ISSN 1479-2621

Plant Genetic Resources; 1–6
doi:10.1017/S147926211700017X

High-yielding winter synthetic hexaploid wheats resistant to multiple diseases and pests

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Received 6 December 2016; Accepted 31 March 2017

Abstract
Development of winter wheat (*Triticum aestivum*) synthetics started at CIMMYT-Mexico in 2004, when winter durum wheat (*Triticum turgidum*) germplasm from Ukraine and Romania was crossed with *Aegilops tauschii* accessions from the Caspian Sea region. Chromosomes were doubled after pollination and embryo rescue, but chromosome number and cytological validation was not performed. F_2 populations were grown in Mexico and were shipped to Turkey in 2008. During 2009–2015, these populations were subjected to rigorous pedigree selection under dry, cold, disease-affected environments of the Central Anatolian Plateau. The wide segregation and partial sterility observed in 2009 gradually decreased and, by 2016, most of the F_8 single spike progenies demonstrated good fertility and agronomic performance. Since 2013, lines have been selected from synthetic populations and evaluated at multiple sites. Superior lines were characterized for resistance to leaf, stripe and stem rust, plant height, and reaction to common bunt and soil-borne pathogens. Thousand kernel weight of many lines exceeded 50 g, compared with the check varieties that barely reached 40 g. Threshability of synthetic lines varied from 0 to 95%, demonstrating genetic variation for this important domestication trait. Screening against Hessian fly, sunny pest and Russian wheat aphid identified several resistant genotypes. Both durum and *Aegilops* parents affected synthetic wheat traits. Several studies are underway to reveal the genetic diversity of synthetic lines and the basis of resistance to diseases and insects. This synthetic germplasm represents a new winter bread wheat parental pool. It is available upon request to interested breeding/research programmes.

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Keywords: biotic/abiotic stresses, genetic resources, wheat improvement

Introduction

Utilizing wheat wild relatives offers a great opportunity for expanding the genetic base to combat climatic challenges and biotic stresses. Synthetic hexaploid wheats ($2n = 6x = 42$; AABBDD), derived from crosses between durum wheat ($2n = 4x = 28$; AABB) and *Aegilops tauschii* syn. *Ae. squarossa* ($2n = 2x = 14$; DD), are widely accepted as important source of useful traits in wheat breeding (Ogbonnaya *et al.*, 2013). Recent studies have proven the value of synthetics in breeding for root traits (Becker *et al.*, 2016) and resistance to multiple insect pests and diseases (El Bouhssini *et al.*, 2013; Jighly *et al.*, 2016).

CIMMYT started developing synthetic spring wheats in the mid-1980s by crossing elite spring semi-dwarf durum wheat germplasm with a large collection of *Ae. tauschii* (Mujeeb-Kazi *et al.*, 2008). The resulting spring primary synthetics were successfully utilized by breeders (Trethowan and Mujeeb-Kazi, 2008). Development of winter synthetics started at CIMMYT in 2004, when winter durum wheat germplasm from Ukraine and Romania was crossed with winter *Ae. tauschii* accessions from the Caspian Sea basin. The chromosome number of the amphiploid plants was not controlled and, hence, 42-chromosome plants were not selected. F_3 populations were shipped to Turkey in 2008 and were subjected to rigorous pedigree selection under the dry, cold, disease-affected environment of the Central Anatolian Plateau during 2009–2015. The wide segregation for height, morphological traits (including awns and spike colour) and partial sterility observed in 2009 gradually decreased. By 2016, most of the F_8 progenies demonstrated high floret fertility and good agronomic performance. This study aimed to identify superior synthetics to be used as breeding material.

