				15R	15MR	—	L	H	0	L	19-May
1752	*Aegilops*	*triuncialis*	Iran	10R	20MR	20MR	L	M	0	0	26-May
				5R	10MR	20MR	M	H	0	0	28-May

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TA	Genus	species	Cou—ry of origin	Leaf rust			Barley yellow dwarf virus		Powdery mildew		Heading date
				5/22	6/9	6/13	5/22	6/6	5/22	6/6	
1753	*Aegilops*	*triuncialis*	Turkey	0	5MR	—	0	H	0	0	26-May
				0	5MR	10MR	L	M	0	L	28-May
1755	*Aegilops*	*triuncialis*	Iran	1R	10MR	—	M	H	0	0	20-May
				10R	10MR	—	M	H	0	L	17-May
2135	*Aegilops*	*triuncialis*	Kazakhstan	5R	15M	—	L	H	0	L	27-May
				15R	15MR	—	M	H	L	M	27-May
2229	*Aegilops*	*triuncialis*	Morocco	0	5MR	5MR	0	H	0	L	27-May
				1R	10MR	—	L	H	0	L	25-May
2279	*Aegilops*	*triuncialis*	Turkey	0	5MR	10MR	L	M	0	L	28-May
				0	5MR	10MR	L	M	0	0	27-May
2304	*Aegilops*	*triuncialis*	Turkey	0	1R	5MR	L	M	0	L	2-Jun
				0	5MR	10MR	L	M	0	L	28-May
2325	*Aegilops*	*triuncialis*	Iraq	5R	10MR	20M	L	H	0	L	27-May
				10R	15M	30S	L	H	0	0	27-May
2622	*Aegilops*	*triuncialis*	Syrian Arab Republic	10R	15M	—	H	H	0	L	20-May
				15R	15M	—	0	M	0	L	19-May
2788	*Aegilops*	*triuncialis*	Croatia	0	1R	—	L	H	0	0	27-May
				0	5MR	—	0	H	0	L	28-May
10013	*Aegilops*	*triuncialis*	Morocco	1R	5MR	—	L	H	0	L	22-May
				0	5MR	20MR	L	H	0	L	2-Jun
10055	*Aegilops*	*triuncialis*	Portugal	0	1R	—	L	H	0	0	27-May
				5R	5R	—	L	M	0	L	28-May
10056	*Aegilops*	*triuncialis*	Greece	0	5MR	10MR	0	M	0	0	4-Jun
				10R	10MR	—	L	M	0	L	28-May
10358	*Aegilops*	*triuncialis*	Tajikistan	10R	20MR	40MR	M	M	0	L	28-May
				25MR	25MR	25MR	M	M	0	0	28-May
1825	*Aegilops*	*umbellulata*	Turkey	0	1R	—	0	H	0	0	2-Jun
				0	10M	—	0	H	0	L	28-May
1831	*Aegilops*	*umbellulata*	Iran	10R	10M	—	0	H	0	L	17-May
				15MR	15MR	—	L	H	0	L	17-May
1850	*Aegilops*	*umbellulata*	Syrian Arab Republic	1R	5MR	—	L	H	0	L	17-May
				5R	10M	—	L	H	0	L	18-May
1851	*Aegilops*	*umbellulata*	Unknown	20R	25MR	—	M	H	0	0	2-Jun
				10R	15MR	—	L	H	0	L	28-May
1852	*Aegilops*	*umbellulata*	Turkey	1R	15M	—	0	H	0	L	27-May
				1R	1MR	—	L	H	0	L	27-May
2641	*Aegilops*	*umbellulata*	Turkey	10R	10MR	—	L	H	0	L	20-May
				15R	15MR	—	L	M	0	L	18-May
10835	*Aegilops*	*umbellulata*	Azerbaijan	5R	—	—	0	H	0	—	20-May
				10R	—	—	0	H	0	—	18-May
2762	*Aegilops*	*uniaristata*	Greece	0	1MR	15M	0	M	0	0	26-May
				1R	10MR	20MR	L	H	0	L	28-May
2766	*Aegilops*	*uniaristata*	Greece	0	5M	—	L	H	M	L	24-May
				1R	5MR	—	L	H	H	H	24-May
2768	*Aegilops*	*uniaristata*	Greece	0	10M	—	0	H	0	0	27-May
				0	5M	—	L	H	0	L	25-May

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TA	Genus	species	Cou—ry of origin	Leaf rust			Barley yellow dwarf virus		Powdery mildew		Heading date
				5/22	6/9	6/13	5/22	6/6	5/22	6/6	
2655	*Aegilops*	*vavilovii*	Jordan	0	10M	—	L	H	M	H	20-May
				0	20MS	—	L	H	L	H	18-May
2210	*Aegilops*	*ventricosa*	Libya	0	—	—	M	—	0	—	17-May
				0	40MS	—	L	H	0	L	20-May
2230	*Aegilops*	*ventricosa*	Morocco	0	1MR	—	0	M	0	M	27-May
				1R	10MR	—	L	H	0	L	26-May
2741	*Ambly-opyrum*	*muticum*	Turkey	0	1R	20MR	0	H	0	0	8-Jun
				0	5MR	—	L	H	0	L	27-May
2199	*Dasypyrum*	*villosum*	Croatia	10R	15MR	—	M	H	0	0	17-May
				15R	15MR	15MR	M	M	0	0	19-May
10225	*Dasypyrum*	*villosum*	Italy	0	1R	5MR	M	H	0	0	19-May
				1R	10MR	—	L	H	0	0	17-May
10226	*Dasypyrum*	*villosum*	Bulgaria	0	0	—	L	H	0	0	24-May
				5R	10R	—	0	H	0	0	22-May
10232	*Dasypyrum*	*villosum*	Italy	0	20MR	—	0	M	0	0	18-May
				10R	15R	—	M	H	0	0	17-May
10235	*Dasypyrum*	*villosum*	France	0	5MR	—	H	H	0	0	19-May
				10R	15R	—	M	H	0	0	20-May
10239	*Dasypyrum*	*villosum*	Turkey	1R	1R	—	M	H	0	0	22-May
				10R	10R	10MR	L	H	0	0	20-May
10273	*Dasypyrum*	*villosum*	Greece	5R	5MR	—	M	H	0	0	18-May
				0	10MR	—	M	H	0	L	16-May
10289	*Dasypyrum*	*villosum*	Greece	0	1MR	—	L	H	0	0	20-May
				10R	10MR	—	M	H	0	0	19-May
10662	*Dasypyrum*	*villosum*	Greece	0	5MR	—	M	H	0	0	18-May
				0	5MR	—	M	H	0	0	16-May
10664	*Dasypyrum*	*villosum*	Ukraine	0	15MR	15MR	0	H	0	0	20-May
				0	5MR	—	L	H	0	0	17-May

VIRGINIA

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2012 wheat production in the Commonwealth of Virginia.

Growing conditions. Late summer 2011 in the Commonwealth brought significant rain to most areas. However, by September, weather was favorable and corn harvest was ahead of the normal pace. This influenced wheat seeding in many areas with 26% of intended wheat acres planted by early October, compared to the 5-yr average of 7%. Precipitation in many areas at the end of October, however, resulted in only about 35% of the intended acreage being planted by the end of the third week of October. By the first week of November, growers had planted 57% of the acres they indicated they planned to plant, which was slightly below the 5-yr average of 61%. Rain in mid to late November meant planting continued at a slightly slower pace, but rain benefitted the early planted wheat and barley. December was warmer and generally wetter than normal. January and February were very mild, which left many fields far advanced and growers concerned about applying N that early and encouraging too much winter growth and increasing the likelihood of spring freeze injury. March and April were quite dry in many areas of the Commonwealth, and many fields likely experienced some yield loss due to inadequate moisture. On 20 April, growers indicated that 70% of the wheat and 57% of the barley crop were in good condition. Grain maturity came early in many areas and by 20 May, virtually all the wheat in the state was headed compared to the 5-yr average of 77% headed by this date. This trend continued and by 17 June, 98% of barley harvest and 58% of wheat harvest was complete. Initial harvest results indicated that yield and quality of the 2012 wheat and barley crops were near the long-term trend, or about average. As of 11 July, 2012, the USDA NASS Virginia Field Office estimated that Virginia's wheat producers expected to average 65 bu/ac in 2012. Wheat production in Virginia was expected to total about 17.6 x 10^6 bushels, down 1% from last year's total wheat crop of 17.8 x 10^6 bushels. Producers expected to harvest 270,000 acres of wheat, 20,000 acres more than in 2011.

