Table 1. Details of some annual/perennial Triticeae species hybrids and genetic stocks with durum wheat cultivars (Ad = amphiploid and BC_1 = backcross 1).

Identification number	Pedigree	Somatic chromosome number	Advance status
ANNUAL TRITICEAE			
96-2704	Laru/*Ae. variabilis*	2n = 8x = 56	Ad
96-2708	Arlin/*Ae. variabilis*	2n = 8x = 56	Ad
96-2805	Altar/*Ae. variabilis*	2n = 8x = 56	Ad
96-2821	Bia/*Ae. variabilis*	2n = 8x = 56	Ad
96-1640	Ceta/*Ae. ventricosa*	2n = 8x = 56	Ad
97-2619	Capelli/*Ae. ovata*	2n = 8x = 56	Ad
96-1250	Capelli/*Ae. triuncialis*	2n = 8x = 56	Ad
96-1254	Capelli/*Ae. speltoides*	2n = 6x = 42	Ad
PERENNIAL TRITICEAE			
96-1	*E. fibrosus*/Cocorit 71	2n = 4x = 28	Ad
96-2	*E. virginicus*/Cocorit 71	2n = 4x = 28	Ad
96-7	Altar 84/*Th. scirpeum*	2n = 4x = 28	Ad
96-65	Capelli/*Th. acutum*	2n = 5x = 35	Ad
96-66 to 69	Yavaros/*Th. acutum*	2n = 5x = 35	Ad
96-70	Cocorit 71/*Th. acutum*	2n = 5x = 35	Ad
96-72	Cocorit 71/*Th. campestre*	2n = 6x = 42	Ad
96-73	Yavaros 79/*Th. intermedium*	2n = 5x = 35	Ad
96-74	Cocorit 71/*Th. intermedium*	2n = 5x = 35	Ad
96-75	Mexicali 75/*Th. intermedium*	2n = 5x = 35	Ad
96-76	Capelli/*Th. intermedium*	2n = 5x = 35	Ad
96-77	Cocorit 71/*Th. junceiforme*	2n = 4x = 28	Ad
96-78	Cocorit 71/*Th. pulcherrimum*	2n = 5x = 35	Ad
96-79	Mexicali 75/*Th. pulcherrimum*	2n = 5x = 35	Ad
96-80	Mexicali 75/*Th. podperae*	2n = 5x = 35	Ad
96-81	Mexicali 75/*Th. trichophorum*	2n = 5x = 35	Ad
96-82	Mexicali 75/*Th. varnense*	2n = 5x = 35	Ad
96-84	Yavaros 79/*Th. varnense*	2n = 5x = 35	Ad
96-85	Capelli/*Th. varnense*	2n = 5x = 35	Ad
96-86	Cocorit 71/*Th. junceum*	2n = 4x = 28	Ad
96-106 to 108	Arlin/*Th. glaucum*	2n = 5x = 35	Ad
96-109 to 113	Croc 1/*Th. glaucum*	2n = 5x = 35	Ad
96-114 to 115	Yavaros 79/*Th. glaucum*	2n = 5x = 35	Ad
96-116 to 117	Dverd 2/*Th. glaucum*	2n = 5x = 35	Ad
96-134 to 135	Arlin/*Th. acutum*	2n = 5x = 35	Ad
96-136	Altar 84//*Th. acutum*/*Th. intermedium*	2n = 5x = 35	Ad
96-137 to 139	Croc 1//*Th. acutum*/*Th. intermedium*	2n = 5x = 35	Ad
96-140 to 142	Laru//*Th. acutum*/*Th. intermedium*	2n = 5x = 35	Ad
96-143	Arlin//*Th. acutum*/*Th. intermedium*	2n = 5x = 35	Ad
96-144 to 146	Arlin 1/*Th. junceiforme*	2n = 4x = 28	Ad
96-147	Altar 84/*Th. junceiforme*	2n = 4x = 28	Ad
96-148 to 149	Croc 1/*Th. junceiforme*	2n = 4x = 28	Ad
96-150	Altar 84/*El. pungens*	2n = 5x = 35	Ad
96-151	Yavaros 79/*Th. scirpeum*	2n = 4x = 28	Ad
96-152	Laru/*P. spicatum*	2n = 5x = 35	Ad
96-153 to 155	Dverd/ *Th. trichophorum*	2n = 5x = 35	Ad
96-156	Croc 1/*Th. trichophorum*	2n = 5x = 35	Ad
96-157	Rok/Kml//*Th. trichophorum*	2n = 5x = 35	Ad
96-158 to 161	Laru/*Th. trichophorum*	2n = 5x = 35	Ad
96-163	Altar 84/*Th. varnense*	2n = 5x = 35	Ad
96-164 to 165	Altar 84//*Th. acutum*/*Th. intermedium*	2n = 5x = 35	Ad
96-166	Laru/*Th. varnense*	2n = 5x = 35	Ad
96-169	Dverd 2/*Ps. juncea*	2n = 4x = 28	Ad + BC_1
96-170	Yavaros 79/*Th. elongatum*	2n = 3x = 21	BC_1
96-171	Croc 1/*Th. scirpeum*	2n = 4x = 28	BC_1
96-173	Yav 3/Scot//Jo69/Cra/3/Yav 79/4/*Th. elongatum*	2n = 3x = 21	BC_1
96-174	Altar 84/*Th. intermedium*	2n = 5x = 35	BC_1