Experimental

The synthetics used in the study (249 lines) originated from 6 winter durum parents and 10 *Ae. tauschii* accessions from Azerbaijan, Iran and Russia (Table 1; online Supplementary Fig. S1). In 2016, comprehensive evaluations were conducted at multiple locations in Turkey, Azerbaijan, Kazakhstan, Lebanon, Morocco and Russia. Field evaluation for rusts was conducted in disease hotspots in Turkey under artificial inoculation: stripe rust (*Puccinia striiformis*) near Ankara, stem rust (*Puccinia graminis*) near Kastamonu (Kast.), leaf rust (*Puccinia recondite*) in Sakarya, Terter (Azerbaijan), Shortandy (Kazakhstan) and Omsk (Russia). Common bunt reaction was evaluated using artificial inoculation in Eskisehir (Turkey). Resistance to cereal cyst nematodes (*Heterodera filipjevi*) and crown rot (*Fusarium culmorum*) was evaluated under artificial inoculation in growth rooms in Eskisehir. Screening for sunny pest (*Eurygaster integriceps*) was conducted under artificial infestation in Lebanon. The synthetics were screened for Hessian fly (*Mayetiola destructor*) resistance using a population from Morocco (effective genes *H5*, *H11*, *H13*, *H14*, *H15*, *H21*, *H22*, *H23*, *H25* and *H26*). Screening for Russian wheat aphid (*Diuraphis noxia*) and barley yellow dwarf virus (BYDV) was conducted under heavy natural field pressure in Konya and Sakarya (Turkey), respectively. Growth habit was evaluated by planting material in the field in early May in Russia and Turkey. A field trial for grain yield was conducted under drought conditions in Konya (110 lines, unreplicated, 5 m^2 plots). Spike productivity traits were evaluated by harvesting five random spikes from synthetics grown at Sakarya and Kastamonu. Spike threshability (% of threshed grains) was evaluated using manual threshing. Turkish rain-fed varieties Gerek-79 and Karahan-99 were used as checks.

Discussion

The majority of the lines studied (219–87.9%) had winter growth habit (Table 1). The highest number of resistant synthetics was identified for stem rust (183–73.5%), followed by stripe rust (118–47.3%), common bunt (92–36.9%), cereal cyst nematodes (64–25.7%), crown rot (57–22.9%) and BYDV (11–4.4%). Most lines combined resistance to more than one pathogen. Both durum and *Aegilops* parents influenced resistance of synthetic wheats. For instance, durum wheat line Leuc. 84693 crossed with *Ae. tauschii* (409) produced a higher frequency of genotypes with individual or combined resistance to diseases, compared with crosses between Pandur and the same *Ae. tauschii* (409). Breeding line Ukr.-Od. 1530.94 was crossed with five accessions of *Aegilops*; the most successful was the cross with *Ae. tauschii* (1027), which resulted in the highest number of lines having high productivity combined with resistance to stripe rust, stem rusts, common bunt and soil pathogens.

Disease resistance – along with spike productivity and other traits – was used to select 85 elite synthetic lines listed in online Supplementary Table S1. The best lines (Table 2) combined high spike productivity traits with resistance to stripe and stem rust, tall stature (100–110 cm), and variable

Table 1. Pedigree of primary hexaploid synthetics and frequency of lines resistant to diseases

Cross ID	Durum parent	*Aegilops tauschii* parent			Number of lines		Number of lines resistant to the following diseases:					
		Accession #	Subspecies	Origin	Tested	Winter habit	Stripe rust[a]	Stem rust[b]	Common bunt[c]	BYDV[d]	CCN[c]	Crown rot[c]
C04GH3	Aisberg	369	*Typica*	Mazandaran, Iran	12	10	5	7	3	0	5	4
C04GH5		511	unknown	Unknown	16	8	8	11	0	1	5	5
C04GH10	Leuc. 84693	409	*Typica*	Dagestan, Russia	10	10	6	5	5	2	2	5
C04GH22	Ukr.-Od. 761.93	392	*Typica*	Shamahi, Azerbaijan	17	17	13	10	10	1	5	4
C04GH61	Ukr.-Od. 952.92	1031	*Typica*	Zanjan, Iran	21	21	12	14	0	0	4	4
C04GH68	Ukr.-Od. 1530.94	310	*Strangulata*	Gilan, Iran	7	5	2	5	2	0	4	2
C04GH71		392	*Typica*	Shamahi, Azerbaijan	12	8	1	9	8	1	1	1
C04GH74		458	unknown	Unknown	14	11	3	12	12	0	3	2
C04GH76		629	*Typica*	Mazandaran, Iran	28	24	6	21	5	1	9	6
C04GH78		1027	*Typica*	Mazandaran, Iran	47	40	36	41	28	0	17	14
C04GH79	Pandur	223	*Typica*	Gilan, Iran	39	39	19	31	14	2	5	6
C04GH81		409	*Typica*	Dagestan, Russia	26	26	7	17	5	3	4	4
Total					249	219	118	183	92	11	64	57