Disease and insect incidence and severity. Entries in Virginia's 2012 state wheat variety trials were rated (0 = no infection to 9 = severe infection) for disease severity at four diverse locations. The 94 entries in the 2012 trial had mean powdery mildew (*Blumeria graminis*) ratings that varied from 0 to 6 over four regions of the Commonwealth in the Southwest (0–5), Southern Piedmont (0–6), Eastern Virginia (0–6), and Eastern Shore (0–5) regions. Barley/Cereal Yellow Dwarf Virus infection was moderate at Blackstone and Warsaw (1–5) and severe at Blacksburg (2–7). Fusarium head blight (*Fusarium graminearum*) was negligible. At two locations, mean trial scores for leaf rust (*Puccinia triticina*) varied from 1.1 to 3.6. Wheat entries received mean ratings from 0 to 8 in the Eastern Virginia (Warsaw) no-till trial and from 0 to 9 in the Eastern Shore (Painter) trial. Cultivars having only genes *Lr24* or *Lr26* were susceptible to leaf rust at the Eastern Shore location, whereas cultivars with gene *Lr9* or this gene combined with other genes were moderately to highly resistant. Race surveys conducted by Dr. James Kolmer of the USDA–ARS Cereal Disease Lab on 19 isolates from four regions in Virginia indentified seven races of leaf rust including MBTNB (Richmond and Suffolk counties), MCTNB (Accomack, Suffolk, and Westmoreland), MCTQB (Accomack Co.), MCTSB (Richmond Co.), MFGJG (Accomack Co.), TBJSB (Suffolk Co.), and TBRKG (Westmoreland Co.). Three races having virulence for gene *Lr26* but avirulent to gene *Lr24* included MCTNB, MCTQB, and MCTSB. Race MFGJG has virulence for genes *Lr24* and *Lr26*. None of the races identified had virulence for resistance gene *Lr9*. Stripe rust (*Puccinia striiformis*) was found in state variety trails at Blackstone, Painter, and Warsaw, VA, in 2012 and samples were sent to Dr. Xianming Chen at USDA–ARS in Pullman, WA, for race identification. Four races were identified including PSTv-37 (virulence for genes *Yr6*, *Yr7*, *Yr8*, *Yr9*, *Yr17*, *Yr27*, *Yr43*, *Yr44*, *YrTr1*, and *YrExp2*) at all three locations, race PSTv-52 (virulence for genes *Yr6*, *Yr7*, *Yr8*, *Yr9*, *Yr17*, *Yr27*, *Yr43*, *Yr44*, and *YrExp2*) at Blackstone and Painter, race PSTv-30 (virulence for genes *Yr6*, *Yr7*, *Yr8*, *Yr9*, *Yr44*, *YrTr1*, and *YrExp2*), and race PSTv-35 (virulence for genes *Yr6*, *Yr7*, *Yr8*, *Yr9*, *Yr17*, *Yr43*, *Yr44*, *YrTr1*, and *YrExp2*) only at Warsaw.

Production. According to the United States Department of Agriculture's National Agriculture Statistical Service (http://www.nass.usda.gov/Statistics_by_State/Virginia/index.asp), in the autumn of 2011 Virginia growers planted 300,000 acres (121,500 hectares) of wheat and in the spring 270,000 acres (109,350 hectares) of wheat were harvested. The average yield was 65 bu/ac (4,367 kg/ha), which was lower than the previous year. Virginia growers harvested a total 17.6 x 10^6 bushels (475,200 metric ton) of wheat in 2012.

State cultivar tests. In the 2011–12 tests, there were a total of 94 entries planted in seven environments across Virginia (http://www.grains.cses.vt.edu/). The test included 55 SRW wheat cultivars and 39 experimental lines. No-till tests, planted after corn, were conducted at Warsaw and Holland, VA. Mean grain yields varied from 67 bu/ac (4,522 kg/ha) in southwestern Virginia to 99 bu/ac (6,652 kg/ha) in the Shenandoah Valley with a mean yield over all seven locations of 78 bu/ac (5,227 kg/ha). Commercial cultivars SS 5205, USG 3555, USG 3120, Pioneer Brand 26R15, Shirley, USG 3612, USG 3251, and Merl all produced yields (82–85 bu/ac, 5,503–5,704 kg/ha) that were significantly higher than the overall trial average. Average grain yields among the 94 entries ranged from 61.4 bu/ac (4,125 kg/ha) for the long-term check cultivar Massey to 84.9 bu/ac (5,704 kg/ha) for SS 5205. Test weight means among the seven locations varied from 57.2 lb/bu (73.7 kg/hl) in the northern Piedmont to 61.3 lb/bu (78.9 kg/hl) in the Shenandoah Valley. Average test weights of the 94 entries over all seven environments ranged from 56.6 lb/bu (72.9 kg/hl) to 60.9 lb/bu (78.4 kg/hl) with an overall trial average of 59.0 lb/bu (76.0 kg/hl).

Other tests. A report at Kansas State University raised concerns about planting wheat following sorghum. In their study, it appeared that sorghum can express allelopathic effects on wheat germination and emergence when planted after sorghum with effects on yield. To evaluate this effect on our sandy soils, seedling counts were taken on 28 January, 2013, on wheat following sorghum and cotton in three fields at the Tidewater AREC, Suffolk, VA. Wheat was broadcast planted in November 2012 as a cover crop at 1 bu/a seed population. In each field, two for sorghum and one for cotton as previous crops, 5 × 1 m^2 areas were counted. The average seedling population was 85/m^2, and there were no significant differences due to previous crop, either sorghum or cotton.

Mapping studies on Fusarium head blight resistance (FHB) in the SRW wheat cultivars Roane and Jamestown were conducted using data from field and greenhouse experiments. Preliminary results indicated that QTL on chromosomes 1A, 2A, 2B, 2D, 5A, and 7B co-segregate for FHB related traits. The QTL on chromosomes 1A and 7B were associated with both resistance and susceptibility for FHB related traits, while the QTL on 2A, 2B, 2D, and 5A were associated with resistance for multiple FHB traits. Once these QTL are validated, diagnostic markers will be recommended for use in marker assisted selection for FHB resistance in wheat breeding programs.

Virginia Wheat Yield Contest Results. The 2012 contest was conducted statewide and the results can be found in the table below. The top contestants all planted no-till following corn. Congratulations to our winners!

2012 Virginia Wheat Yield Challenge Winners						
Place	Grower	FARM	COUNTY	YIELD BU/AC	PLANTING DATE	VARIETY
1	**Ronnie Russell**	Corbin Hall Farm	Middlesex	111.9	10/10/2011	Pioneer 26R20
2	**Evan Perry**	Corbin Hall Farm	Middlesex	111.1	10/12/2011	USG 3555
3	**Bill Nelson**	Colonial Acres Farm, LLC	Henrico	110.9	10/25/2011	Shirley
4	**John Copland, Jr.**	North Bend Farm	Charles City	105.9	10/17/2011	Shirley
Other Entries in the 2012 Virginia Wheat Yield Challenge						
	Tony Jones	Corbin Hall Farm	Middlesex	108.7**	10/15/2011	Pioneer 26R22
** Contest rules state that there can be no more than two prize-willing entries from the same operation						

Release of the soft red winter wheat cultivar 2013412.

The soft red winter (SRW) wheat cultivar **2013412**, formerly designated as VA06W-412, was derived from the cross 'Tribute (PI 632689) / AGS 2000 (PI 612956) // VAN99W-20 (VA90-54-631 / VA90-52-49)'. Cultivar 2013412 is a

broadly adapted, high yielding, full-season, short height semi-dwarf (gene *Rht2*) producing grain that is well suited for dual end uses in both pastry and cracker products. Spikes of cultivar 2013412 are apically awnletted, inclined, mid-dense, tapering in shape, and creamy white in color at maturity. Straw is yellow in color with trace anthocyanin visible near physiological maturity.

In the southern SRW wheat region, average head emergence of cultivar 2013412 (119 days) was 1 day later than USG3555. Average mature plant height of cultivar 2013412 has varied from 32 to 36 inches (81.3–91.4 cm) and is about 1.5 inches (3.8 cm) taller than USG 3555 and 2 inches (5.1 cm) shorter than Coker 9553. On average, straw strength (0 = erect to 9 = completely lodged) of cultivar 2013412 (0–1.3) is very good being most similar to that of Shirley (0.7–1.0) and better than that of cultivar 5187J (1.9–3.8). Cultivar 2013412 has moderate winter hardiness. In Virginia's State Variety Trial, cultivar 2013412 had a three-year (2009–2011) mean grain yield of 84.9 bu/ac (5,704 kg/ha) that was significantly ($P < 0.05$) higher than the overall trial average. Grain volume weight of cultivar 2013412 (59.6 lb/bu, 76.7 kg/hl) was significantly higher than those of nine of the ten top-yielding cultivars.

Cultivar 2013412 is resistant to powdery mildew, leaf rust, and stem rust, and has genes *Pm17*, *Lr*/*Sr24*, and the T1RS·1AL rye–wheat translocation. Cultivar 2013412 expresses an intermediate level of resistance to stripe rust in the adult-plant stages. Cultivar 2013412 is resistant to Barley and Cereal Yellow Dwarf Viruses and moderately resistant to Wheat Soil Borne Mosaic Virus and Wheat Spindle Streak Mosaic Virus. Cultivar 2013412 is moderately resistant to Septoria tritici leaf blotch, Stagonospora nodorum glume blotch, and Fusarium head blight. Although seedlings of cultivar 2013412 are susceptible to Hessian fly (*Mayetiola destructor*) biotypes B, O, and L, it expresses moderate resistance in the field.

Release of soft red winter wheat cultivar Yorktown.