desired combination for applied purposes via addition/substitution lines and then introgressing the required trait by cytogenetic manipulation. The use of the *ph* genetic stock of Capelli is in its infancy in our program and is projected as a fast source to enforce alien transfers when it is the backcross parent for the F_1 hybrid, yielding *Phph* heterozygote progeny. Selecting the *ph* recessive and achieving the alien transfer are anticipated and being studied.

Of the annual Triticeae species, those hybridized with durum cultivars are *Ae. peregrina*, *Ae. ventricosa*, *Ae. geniculata*, *Ae. triuncialis*, and *Ae. speltoides*. Amphiploids were produced from all the above hybrid combinations (see Table 1).

Intergeneric F_1 hybrids of some bread wheat cultivars with annual and perennial Triticeae species: germ plasm status and utilization in wheat breeding.

A. Mujeeb-Kazi, V. Rosas, and R. Delgado.

In a bread wheat-based, intergeneric, hybridization program with a focus on applied agricultural objectives, outputs are necessary that address several biotic and abiotic stresses. This is indeed a tall order, because the initial hybrid production in itself is so complex but a necessary starting point. Next in order come the crucial steps of transfering the desirable stress genes and dealing with the genetic distance between wheat and the species involved. We have been involved in F_1-hybrid production of wheat with annual and perennial Triticeae for over two decades. The annual species/wheat cultivar hybrids were relatively easy to produce, and their amphiploid products also obtained easily (Table 2). The complexity resided in the combinations of wheat with species that were not in the primary gene pool,

which is not the case for hybrid production alone, but is a major factor when alien genes have to be transferred from the distant species into wheat.

Our hybrid production protocols have been simplified enough that quite complex cross-combinations have been realized. The hybrids possessing a perennial habit are maintained with cytological documentation and physical cloning twice each year. Clones are further field evaluated for resistance/tolerance to stresses and production of amphiploids, wherever possible, is a routine biannual process. The complete list of these intergeneric combinations and some supporting details are provided in Table 2. The diversity is significant in that it allows researchers to produce desired BC_1 progeny with virtually any bread wheat cultivar that may be site/country specific and to use our genetic-stock base more efficiently and avoid remaking the F_1 hybrids; which is a difficult, if not cumbersome process. This information is shared to facilitate the broad usage of our germ plasm. To some extent, we have been producing special BC_1 progeny for international colleagues who use their cultivars as the parents and our perennial F_1 hybrids.

The most important stress traits linked to a species and combination are identified. The data that we have accumulated allow us to use a combination for multiple objectives with the same manipulation strategy. That strategy is pursued intensively and revolves around the alien species *Th. bessarabicum*. Combining this species with wheat has potential for salt tolerance and *F. graminearum* resistance, two major constraints on wheat production. *Thinopyrum elongatum* is another such preferred species. Tolerance/resistance in these species resides on more than one chromosome. A living collection allows us to swiftly go to the

Table 2. Details of some annual/perennial Triticeae species hybrids and genetic stocks with bread wheat cultivars (Ad = amphiploid and BC_1 = backcross 1)..