BYDV, barley yellow dwarf virus; CCN, cereal cyst nematode.
[a]Data from Haymana station near Ankara; stripe rust-resistant lines were identified as those with 30% or less severity. Stripe rust population was avirulent on genes *Yr5*, *Yr8*, *Yr10*, *Yr15*, *Yr24*, *Yr26* and *Yr27*.
[b]Data from Kastamonu province; stem rust-resistant lines were identified as those with 30% or less severity. Stripe rust population was avirulent on genes *Sr13*, *Sr24*, *Sr31*, *Sr36* and *Sr38*.
[c]Data from Eskisehir province; common bunt-resistant lines were identified as those with <5% infected spikes (population avirulent on genes *Bt1*, *Bt5*, *Bt8*, *Bt9*, *Bt10*, *Bt11* and *Bt13*); CCN-resistant lines from groups 1 and 2 based on number of cysts/plant and crown rot-resistant lines from groups 1 and 2 based on the visual score.
[d]Data from Sakarya province; BYDV-resistant lines were identified with score 1 based on visual evaluations.

Table 2. Superior primary hexaploid synthetics with combination of disease resistance and productive spikes, Turkey, 2016

			Ankara				Eskisehir			Sakarya				
Cross ID	2016 plot #	Konya Growth habit	Days to heading	Plant height, cm	Stripe rust, %	Kast. Stem rust, %	Common bunt, %	CCN, group	Crown rot, group	Spike length, cm	Threshability, %	Grains/ spike	Grain weight/ spike, g	1000 kernel weight, g
Gerek		W	139	93	100	72	51.3	4	3	11.5	76	40.2	1.31	32.9
Karahan		F	141	109	0	43	0.6	3	2	11.2	85	40.0	1.63	40.7
C04GH3	62	F	150	90	20	20	15.7	4	2	11.5	57	44.8	2.28	50.7
	68	F	144	105	0	20	11.9	2	2	12.6	31	52.6	2.62	49.6
C04GH5	81	W	145	115	30	5	45.3	5	2	13.6	8	40.0	1.51	38.7
	144	W	140	95	30	30	14.2	1	1	13.2	26	26.2	0.77	28.5
C04GH10	14	W	139	100	0	40	4.7	5	3	10.1	52	31.0	1.80	58.1
	162	W	143	110	0	20	8.7	2	3	12.3	10	36.4	1.33	36.2
C04GH22	51	W	146	110	5	30	1.4	2	2	11.4	47	57.6	2.42	43.1
	173	W	146	110	10	40	0.8	5	3	10.4	28	38.6	2.05	53.2
C04GH61	3	F	140	130	0	70	28.9	3	1	11.9	87	43.2	1.79	43.0
	95	F	148	115	0	20	58.5	5	3	11.1	54	36.8	1.81	49.1
C04GH68S	94	F	147	110	40	0	19.2	4	3	10.6	72	41.2	1.65	40.5
	195	S	145	110	0	5	2.0	1	2	12.1	26	38.8	1.54	39.6
C04GH71	198	W	143	105	40	5	0.0	3	3	10.9	43	40.2	1.70	42.2
	199	W	149	115	0	30	0.7	4	1	9.8	34	33.0	1.65	50.0
C04GH74	84	F	139	85	30	30	0.0	4	3	12.3	20	45.2	1.78	38.7
	207	W	139	110	40	5	0.0	1	1	12.0	14	35.4	1.16	31.7
C04GH76	211	W	145	80	40	20	0.6	4	3	11.7	12	35.6	1.22	32.2
	218	W	144	110	50	0	1.8	1	3	10.8	44	34.6	1.58	46.2
C04GH78	16	W	145	120	10	20	2.5	1	3	7.4	90	47.8	2.18	46.6
	34	W	137	97	0	5	4.5	2	4	9.6	93	25.8	1.36	53.8
	233	F	142	105	0	10	0.0	1	2	10.7	24	37.6	1.56	41.2
	235	F	136	105	0	30	1.3	3	3	11.8	4.1	35.8	1.93	55.1
	251	F	137	100	0	5	0.0	5	2	13.0	1	35.8	1.49	41.8