Soft red winter wheat cultivar **Yorktown**, previously designated as VA08W-294, was derived from the cross '38158 (PI 19052) / VA99W-188 [(VA91-54-343 / Roane (PI 612958) sib] // Tribute (PI 632689)'. Yorktown is a broadly adapted, high yielding, full-season, short height semi-dwarf (gene *Rht2*). At maturity Yorktown has yellow colored straw, creamy white colored, slightly-tapering strap shaped, awnletted spikes. On average, head emergence of Yorktown (133 days) in the eastern SRW wheat region is similar to that of Shirley, whereas in the southern SRW wheat region, average head emergence of Yorktown (117 days) has been 1 day later than USG 3555. Average mature plant height of Yorktown has varied from 30 to 35 inches (76–89 cm) and is about one inch (2.5 cm) taller than Branson and two inches (5 cm) shorter than Bess and AGS 2000. On average, straw strength (0 = erect to 9 = completely lodged) of Yorktown (0.8–2.0) is good being most similar to that of USG 3555 (0.7–2.2) and better than that of Bess (1.2 vs. 2.3) and AGS 2000 (1.9 vs. 3.2). Winter hardiness of Yorktown is greater than that of AGS 2000 and USG 3555. Yorktown has exhibited milling and baking qualities that are most similar to those of the strong gluten cultivars Coker 9553, USG 3555, and 5187J. Yorktown was evaluated at 26 locations in the 2011 USDA–ARS Uniform Southern SRW Wheat Nursery, and ranked third in grain yield (77.8 bu/ac, 5,227 kg/ha) among 28 entries. Average grain volume weight of Yorktown (59.6 lb/bu, 76.7 kg/hl) was significantly ($P < 0.05$) higher than that of USG 3555 (58.0 lb/bu, 74.7 kg/hl). In the 2011 USDA–ARS Uniform Eastern SRW Wheat Nursery, Yorktown ranked tenth in grain yield (72.4 bu/ac, 4,865 kg/ha) among 38 entries evaluated over 28 environments. Average grain volume weight of Yorktown (58.8 lb/bu, 75.7 kg/hl) was significantly ($P < 0.05$) higher than those of the check cultivars Shirley (55.8 lb/bu, 71.9 kg/hl) and Branson (56.7 lb/bu, 73.0 kg/hl).

Yorktown is resistant to powdery mildew, leaf rust, and stem rust and has genes *Pm17* and *Lr9* and the T1RS·1AL rye–wheat translocation. Yorktown is moderately resistant in the adult plant stages to stripe rust races (PST-114 and PST-100) prevalent in the eastern U.S. Yorktown is resistant to Barley and Cereal Yellow Dwarf Viruses, but is susceptible to Wheat Soil Borne Mosaic Virus. Yorktown is moderately resistant to Septoria tritici leaf blotch and to Stagonospora nodorum leaf and glume blotch. Yorktown is most similar to the moderately resistant check cultivar Ernie in reaction to Fusarium head blight. Although seedlings of Yorktown are susceptible to Hessian fly biotypes B, C, D, O, and L, it expresses moderate resistance in the field.

Publications.

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WASHINGTON

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A natural dietary-therapy for gluten intolerance, sensitivity, and allergenicity.

Wheat, with its various cytological forms and different ploidy levels, is nourishing the world population from millennia. Wheat is believed to be a determining factor in the human transition from a hunting gathering to an agricultural society. Domestication of durum and bread wheats was a boon to humans, but it also brought unforeseen problems to the society, and continued to be a major cause of premature deaths until recently. The cause of these abdominal disorders, named 'koiliakos' by Aretaeus of Cappadocia, was long unknown until l953 when Willem-Karel Dicke declared 'gluten' from wheat and rye to be the source of these disorders. With the improvement in understanding of etiologies of a number of food-born disorders, and refinement in the diagnostic procedures we now know that 'gluten' is a cause of many diet induced health issues including gluten intolerance, sensitivity, allergies, ataxia, and dermatitis herpetiformis with a cumulative incidence of >8% in the US population.

In view of the magnitude of the problem, we undertook a multipronged approach to develop a natural dietary therapy for the gluten-induced disorders collectively referred as 'gluten syndrome'. The three approaches undertaken in this direction are i) epigenetic elimination of immunogenic prolamins (namely low-molecular-weight (LMW) glutenins and gliadins) using transgenic and nontransgenic approaches, ii) post transcriptional silencing of immunogenic prolamins via RNA interference, and iii) post translational detoxification of prolamins by ectopic expression of 'glutenases' in the wheat grains (the enzymes were selected such that they will degrade prolamins in the human gut only after their intake with food but not within the grains).

Epigenetic elimination of immunogenic prolamins. Earlier research performed on the high-lysine barley mutant Risø 1508 (*lys3a*) revealed two-types of transcriptional regulation for the prolamin genes in the barley endosperm, i) genes whose transcription depend on demethylation of their promoters in the endosperm, such as LMW glutenins and gliadins, and ii) genes whose transcription solely depend on expression of specific transcription factors, such as high molecular weight (HMW) glutenins. Based on the functional similarity between the barley *Lys3* and Arabidopsis *DEMETER* genes, we undertook cloning of barley and wheat homologues of the *DEMETER* gene (Wen et al. 2012 for details).

Induced mutagenesis of DEMETER homoeologues. The *DEMETER* sequences obtained from the wheat genome were used to develop homoeologue specific primers that were utilized to screen for mutations in the hexaploid wheat Express and tetraploid wheat Kronos TILLING populations. The screen for mutants resulted in identification of a total of 191 mutants in the Express and 77 mutants in the Kronos backgrounds.

Single mutations identified in the A- and B-subgenome *DEMETER* homoeologues of bread and durum wheat, were crossed in combinations to obtain *DEMETER* double mutants. All crosses were made reciprocally and in duplicates. The F_1 and, later, F_2 seeds were obtained for three and eight different mutant combinations respectively in Kronos and Express backgrounds. The mutations for crossing were selected on the basis of their molecular nature (i.e., substitution, splice site variation or premature stop codon), respective location in the *DEMETER* active site, and level of transcriptional suppression of *DEMETER* homoeologues and accumulation of prolamins. The F_2 grains obtained from the aforementioned crosses were propagated in 48-well flats and are currently being analyzed for homo-/heterozygous double mutations by PCR followed by sequencing of the PCR products. Preliminary analysis allowed identification of 20 double mutations in Kronos background and 14 double mutants in Express background. These double mutants were transferred to larger pots and raised till maturity to obtain F_3 seed. Spikes of 19 *DEMETER* double mutants identified in Kronos background were collected to extract RNA from immature grains followed by qRT–PCR analysis. Results of qRT–PCR showed reduction in *DEMETER* transcript abundance in relation to actin and glyceraldehyde 3-phosphate dehydrogenase in eight cases. We have not expected to see transcriptional suppression in the double mutants, because single mutants used to obtain these double mutants are either amino acid substitutions or splice-site variants, which in principle should not lead to transcriptional suppression, although we expected reduction in amount of immunogenic prolamins accumulated in the endosperm. Very little reduction in amount of immunogenic prolamins was observed for the double mutants, which, in all confirmed cases, showed pyramiding of amino acid substitutions.

Interestingly, in contrary to the barley high-lysine mutants, *DEMETER* mutants carrying premature stop codons or splice site variants especially in tetraploid wheat background showed reduction in anther size, locule size, number of fertile pollens, and pollen germination rate. Reduced level of pollen germination adversely affected competitiveness of the mutant pollens in relation to the wild type pollens causing severe segregation distortion. In fact, analysis of >2000 F_2 plants from crosses between A- and B-subgenome *DEMETER* mutants with premature stop codons have so far yielded either mutant allele at a single loci or wild type alleles at both homoeoloci. A reduction in seedling vigor of the *DEMETER* mutants with stop codons was also observed. Both of these observations indicated towards the vital role of *DEMETER* during microspore/pollen development and seed germination. This is suggesting a RNA interference (RNAi) based approach, expressing *DEMETER* silencing microRNAs in a spatio–temporal fashion as an alternative.

Silencing wheat DEMETER genes using RNA interference (RNAi). A RNAi based approach was followed to develop *DEMETER*-deficient wheat lines with no influence on other vital plant processes. In order to achieve this objective, transformants exclusively expressing *DEMETER* targeting micro/small interfering RNAs in wheat endosperm where developed. A total of 333 candidate transformants were obtained by four rounds of biolistic transformations using five different constructs, where three express artificial microRNAs (amiRNAs, namely pRB104, pRB105, and pRB106) and two express hairpin RNA (hpRNA, namely p728 and pDRB6). All of the above single-cassette vectors were co-transformed in 2:1 proportion with another single-cassette vector (pDPG165) expressing bialaphos resistance gene (*bar*) gene under the control of the Cauliflower Mosaic Virus 35S promoter and nopaline synthase (nos) terminator. A biolistic-based approach was used to transform scutellar calli derived from two soft white winter wheat cultivars Brundage 96 and Simon with the above mentioned constructs. Plants recovered from the tissue culture were transferred to soil and vernalized for 8-12 weeks. After vernalization, the plants were transplanted to 6-inch pots, and their leaves were painted with 2% Ignite solution (active ingredient biolaphos) and data were recorded for injury on 0–5 scale, where 0 represents no injury and 5 represents dead tissue. Two weeks after transplanting, leaf tissue was collected from candidate transformants (T_0), and DNA was extracted to study integration of artificial microRNA/hairpin RNA expressing cassettes in the wheat genome. RNA was also extracted from developing T_1 seed (harvested 17±3 days post anthesis) of the putative transformants. The results of qRT–PCR showed suppression in *DEMETER* transcript abundance ranging from 3.0 to 85.2% in individual transformants.

On the basis of the results of PCR and quantitative real time PCR (qRT–PCR) analysis a set of 52 transformants (T_0) was selected for further analysis. Out of the 52 plants, 50 gave seed (T_1) that were used for protein extraction following the protocol described in Wieser et al. (1998) with minor modifications. The three different fractions albumins/globulins (salt soluble fraction), gliadins (aqueous alcohol soluble fraction), and glutenins (soluble in aqueous alcohol with reducing agents) were extracted from the T_1 seed obtained from each transformant and analyzed using denaturing sodium dodecyl sulfate–polyacrylamide gel electrophoresis (SDS–PAGE).

The extracted proteins were first quantified using Bradford colorimetric assay followed by quantitative (by loading equal volume of extracted proteins obtained from same amount of starting seed material) and qualitative analysis (by loading equimolar amounts of proteins) on PAGE and/or high-performance liquid chromatography (HPLC). Preliminary results of PAGE analysis and reverse phase (RP)–HPLC revealed elimination of specific gliadins (generally in γ- and/or

α/β-gliadin fractions) and glutenins (in LMW glutenin fraction) instead of gross eliminations. These results were validated by improving resolution of the analysis by collecting and analyzing three HPLC traces (based on retention times) from gliadin fraction and five HPLC traces from glutenin fraction on the time-of-flight mass spectrometer. Elimination of specific prolamins instead of gross eliminations was attributed to the bulk harvest of all T_1 seed from individual T_0 plants, which were most likely chimeric in nature, and have resulted in dilution effect in the protein gels and RP–HPLC profiles. In order to deal with the problem of chimerism, we propagated 2,620 T_1 progeny plants in a glasshouse, which represents 10% of the seed derived the 50 T_0 transformants. DNA was extracted from these T_1s and faithful inheritance of transgene integrations (expressing *DEMETER* silencing hp/amiRNAs) was confirmed in 522 of 2,620 cases using construct-specific primers. Out of 522 cases, 242 represent T_1s expressing one of the three amiRNAs (namely pRB104, pRB105, and pRB106), and 280 represent T_1s expressing the hpRNA (plasmid 728). The T_1 plants showing inheritance of transgene integration(s) in hemi-/homozygous state were transferred to individual pots, and the spikes carrying immature T_2 seed (collected after 17±3 days post anthesis) were collected from glasshouse to analyze for transcriptional suppression of *DEMETER* homoeologues. Mature T_2 grains were collected from these transformants to study effect of *DEMETER* suppression/silencing on prolamin accumulation.

DNA analysis of the T_1 plants showed faithful inheritance of the transgene in 2.8 to 71.7% of progeny plants belonging to different transformants. A wide range of plants showing transgene inheritance in the T_1 progeny confirmed chimeric origin of these plants. Similarly, the results of qRT–PCR analysis performed on the cDNAs prepared from the immature T_2 seed belonging to different transformant families showed a wide suppression range from 3.5 to 85.6%.

The selected transformants showing suppression in *DEMETER* transcript abundance close to or more than 50% were examined for the accumulation of immunogenic prolamins in their endosperm by sequential extraction of seed storage proteins followed by SDS–PAGE and RP–HPLC. The PAGE and HPLC results revealed that different transformants show elimination of different prolamin groups, for instance P32F2 and P31D12, progeny of T_0 transformant 10-728, showed reduced accumulation of α-gliadins respectively by 56.2 and 37.6% and γ-gliadins by 21.8% and 20.7%. These lines also show reduced accumulation for LMWgs, which together with gliadins, account for ~67% reduction in the amount of total immunogenic prolamins. However a slight increase in the amount of ω-gliadins and a significant increase in the amount of HMWgs was also observed in these lines, which will be beneficial in maintaining the rheological properties of these lines. Not only reduction in amount of immunogenic prolamins but also total elimination of specific gliadin members/groups and/or LMWg subunits was also observed in different transformants.

The level of reduction for immunogenic prolamins (LMWgs and gliadins) calculated using the area covered by specific prolamin groups on the HPLC chromatograms ranged from 45.2–76.4%, when used as independent variable to regress on the values obtained for DEMETER transcript level expressed as percent suppression. The two variables regressed very well on each other with a r^2 value of 0.877 suggesting 87.7% correspondence among the two variables indicating that level of *DEMETER* transcript determines the amount and type of immunogenic prolamins accumulated in the wheat grain.

In summary, the qRT–PCR analysis with immature T_2 seed followed by protein profiling with three individual mature seed/T_1 showed reduced accumulation of immunogenic prolamins in 48 cases expressing amiRNAs (representing 22 transformation events) and 50 cases expressing hpRNA (representing 13 transformation events). Out of these 98 T_1s, 19 (representing a total of 15 transformation events, 10 expressing amiRNA and five expressing hpRNA) showed the highest level of reduction in the amount of immunogenic prolamins, which were selected for propagation in the glasshouse. In view of multiple integrations and zygosity of transgene at each integration site 10 T_2 seed/T_1 were propagated in the glasshouse. The plants propagated in glasshouse will be converted to doubled haploids (DHs) using the microspore culture method standardized in our laboratory (Santra et al. 2012). The plants are expected to attain the desired developmental stage (i.e., Feeke's stage 10–10.1) for microspore collection by the end of May 2013. Once obtained each doubled haploid (DH) line will be evaluated for its prolamin content using SDS–/Acid–PAGE followed by RP–HPLC. The DH lines showing elimination of different prolamin families will be crossed to pyramid their effect in a single genotype.

Chimeric TALE (transcription-activator like effector) suppressor for complete silencing of DEMETER homoeologues. With the current transformation methods, there is no control on the number and site of transgene integration in the genome, thus, it is difficult to obtain a transformant showing complete or near complete suppression of the targeted gene. Specifically in cases when dealing with a gene family with members having redundant or overlapping functions, and/or working with a polyploidy organism. However, pyramiding the effects of different transformants each showing partial suppression of gene activity is a viable option, which will be followed, but it is a labor-intensive and time-consuming approach. To assure elimination of all immunogenic prolamins, we are currently developing chimeric suppressor

of *DEMETER* activity. By engineering the DNA binding domain of a *Xanthomonas transcription*-activator like effector (TALE) to exclusively recognize wheat *DEMETER* homoeologues, and attaching it with the *Arabidopsis* EAR-repression domain (SRDX). This chimeric suppressor will be provided with wheat endosperm specific HMW glutenin 1Dy promoter and nos terminator to specifically silence *DEMETER* homoeologues in the developing endosperm. The above construct will be introduced in the already identified transformants showing >75% reduction in the accumulation of immunogenic prolamin to achieve 100% elimination.

TAL effectors are a novel class of DNA binding proteins produced in the plant pathogenic bacteria of the genus Xanthomonas. A cocktail of different virulent effectors are transferred via a type-III secretion system into plant cells, where they translocate to the nucleus with the aid of a nucleus localization signal, bind to the target promoter, and induce expression of the plant gene. A domain of 11 to 18 repeats of 34 amino acids each determines the DNA-binding specificity of these effectors. Each repeat binds to one base pair determined by the repeat variable diresidue at position 12 and 13 that is Asn**x**Ile to A, His**x**Asp to C, Asn**x**Gly to T, and Asn**x**Lys to G. Rearrangement of the repeat sequences in the DNA binding domain allow its binding to any desired DNA sequence in the genome.

Post transcriptional silencing of immunogenic prolamins via RNA interference. For the post-transcriptional silencing of the immunogenic prolamins a chimeric hairpin construct was assembled by putting together the conserved target sequences identified for different gliadin groups and the LMW glutenin subunits. The hairpin was provided with the endosperm specific HMW glutenin 1Dy promoter, and introduced into the wheat genome by biolistic approach or via microspore electroporation based transformation method. The T_1 seed of the selected transformants are currently being examined for their prolamin content.

Post translational detoxification of prolamins by expression of 'glutenases' in wheat grains. *In vitro* and *in vivo* studies have demonstrated that sequential treatment of prolamins with a glutamine specific endoprotease and a post-proline cleaving endopeptidase completely detoxifies prolamins, and the enzyme combination can be used as a dietary therapy for the celiac disease. Thus, to make this therapy easily assessable to the patients, we undertook a transgenic approach to introduce an endoprotease and an endopeptidase in the wheat grains. In order to achieve this objective, evaluation of different 'glutenases' was undertaken, in combination with expression and stability analysis of the selected enzymes. To identify the best combination of glutenases, a virtual digestion of 1,336 prolamin sequences under simulated gastric conditions was performed. These sequences include α/β-, γ- and ω-gliadins, low- and high-molecular-weight glutenins from wheat, B-, C-, γ-, and D-hordeins from barley, and γ-, ω-, and high-molecular-weight secalins from rye. The virtual digestions were performed using a suit of Visual Basic (version 6) based macros. These macros were designed using information available for the cleavage site specificities of different digestive enzyme and glutenases. After virtual digestion, resulting peptides were checked for the presence of 44 immunogenic peptides documented in the literature (cf. Osorio et al. 2012). Results of this analysis revealed *Flavobacterium meningosepticum* prolyl endopeptidase (Fm-PEP) and barley cysteine endoprotease B2 (EP-B2) as the most efficient combination of enzymes for complete/near complete detoxification of gluten proteins. Based on these results a construct expressing Fm-PEP and EP-B2 under the control of wheat high molecular weight glutenin 1Dy promoter was assembled and introduced in the wheat scutellar calli using biolistic approach. To direct these enzymes to the protein storage vacuoles the genes encoding these proteins were fused with the sequence coding for the barley D-hordein signal peptide. Integration of these genes in the wheat genome was confirmed at the T_0 and T_1 generations, and the transformants showing gene integrations are currently being investigated for the expression of these enzymes.