C04GH79	6	W	139	105	20	5	8.0	3	3	12.0	2	22.4	1.14	49.6
	58	W	141	110	0	20	5.4	3	2	13.4	83	36.8	1.89	51.1
	89	W	141	95	20	5	1.3	5	4	13.8	23	47.2	2.17	45.6
	267	W	145	110	30	10	12.7	3	3	13.7	4	34.8	1.91	54.8
	270	F	141	90	20	20	1.3	1	3	12.9	36	41.2	2.03	50.1
C04GH81	286	W	146	95	30	10	1.4	5	3	10.9	6	36.0	1.70	47.5
	289	F	141	110	30	5	14.8	4	3	11.9	2	36.2	1.94	53.3
LSD ($P < 0.5$)1	–	2	4	–	–	–	–	–	0.7	10	2.8	0.19	3.5	

Kast., Kastamonu; CCN, cereal cyst nematode; LSD, least significant difference.
LSD value was computed based on augmented design with replicated checks (Gerek and Karahan) using PROC GLM in SAS 9.4.

reactions to common bunt and soil borne pathogens. More than 100 lines were identified with resistance to leaf rust. Screening against Hessian fly and sunny pest identified seven and eight resistant synthetic lines, respectively, including three lines with resistance to both insects. Confirmation of this resistance is underway. Ten lines (plots 14–17; 22; 41; 44; 81; 114; 142) showed high resistance to Russian wheat aphid under severe pest pressure.

Synthetic lines' spikes were longer than the checks, with more spikelets per spike. The number of grains per spike varied from 20 to 58, though many exceeded the checks (40 grains/spike). Thousand kernel weight of many synthetic lines exceeded 50 g, while checks rarely reached 40 g. Threshability of synthetic lines varied from 0 to 95%, demonstrating genetic variation for this important domestication trait. The best five lines demonstrated grain yield exceeding 400 g/m^2 (online Supplementary Fig. S2).

Developing primary synthetics normally involves selection of plants with 42 chromosomes, resulting in a stable uniform genotype per cross. The primary synthetics developed in this study underwent the gradual process of chromosome stabilization. Plants with chromosome irregularities were eliminated by natural and artificial selection. Genetic material was exchanged between the chromosomes, resulting in diversity within each population for morphological and agronomic traits, which enabled targeted selection for resistance to diseases, pests and abiotic stresses. This resulted in a diverse set of valuable germplasm (120 lines) with resistance to multiple diseases and pests, while maintaining superior spike productivity. Several studies are underway to reveal the synthetics' genetic diversity and genetic basis of resistance to diseases and insects. The germplasm represents a new, unique winter bread wheat parental pool and is available to breeding/research programmes upon request.

Supplementary material

The supplementary material for this article can be found at https://doi.org/10.1017/S147926211700017X.

Acknowledgments

The International Winter Wheat Improvement Program (IWWIP) is supported by Turkey's Ministry of Food, Agriculture and Livestock and CRP WHEAT. CIMMYT thanks the Bill and Melinda Gates Foundation (BMGF) and UK Department for International Development (DFID) for providing financial support through the grant OPP1133199. Germplasm evaluation in Russia was supported by the Russian Science Foundation (Project No. 16-16-10005 signed 10.05.2016). Technical editing by Emma Quilligan is highly appreciated.

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