To ensure survival of these enzymes during the baking process a site-directed mutagenesis approach was followed to introduce mutations in the first and the seventh blades of the b-propeller domain of the Fm-PEP, and at the junction of left and right-domains in EP-B2. Additionally, EP-B2 was introduced as a proenzyme in the wheat endosperm, where the pro-peptide is known to function as a chaperone during enzyme refolding after denaturation. The pro-peptide is removed from the enzyme by an autocatalytic activity that takes place only at a pH below 4, which does not encounter either during the grain development or dough making process. This property of proEP-B2 is highly beneficial for the present research in two ways; i) by providing enzyme stability under the baking conditions and ii) by avoiding degradation of prolamins within the grains and dough-making process.

Using a sequence homology-based approach, nine loci were selected to introduce point mutations in the Fm-PEP gene, and a PCR-based method was followed to introduce these mutations. The process has resulted in generation of a library of 66 mutants. Expression of these enzyme isoforms was performed in *E. coli*. After purification the enzyme isoforms were tested for their thermostability under temperatures ranging from 60–90°C with an increment of 10°C.

Kinetics parameters were calculated using synthetic substrate. Results of this analysis demonstrate that mutations at residues 412, 413, 414, and 415 increase thermostability of Fm-PEP under in vitro conditions. Digestion of reference gluten material under simulated gastrointestinal conditions using the thermostable Fm-PEP isoforms is currently underway.

Similarly, in view of the structural and functional similarities between a thermostable Ervatamin (ERV), ERVC and barley cysteine endoprotease B2, we used ERVC as a template to make alterations in EP-B2 with an expectation to increase its thermostability. Primers were designed to introduce mutations at the Val32, Gly36, and Lys174 to Ser, Ser, and Arg. These three substitutions were already introduced in the *EP-B2* gene by specifically designed primers and assembled in one fragment by splicing by overlap extension (SOE) PCR, and the enzyme isoform is currently being tested for its thermostability.

In a nutshell our results show a great potential in obtaining non-immunogenic wheat cultivars, which will have a broad impact on the quality of lives of a vast majority of the wheat consumers.

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The mission of the lab is two-fold: conduct milling, baking, and end-use quality evaluations on wheat breeding lines, and conduct research on wheat grain quality and utilization. Our web site: http://www.wsu.edu/~wwql/php/index.php provides great access to our research and methodology. Our research publications are available on our web site.

Morris and Engle lead the Pacific Northwest Wheat Quality Council, a consortium of collaborators who evaluate the quality of new cultivars and advanced breeding lines. We also conduct the U.S. Wheat Associates' Overseas Varietal Analysis Program for Soft White and Club Wheat. Our current activities and projects include grain hardness and puroindolines, waxy wheat, polyphenol oxidase (PPO), arabinoxylans, SDS sedimentation test, and soft durum wheat.

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National Small Grains Collection activities.

H.E. Bockelman, Agronomist and Curator.

Recent PI Assignments in Triticum, X Triticosecale, Aegilops, and Secale.

Passport and descriptor data for these new accessions can be found on the Germplasm Resources Information Network (GRIN): http://www.ars-grin.gov/npgs. Certain accessions may not be available from the National Small Grains Collection due to intellectual property rights, quarantine, or insufficient inventories. Accessions registered in the *Journal of Plant Registrations* or *Crop Science* are available by contacting the developers.

Table 1. Recent PI assignments in *Triticum, Aegilops*, and *X Triticosecale*. There were no PI assignments in *Aegilops* or *Secale* in the past year.

PI number	Taxonomy	Cultivar name or identifier	Country	State/Province
664944 PVPO	*Triticum aestivum* subsp. *aestivum*	SY 901	United States	Colorado
664945 PVPO	*Triticum aestivum* subsp. *aestivum*	SY Southwind	United States	Colorado
664944 PVPO	*Triticum aestivum* subsp. *aestivum*	SY 901	United States	Colorado
664945 PVPO	*Triticum aestivum* subsp. *aestivum*	SY Southwind	United States	Colorado
664946 PVPO	*Triticum aestivum* subsp. *aestivum*	SY Harrison	United States	Colorado
664947 PVPO	*Triticum aestivum* subsp. *aestivum*	SY 483	United States	Colorado
664948 PVPO	*Triticum aestivum* subsp. *aestivum*	SY 107	United States	Colorado
664949 PVPO	*Triticum aestivum* subsp. *aestivum*	W020088N1	United States	Indiana
664950 PVPO	*Triticum aestivum* subsp. *aestivum*	W020054B1	United States	Indiana
664951 PVPO	*Triticum aestivum* subsp. *aestivum*	W010712A1	United States	Indiana
664952 PVPO	*Triticum aestivum* subsp. *aestivum*	W010684M1	United States	Indiana
665019 PVPO	*Triticum aestivum* subsp. *aestivum*	LCS Powerplay	United States	Colorado
665038	*Triticum aestivum* subsp. *aestivum*	12V51	United States	Virginia
665039	*Triticum aestivum* subsp. *aestivum*	5187J	United States	Virginia
665040	*Triticum aestivum* subsp. *aestivum*	2013911	United States	Virginia
665047	*Triticum aestivum* subsp. *aestivum*	ARS-Amber	United States	Washington
665048	*Triticum aestivum* subsp. *compactum*	ARS-Crescent	United States	Washington
665049	*Triticum aestivum* subsp. *compactum*	ARS-Chrystal	United States	Washington
665061 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-Prestea	United States	Montana
665062 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-Grainfield	United States	Kansas
665063 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-Redhawk	United States	Kansas
665064 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-Gunnison	United States	Montana
665065 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-Hartline	United States	Idaho
665066 PVPO	*Triticum turgidum* subsp. *durum*	Westmore	United States	Arizona
665417 PVPO	*Triticum aestivum* subsp. *aestivum*	Velva	United States	North Dakota
665473	*Triticum turgidum*	DGE-3	United States	North Dakota
665493 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-700	United States	Indiana

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PI number	Taxonomy	Cultivar name or identifier	Country	State/Province
665494 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-DeuceCL+	United States	Kansas
665495 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-1035CL+	United States	Washington
665696	*Triticum aestivum* subsp. *aestivum*	JS13	United States	Montana
665697	*Triticum aestivum* subsp. *aestivum*	JS14	United States	Montana
665698	*Triticum aestivum* subsp. *aestivum*	JS15	United States	Montana
665699	*Triticum aestivum* subsp. *aestivum*	JS16	United States	Montana
665700	*Triticum aestivum* subsp. *aestivum*	JS18	United States	Montana
665701	*Triticum aestivum* subsp. *aestivum*	JS2	United States	Montana
665702	*Triticum aestivum* subsp. *aestivum*	JS28	United States	Montana
665703	*Triticum aestivum* subsp. *aestivum*	JS32	United States	Montana
665704	*Triticum aestivum* subsp. *aestivum*	JS33	United States	Montana
665705	*Triticum aestivum* subsp. *aestivum*	JS34	United States	Montana
665706	*Triticum aestivum* subsp. *aestivum*	JS35	United States	Montana
665707	*Triticum aestivum* subsp. *aestivum*	JS36	United States	Montana
665708	*Triticum aestivum* subsp. *aestivum*	JS37	United States	Montana
665709	*Triticum aestivum* subsp. *aestivum*	JS40	United States	Montana
665710	*Triticum aestivum* subsp. *aestivum*	JS49	United States	Montana
665711	*Triticum aestivum* subsp. *aestivum*	JS5	United States	Montana
665931	*Triticum aestivum* subsp. *aestivum*	ND 803	United States	North Dakota
665932 PVPO	*Triticum aestivum* subsp. *aestivum*	Greenville	United States	Utah
665947 PVPO	*Triticum aestivum* subsp. *aestivum*	Mattern	United States	Nebraska
665948 PVPO	*Triticum aestivum* subsp. *aestivum*	Clara CL	United States	Kansas
665951	*Triticum turgidum* subsp. *durum*	Kandur 1101	United States	Kansas
665952	*Triticum turgidum* subsp. *durum*	Kandur 1102	United States	Kansas
665953	*Triticum turgidum* subsp. *durum*	Kandur 1103	United States	Kansas
665954	*Triticum turgidum* subsp. *durum*	Kandur 1104	United States	Kansas
665955	*Triticum turgidum* subsp. *durum*	Kandur 1105	United States	Kansas
665956	*Triticum turgidum* subsp. *durum*	Kandur 1106	United States	Kansas
665957	*Triticum turgidum* subsp. *durum*	Kandur 1107	United States	Kansas
665958	*Triticum turgidum* subsp. *durum*	Kandur 1108	United States	Kansas
665959	*Triticum turgidum* subsp. *durum*	Kandur 1109	United States	Kansas
665960	*Triticum turgidum* subsp. *durum*	Kandur 1110	United States	Kansas
665961	*Triticum turgidum* subsp. *durum*	Kandur 1111	United States	Kansas
665962	*Triticum turgidum* subsp. *durum*	Kandur 1112	United States	Kansas
665963	*Triticum turgidum* subsp. *durum*	Kandur 1113	United States	Kansas
665964	*Triticum turgidum* subsp. *durum*	Kandur 1114	United States	Kansas
665965	*Triticum turgidum* subsp. *durum*	Kandur 1115	United States	Kansas
665966	*Triticum turgidum* subsp. *durum*	Kandur 1116	United States	Kansas
665967	*Triticum turgidum* subsp. *durum*	Kandur 1117	United States	Kansas
665968	*Triticum turgidum* subsp. *durum*	Kandur 1118	United States	Kansas
665969	*Triticum turgidum* subsp. *durum*	Kandur 1119	United States	Kansas
665970	*Triticum turgidum* subsp. *durum*	Kandur 1120	United States	Kansas
665971	*Triticum turgidum* subsp. *durum*	Kandur 1121	United States	Kansas
665972	*Triticum turgidum* subsp. *durum*	Kandur 1122	United States	Kansas
665973	*Triticum turgidum* subsp. *durum*	Kandur 1123	United States	Kansas
665974	*Triticum turgidum* subsp. *durum*	Kandur 1124	United States	Kansas
665975	*Triticum turgidum* subsp. *durum*	Kandur 1125	United States	Kansas

Table 1. Recent PI assignments in *Triticum, Aegilops*, and *X Triticosecale*. There were no PI assignments in *Aegilops* or *Secale* in the past year.

PI number	Taxonomy	Cultivar name or identifier	Country	State/Province
665976	*Triticum turgidum* subsp. *durum*	Kandur 1126	United States	Kansas
665977	*Triticum turgidum* subsp. *durum*	Kandur 1127	United States	Kansas
665978	*Triticum turgidum* subsp. *durum*	Kandur 1128	United States	Kansas
665979	*Triticum turgidum* subsp. *durum*	Kandur 1129	United States	Kansas
665980	*Triticum turgidum* subsp. *durum*	Kandur 1130	United States	Kansas
665981	*Triticum turgidum* subsp. *durum*	Kandur 1131	United States	Kansas
665982	*Triticum turgidum* subsp. *durum*	Kandur 1132	United States	Kansas
665983	*Triticum turgidum* subsp. *durum*	Kandur 1133	United States	Kansas
665984	*Triticum turgidum* subsp. *durum*	Kandur 1134	United States	Kansas
665985	*Triticum turgidum* subsp. *durum*	Kandur 1135	United States	Kansas
665986	*Triticum turgidum* subsp. *durum*	Kandur 1136	United States	Kansas
665987	*Triticum turgidum* subsp. *durum*	Kandur 1137	United States	Kansas
665988	*Triticum turgidum* subsp. *durum*	Kandur 1138	United States	Kansas
665989	*Triticum turgidum* subsp. *durum*	Kandur 1139	United States	Kansas
665990	*Triticum turgidum* subsp. *durum*	Kandur 1140	United States	Kansas
665991	*Triticum turgidum* subsp. *durum*	Kandur 1141	United States	Kansas
665992	*Triticum turgidum* subsp. *durum*	Kandur 1142	United States	Kansas
665993	*Triticum turgidum* subsp. *durum*	Kandur 2071	United States	Kansas
665994	*Triticum turgidum* subsp. *durum*	Kandur Population B1/ B6 United States Kansas	Syria	
665995	*Triticum turgidum* subsp. *durum*	Kandur Population E8/ E545 United States Kansas	Syria	
666046 PVPO	*Triticum aestivum* subsp. *aestivum*	WB9879CLP	United States	Montana
666125 PVPO	*Triticum aestivum* subsp. *aestivum*	TAM 113	United States	Texas
666126 PVPO	*Triticum aestivum* subsp. *aestivum*	WB9229	United States	California
666127 PVPO	*Triticum turgidum* subsp. *durum*	WB-Mohave	United States	Arizona
666128 PVPO	*X Triticosecale spp*.	HyOctane	Canada	Ontario
666365 PVPO	*Triticum aestivum* subsp. *aestivum*	XW10Q	United States	Indiana
666366 PVPO	*Triticum aestivum* subsp. *aestivum*	XW10S	United States	Indiana
666367 PVPO	*Triticum aestivum* subsp. *aestivum*	XW10V	United States	Indiana
666368 PVPO	*Triticum aestivum* subsp. *aestivum*	XW10T	United States	Indiana
666369 PVPO	*Triticum aestivum* subsp. *aestivum*	W010F00J3	United States	Indiana
666370 PVPO	*Triticum aestivum* subsp. *aestivum*	W020189J1	United States	Indiana
666371 PVPO	*Triticum aestivum* subsp. *aestivum*	W011031I1	United States	Indiana
666372 PVPO	*Triticum aestivum* subsp. *aestivum*	W020544C1	United States	Indiana
666373 PVPO	*Triticum aestivum* subsp. *aestivum*	Mary	United States	Oregon
666374 PVPO	*Triticum aestivum* subsp. *aestivum*	W030085A1	United States	Indiana
666375 PVPO	*Triticum aestivum* subsp. *aestivum*	W020852C1	United States	Indiana
666376 PVPO	*Triticum aestivum* subsp. *aestivum*	W020757F1	United States	Indiana
666377 PVPO	*Triticum aestivum* subsp. *aestivum*	W020556F1	United States	Indiana
666378 PVPO	*Triticum aestivum* subsp. *aestivum*	W020536B1	United States	Indiana
666379 PVPO	*Triticum aestivum* subsp. *aestivum*	T173	United States	Colorado
666940	*Triticum aestivum* subsp. *aestivum*	Glee	United States	Washington
666941	*Triticum aestivum* subsp. *aestivum*	Dayn	United States	Washington
666962 PVPO	*Triticum aestivum* subsp. *aestivum*	Patwin-515	United States	California
667038	*Triticum aestivum* subsp. *aestivum*	NE06545	United States	Nebraska
667096	*Triticum aestivum* subsp. *aestivum*	Dy10-DLC	United States	California

Table 1. Recent PI assignments in *Triticum*, *Aegilops*, and *X Triticosecale*. There were no PI assignments in *Aegilops* or *Secale* in the past year.

PI number	Taxonomy	Cultivar name or identifier	Country	State/Province
667102 PVPO	*Triticum aestivum* subsp. *aestivum*	LCS Artdeco	United States	Colorado
667103 PVPO	*Triticum aestivum* subsp. *aestivum*	LCS Breakaway	United States	Minnesota
667104 PVPO	*Triticum aestivum* subsp. *aestivum*	Norden	United States	Minnesota
667557	*Triticum aestivum* subsp. *aestivum*	Otto	United States	Washington
667569 PVPO	*Triticum aestivum* subsp. *aestivum*	Gallagher	United States	Oklahoma
667570 PVPO	*Triticum aestivum* subsp. *aestivum*	Iba	United States	Oklahoma
667606	*Triticum turgidum* subsp. *durum*	NW03Y1066	United States	Nebraska
667607	*Triticum turgidum* subsp. *durum*	NW03Y1069	United States	Nebraska
667608	*Triticum turgidum* subsp. *durum*	NW03Y1072	United States	Nebraska
667609	*Triticum turgidum* subsp. *durum*	NW03Y1088	United States	Nebraska
667610	*Triticum turgidum* subsp. *durum*	NW03Y1090	United States	Nebraska
667611	*Triticum turgidum* subsp. *durum*	NW03Y1092	United States	Nebraska
667612	*Triticum turgidum* subsp. *durum*	NW03Y1094	United States	Nebraska
667613	*Triticum turgidum* subsp. *durum*	NW03Y1099	United States	Nebraska
667614	*Triticum turgidum* subsp. *durum*	NW03Y1123	United States	Nebraska
667615	*Triticum turgidum* subsp. *durum*	NW03Y1127	United States	Nebraska
667616	*Triticum turgidum* subsp. *durum*	NW03Y1129	United States	Nebraska
667617	*Triticum turgidum* subsp. *durum*	NW03Y1132	United States	Nebraska
667618	*Triticum turgidum* subsp. *durum*	NW03Y1131	United States	Nebraska
667619	*Triticum turgidum* subsp. *durum*	NW03Y1135	United States	Nebraska
667620	*Triticum turgidum* subsp. *durum*	NW03Y1143	United States	Nebraska
667621	*Triticum turgidum* subsp. *durum*	NW03Y1144	United States	Nebraska
667642 PVPO	*Triticum aestivum* subsp. *aestivum*	Vision 45	United States	Virginia
667643 PVPO	*Triticum aestivum* subsp. *aestivum*	Yorktown	United States	Virginia
667644 PVPO	*Triticum aestivum* subsp. *aestivum*	2013412	United States	Virginia
667645 PVPO	*Triticum turgidum* subsp. *durum*	VT Peak	Canada	Saskatchewan
667680 PVPO	*Triticum aestivum* subsp. *aestivum*	WB4458	United States	Illinois
667681 PVPO	*Triticum aestivum* subsp. *aestivum*	WB1997	United States	Illinois
667682 PVPO	*Triticum aestivum* subsp. *aestivum*	WB2334	United States	Illinois
663870	*Triticum aestivum* subsp. *aestivum*	KS12WGGRC55	United States	Kansas
663945	*Triticum aestivum* subsp. *aestivum*	Kaixian-luohanmai	China	Sichuan
663950	*Triticum aestivum* subsp. *aestivum*	Fieldstar	Canada	Manitoba
664028	*Triticum aestivum* subsp. *aestivum*		Kharoba	Morocco
664077	*Triticum aestivum* subsp. *aestivum*	PI350078-sel-awl	United States	Idaho
664078	*Triticum aestivum* subsp. *aestivum*	E5024	United States	Michigan
664079	*Triticum aestivum* subsp. *aestivum*	Jupiter	United States	Michigan
664088 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-Tucson	United States	Montana
664089 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-Perla	United States	Arizona
664090 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-Patron	United States	Arizona
664091 PVPO	*Triticum turgidum* subsp. *durum*	WB-Mead	United States	Arizona
664092 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-1070CL	United States	Montana
664093 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-Quake	United States	Montana
664094 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-249	United States	Indiana
664095 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-196	United States	Indiana
664096 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-139	United States	Indiana
664097 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-112	United States	Indiana
664098 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-Joaquin Oro	United States	Arizona

Table 1. Recent PI assignments in *Triticum, Aegilops*, and *X Triticosecale*. There were no PI assignments in *Aegilops* or *Secale* in the past year.

PI number	Taxonomy	Cultivar name or identifier	Country	State/Province
664099 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-1066CL	United States	Montana
664100 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-Junction	United States	Montana
664101 PVPO	*Triticum aestivum* subsp. *aestivum*	WB-Arrowhead	United States	Montana
664102 PVPO	*Triticum turgidum* subsp. *durum*	Tipai	United States	California
664103 PVPO	*Triticum aestivum* subsp. *aestivum*	Dinero	United States	Colorado
664250	*Triticum aestivum* subsp. *aestivum*	NC10-23603	United States	North Carolina
664251	*Triticum aestivum* subsp. *aestivum*	NC8889-8	United States	North Carolina
664252	*Triticum aestivum* subsp. *aestivum*	NC8860-5	United States	North Carolina
664253	*Triticum aestivum* subsp. *aestivum*	NC8889-2A	United States	North Carolina
664254	*Triticum aestivum* subsp. *aestivum*	NC8889-3	United States	North Carolina
664255 PVPO	*Triticum aestivum* subsp. *aestivum*	Brawl CL Plus	United States	Colorado
664256	*Triticum aestivum* subsp. *aestivum*	Denali	United States	Colorado
664257	*Triticum aestivum* subsp. *aestivum*	Byrd	United States	Colorado
664268 PVPO	*Triticum aestivum* subsp. *aestivum*	New Dirkwin	United States	California
664269 PVPO	*Triticum aestivum* subsp. *aestivum*	W000582Z2	United States	Indiana
664270 PVPO	*Triticum aestivum* subsp. *aestivum*	XW09H	United States	Indiana
664271 PVPO	*Triticum aestivum* subsp. *aestivum*	Rimrock	Canada	Ontario
664272 PVPO	*Triticum aestivum* subsp. *aestivum*	VA258	United States	Virginia
664301	*Triticum aestivum* subsp. *aestivum*	CO08RWA050	United States	Colorado
664304 PVPO	*Triticum aestivum* subsp. *aestivum*	Bruneau	United States	Idaho
664480 MAP	*Triticum aestivum* subsp. *aestivum*	Becker	United States	Virginia
664481 MAP	*Triticum aestivum* subsp. *aestivum*	Massey	United States	Virginia
664482	*Triticum aestivum* subsp. *aestivum*	Advance	United States	South Dakota
664483	*Triticum aestivum* subsp. *aestivum*	Forefront	United States	South Dakota
664549	*Triticum aestivum* subsp. *aestivum*	UC1110 (5+10) + *Yr36*-GPC	United States	California
664556 PVPO	*Triticum aestivum* subsp. *aestivum*	SY 314	United States	California
664557 PVPO	*Triticum aestivum* subsp. *aestivum*	SY Summit 515	United States	California
664558 PVPO	*X Triticosecale spp*.	SY 115T	United States	California
664559 PVPO	*X Triticosecale spp*.	SY 158T	United States	California
664560 PVPO	*X Triticosecale spp*.	SY 141T	United States	California
GSTR (Genetic Stocks – *Triticum*)				
401	*Triticum aestivum* subsp. *aestivum*	*Lr1*	Canada	Manitoba
402	*Triticum aestivum* subsp. *aestivum*	*Lr1*	Canada	Manitoba
403	*Triticum aestivum* subsp. *aestivum*	*Lr2a*	Canada	Manitoba
404	*Triticum aestivum* subsp. *aestivum*	*Lr2b*	Canada	Manitoba
405	*Triticum aestivum* subsp. *aestivum*	*Lr2c*	Canada	Manitoba
406	*Triticum aestivum* subsp. *aestivum*	*Lr3*	Canada	Manitoba
407	*Triticum aestivum* subsp. *aestivum*	*Lr3bg*	Canada	Manitoba
408	*Triticum aestivum* subsp. *aestivum*	*Lr3ka*	Canada	Manitoba
409	*Triticum aestivum* subsp. *aestivum*	*Lr9*	Canada	Manitoba
410	*Triticum aestivum* subsp. *aestivum*	*Lr10*	Canada	Manitoba
411	*Triticum aestivum* subsp. *aestivum*	*Lr11*	Canada	Manitoba
412	*Triticum aestivum* subsp. *aestivum*	*Lr12*	Canada	Manitoba
413	*Triticum aestivum* subsp. *aestivum*	*Lr13*	Canada	Manitoba
414	*Triticum aestivum* subsp. *aestivum*	*Lr14a*	Canada	Manitoba
415	*Triticum aestivum* subsp. *aestivum*	*Lr14b*	Canada	Manitoba
416	*Triticum aestivum* subsp. *aestivum*	*Lr15*	Canada	Manitoba

Table 1. Recent PI assignments in *Triticum*, *Aegilops*, and *X Triticosecale*. There were no PI assignments in *Aegilops* or *Secale* in the past year.

PI number	Taxonomy	Cultivar name or identifier	Country	State/Province
417	*Triticum aestivum* subsp. *aestivum*	*Lr16*	Canada	Manitoba
418	*Triticum aestivum* subsp. *aestivum*	*Lr17*	Canada	Manitoba
419	*Triticum aestivum* subsp. *aestivum*	*Lr18*	Canada	Manitoba
420	*Triticum aestivum* subsp. *aestivum*	*Lr19*	Canada	Manitoba
421	*Triticum aestivum* subsp. *aestivum*	*Lr20*	Canada	Manitoba
422	*Triticum aestivum* subsp. *aestivum*	*Lr21*	Canada	Manitoba
423	*Triticum aestivum* subsp. *aestivum*	*Lr22a*	Canada	Manitoba
424	*Triticum aestivum* subsp. *aestivum*	*Lr23*	Canada	Manitoba
425	*Triticum aestivum* subsp. *aestivum*	*Lr24*	Canada	Manitoba
426	*Triticum aestivum* subsp. *aestivum*	*Lr25*	Canada	Manitoba
427	*Triticum aestivum* subsp. *aestivum*	*Lr26*	Canada	Manitoba
428	*Triticum aestivum* subsp. *aestivum*	*Lr28*	Canada	Manitoba
429	*Triticum aestivum* subsp. *aestivum*	*Lr29*	Canada	Manitoba
430	*Triticum aestivum* subsp. *aestivum*	*Lr30*	Canada	Manitoba
431	*Triticum aestivum* subsp. *aestivum*	*Lr32*	Canada	Manitoba
432	*Triticum aestivum* subsp. *aestivum*	*Lr33*	Canada	Manitoba
433	*Triticum aestivum* subsp. *aestivum*	*Lr34*	Canada	Manitoba
434	*Triticum aestivum* subsp. *aestivum*	*Lr35*	Canada	Manitoba
435	*Triticum aestivum* subsp. *aestivum*	*Lr36*	Canada	Manitoba
436	*Triticum aestivum* subsp. *aestivum*	*Lr37*	Canada	Manitoba
437	*Triticum aestivum* subsp. *aestivum*	*Lr38*	Canada	Manitoba
438	*Triticum aestivum* subsp. *aestivum*	*Lr44*	Canada	Manitoba
439	*Triticum aestivum* subsp. *aestivum*	*Lr45*	Canada	Manitoba
440	*Triticum aestivum* subsp. *aestivum*	*Lr47*	United States	Minnesota
441	*Triticum aestivum* subsp. *aestivum*	*Lr51*	United States	California
442	*Triticum aestivum* subsp. *aestivum*	*Lr52*	Canada	Manitoba
443	*Triticum aestivum* subsp. *aestivum*	*Lr60*	Canada	Manitoba
444	*Triticum aestivum* subsp. *aestivum*	*Lr63*	Canada	Manitoba
445	*Triticum aestivum* subsp. *aestivum*	*Lr64*	Canada	Manitoba
446	*Triticum aestivum* subsp. *aestivum*	*LrB*	Canada	Manitoba

V. ABBREVIATIONS USED IN THIS VOLUME.

PLANT DISEASES, PESTS, AND PATHOGENS:

BYDV = barley yellow dwarf virus
BMV = barley mosaic virus
CCN = cereal cyst nematode, *Heterodera avenae*
FHB = Fusarium head blight
RWA = Russian wheat aphid
SBMV = soilborne mosaic virus
SLB = Septoria leaf blotch
TMV = *Triticum* mosaic virus
WDF = wheat dwarf mosaic
WSBMV = wheat soilborne mosaic virus
WSMV = wheat streak mosaic virus
WSSMV = wheat spindle streak mosaci virus
WYMV = wheat yellow mosaic virus
E. graminis* f.sp. *tritici = *Erysiphe graminis* f.sp. *tritici* = the powdery mildew fungus
F. graminearum = *Fusarium graminearum* = head scab fungus
F. nivale = *Fusarium nivale* = snow mold fungus
H. avenae = *Heterodera avenae* = cereal cyst nematode
P. graminis = *Polymyxa graminis* = wheat soilborne mosaic virus vector
P. striiformis* f.sp. *tritici = *Puccinia striiformis* f.sp. *tritici* = strip rust fungus
P. triticina = *Puccinia triticina* = *P. recondita* f.sp. *tritici* = leaf rust fungus
R. cerealis = *Rhizoctonia cerealis* = sharp eyespot
R. solani = *Rhizoctonia solani* = Rhizoctonia root rot
R. padi = *Rhonpalosiphum padi* = bird cherry-oat aphid
S. tritici = *Septorai tritici* = Septoria leaf spot fungus
S. graminearum = *Schizaphus graminearum* = greenbug
St. nodorum = *Stagonospora nodorum* = Stagonospora glume blotch
T. indica = *Tilletia indica* = Karnal bunt fungus

SCIENTIFIC NAMES AND SYNONYMS OF GRASS SPECIES (NOTE: CLASSIFICATION ACCORDING TO VAN SLAGEREN, 1994):

A. strigosa = *Avena strigosa*
Ae. cylindrica = *Aegilops cylindrica* = *Triticum cylindricum*
Ae. geniculata = *Aegilops geniculata* = *Aegilops ovata* = *Triticum ovatum*
Ae. longissima = *Aegilops longissima* = *Triticum longissimum*
Ae. markgrafii = *Aegilops markgrafii* = *Aegilops caudata* = *Triticum caudatum*
Ae. speltoides = *Aegilops speltoides* = *Triticum speltoides*
Ae. tauschii = *Aegilops tauschii* = *Aegilops squarrosa* = *Triticum tauschii*
Ae. triuncialis = *Aegilops triuncialis* = *Triticum triunciale*
Ae. umbellulata = *Aegilops umbellulata* = *Triticum umbellulatum*
Ae. peregrina = *Aegilops peregrina* = *Aegilops variabilis* = *Triticum peregrinum*
Ae. searsii = *Aegilops searsii* = *Triticum searsii*
Ae. ventricosa = *Aegilops ventricosa* = *Triticum ventricosum*
D. villosum = *Dasypyrum villosum* = *Haynaldia villosa*
S. cereale = *Secale cereale* = rye
T. aestivum* subsp. *aestivum = *Triticum aestivum* = hexaploid, bread, or common wheat
T. aestivum* subsp. *macha = *Triticum macha*
T. aestivum* subsp. *spelta = *Triticum spelta*
T. militinae = *Triticum militinae*
T. monococcum* subsp. *aegilopoides = *Triticum boeoticum*
T. timopheevii* subsp. *timopheevii = *Triticum timopheevii*
T. timopheevii* subsp. *armeniacum = *Triticum araraticum* = *T. araraticum*
T. turgidum* subsp. *dicoccoides = *Triticum dicoccoides* = wild emmer wheat

T. turgidum* subsp. *dicoccum = *Triticum dicoccum*
T. turgidum* subsp. *durum = *Triticum durum* = durum, pasta, or macaroni wheat
T. urartu = *Triticum urartu*
Th. bessarabicum = *Thinopyrum bassarabicum*
Th. elongatum = *Thinopyrum elongatum* = *Agropyron elongatum*
Th. intermedium = *Thinopyrum intermedium* = *Agropyron intermedium*

Scientific journals and publications:

Agron Abstr = Agronomy Abstracts
Ann Wheat Newslet = *Annual Wheat Newsletter*
Aus J Agric Res = *Australian Journal of Agricultural Research*
Can J Plant Sci = *Canadian Journal of Plant Science*
Cereal Chem = *Cereal Chemistry*
Cereal Res Commun = *Cereal Research Communications*
Curr Biol = *Current Biology*
Eur J Plant Path = *European Journal of Plant Pathology*
Funct Integ Genomics = *Functional Integrative Genomics*
Ind J Agric Sci = *Indian Journal of Agricultural Science*
Int J Plant Sci = *International Journal of Plant Science*
J Agric Sci Technol = *Journal of Agricultural Science and Technology*
J Cereal Sci = *Journal of Cereal Science*
J Hered = *Journal of Heredity*
J Phytopath = *Journal of Phytopathology*
J Plant Phys = *Journal of Plant Physiology*
Mol Gen Genet = *Molecular and General Genetics*
Nat Genet = *Nature Genetics*
PAG = Plant and Animal Genome (abstracts from meetings)
Phytopath = *Phytopathology*
Plant Breed = *Plant Breeding*
Plant, Cell and Envir = *Plant, Cell and Environment*
Plant Cell Rep = *Plant Cell Reporter*
Plant Dis = *Plant Disease*
Plant Physiol = *Plant Physiology*
Proc Ind Acad Sci = *Proceedings of the Indian Academy of Sciences*
Proc Natl Acad Sci USA = Proceedings of the National Academy of Sciences USA
Sci Agric Sinica = *Scientia Agricultura Sinica*
Theor Appl Genet = *Theoretical and Applied Genetics*
Wheat Inf Serv = *Wheat Information Service*

Units of measurement:

bp = base pairs
bu = bushels
cM = centimorgan
ha = hectares
kDa = kiloDaltons
m^2 = square meters
m^3 = cubic meters
μ = micron
masl = meters above sea level
me = milli-equivalents
mL = milliliters
mmt = million metric tons
mt = metric tons
Q = quintals
T = tons

MISCELLANEOUS TERMS:

Al = aluminum
AFLP = amplified fragment length polymorphism
ANOVA = analysis of variance
A-PAGE = acid polyacrylamide gel electrophoresis
APR = adult-plant resistance
AUDPC = area under the disease progress curve
BC = back cross
BW = bread wheat
CHA = chemical hybridizing agent
CMS = cytoplasmic male sterile
CPS = Canadian Prairie spring wheat
DH = doubled haploid
DON = deoxynivalenol
ELISA = enzyme-linked immunosorbent assay
EMS = ethyl methanesulfonate
EST = expressed sequence tag
FAWWON = Facultative and Winter Wheat Observation Nursery
GA = gibberellic acid
GIS = geographic-information system
GM = genetically modified
GRIN = Germplasm Resources Information Network
HPLC = high pressure liquid chromatography
HMW = high-molecular weight (glutenins)
HRSW = hard red spring wheat
HRRW = hard red winter wheat
HWSW = hard white spring wheat
HWWW = hard white winter wheat
ISSR = inter-simple sequence repeat
IT = infection type
kD = kilodalton
LMW = low molecular weight (glutenins)
MAS = marker-assisted selection
NSF = National Science Foundation
NILs = near-isogenic lines
NIR = near infrared
NSW = New South Wales, region of Australia
PAGE = polyacrylamide gel electrophoresis
PCR = polymerase chain reaction
PFGE = pulsed-field gel electrophoresis
PMCs = pollen mother cells
PNW = Pacific Northwest (a region of North America including the states of Oregon and Washington in the U.S. and the province of Vancouver in Canada)
PPO = polyphenol oxidase
QTL = quantative trait loci
RAPD = random amplified polymorphic DNA
RCB = randomized-complete block
RFLP = restriction fragment length polymorphism
RILs = recombinant inbred lines
RT-PCR = real-time polymerase-chain reaction
SAMPL = selective amplification of microsatellite polymorphic loci
SAUDPC = standardized area under the disease progress curve
SCAR = sequence-characterized amplified region
SDS-PAGE = sodium dodecyl sulphate polyacrylamide gel electrophoresis
SE-HPLE = size-exclusion high-performance liquid chromatography
SH = synthetic hexaploid

SNP = single nucleotide polymorphism
SRPN = Southern Regional Performance Nursery
SRWW = soft red winter wheat
SRSW = soft red spring wheat
STMA = sequence tagges microsatellite site
SWWW = soft white winter wheat
SSD = single-seed descent
SSR = simple-sequence repeat
STS = sequence-tagged site
TKW = 1,000-kernel weight
UESRWWN = Uniform Experimental Soft Red Winter Wheat Nursery
VIGS = virus-induced gene silencing

VI. ADDRESSES OF CONTRIBUTORS.

The email addresses of contributors denoted with a '*' are included in section VII.

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VII. VOLUME 60 MANUSCRIPT GUIDELINES.

Manuscript guidelines for the *Annual Wheat Newsletter*, volume 60. The required format for Volume 60 of the *Annual Wheat Newsletter* will be similar to previous editions edited from Kansas State University.

CONTRIBUTIONS MAY INCLUDE:

—Current activities on your projects.
—New cultivars and germ plasm released.
—Special reports of particular interest, new ideas, etc., normally not acceptable for scientific journals.
—A list of recent publications.
—News: new positions, advancements, retirements, necrology.
—Wheat stocks; lines for distribution, special equipment, computer software, breeding procedures, techniques, etc.

FORMATTING & SUBMITTING MANUSCRIPTS:

Follow the format in volume 44–59 of the *Newsletter* in coördinating and preparing your contribution, particularly for state, station, contributor names, and headings. Use Microsoft Word™ or send an RTF file that can be converted. Please include a separate jpg, gif, or equivalent file of any graphic in the contribution. Submit by E-mail to jraupp@k-state.edu.

DISTRIBUTION:

The only method of distribution of Volume 60 will be electronic PDF either by email or through download from the GrainGenes database (http://wheat.pw.usda.gov/ggpages/awn/). The volume can be found in both PDF and HTML formats. The HTML files can be read with any internet browser.

The *Annual Wheat Newsletter* will continue to be available (Vol. 37–59) through the Internet on GrainGenes, the USDA–ARS Wheat Database at http://wheat.pw.usda.gov/ggpages/awn/.