Identification number	Pedigree	Somatic chromosome number	Advance status
Annual Triticeae			
96-2684	Chinese Spring (CS)/*Ae. variabilis*	2n = 10x = 70	Ad*
96-2705	Alondra/Pavon//*Ae. variabilis*	2n = 10x = 70	Ad
96-2832	Faisalabad/*Ae. variabilis*	2n = 10x = 70	Ad
96-2840	Jauhar/*Ae. variabilis*	2n = 10x = 70	Ad
96-2811	Asakazekomugi/*Ae. variabilis*	2n = 10x = 70	Ad
96-2836	Fukohokomugi/*Ae. variabilis*	2n = 10x = 70	Ad
96-2850	Lu 26/*Ae. variabilis*	2n = 10x = 70	Ad
96-2861	Pak 81/*Ae. variabilis*	2n = 10x = 70	Ad
96-2870	Punjab/*Ae. variabilis*	2n = 10x = 70	Ad
Perennial Triticeae			
96-3	CS/*Th. bessarabicum*	2n = 4x = 28	Ad + BC_1
96-5	Genaro/*Th. trichophorum*	2n = 6x = 42	
96-6	Fremont (FMN)/*E. angustus*	2n = 9x = 63	
96-8	CS/*Th. acutum*	2n = 6x = 42	
96-9	FMN/*Th. acutum*	2n = 6x = 42	
96-10	Nacozari/*Th. acutum*	2n = 6x = 42	BC_1
96-11 to 12	CS/*Th. caespitosum*	2n = 5x = 35	
96-13	CS/*Th. curvifolium*	2n = 5x = 35	BC_1
96-14 to 15	CS/*Th. campestre*	2n = 7x = 49	
96-16	CS/*E. cinereus*	2n =5x = 35	
96-17 to 18	CS/*L. racemosus*	2n = 5x = 35	
96-19 to 20	CS/*Th. intermedium*	2n = 6x = 42	BC_1
96-21	Nacozari/*Th. intermedium*	2n = 6x = 42	
96-22	Glennson/*Th. intermedium*	2n = 6x = 42	
96-23 to 24	FMN/*Th. junceiforme*	2n = 5x = 35	
96-25	Fielder (FDR)/*Th. junceiforme*	2n = 5x = 35	BC_1
96-26	Pavon/*Th. junceiforme*	2n = 5x = 35	
96-27	CS/*Th. junceum*	2n = 6x = 42	
96-28	CS/*Th. pulcherrimum*	2n = 6x = 42	BC_1
96-29	CS/*Th. podperae*	2n = 6x = 42	Ad + BC_1
96-30 to 57	CS//*Th. repens*/A. *desertorum*	2n = 8x = 56	BC_1
96-59	CS/*Th. scirpeum*	2n = 5x = 35	Ad + BC_1
96-60	CS/*Th. trichophorum*	2n = 6x = 42	Ad
96-61	CS/*E. triticoides*	2n = 5x = 35	
96-62	CS/*Th. varnense*	2n = 6x = 42	
96-63	Nacozari/*Th. varnense*	2n = 6x = 42	BC_1
96-64	Pavon/*Th. varnense*	2n = 6x = 42	
96-87	Tobari 66/*Th. junceum*	2n = 6x = 42	
96-92	*E. agropyroides*/Nyubay	2n = 6x = 42	
96-93	*E. agropyroides*/Zaragoza75	2n = 6x = 42	
96-97	Pavon/*Th. elongatum*	2n = 8x = 56	
96-98	Zaragoza 75/*Th. elongatum*	2n = 8x = 56	
96-99	CS/*Th. elongatum*	2n = 8x = 56	
96-100	CS/Ciano(enano)//*Th. elongatum*	2n = 8x = 56	
96-101	Nyubay/*Th. elongatum*	2n = 8x = 56	
96-103	CS/*Th. repens*	2n = 6x = 42	
96-104	Mrl/Buc//*Th. campestre*	2n = 7x = 49	
96-105	Yaco/*Th. campestre*	2n = 7x = 49	
96-118	Buc/*Th. glaucum*	2n = 6x = 42	
96-119	*E. fibrosus*/GHK	2n = 5x = 35	Ad + BC_1
96-120 to 122	Buc/Bjy//*Th. acutum*/*Th. intermedium*	2n = 6x = 42	
96-123 to 124	Dove/Buc//*Th. acutum*/*Th. intermedium*	2n = 6x = 42	
96-125 to 127	Yaco//*Th. acutum*/*Th. intermedium*	2n = 6x = 42	
96-128	Buc/Pvn//*Th. junceum*	2n = 6x = 42	
96-129 to 130	Dove/Buc//*Th. trichophorum*	2n = 6x = 42	
96-131 to 132	Yaco/*Th. trichophorum*	2n = 6x = 42	
96-133	Buc/*Th. varnense*	2n = 6x = 42	
96-168	CS/*A. cristatum*	2n = 4x = 28	
96-172	*E. fibrosus*/Pavon	2n = 2x = 14	

desired F_1 hybrid that has the *Ph* gene and produce BC_1 *Phph* heterozygote derivatives by crossing the *Ph* F_1 with the Chinese Spring *phph* genetic stock. A PCR-based protocol then allows us to identify the progeny that possesses the *ph* recessive locus, and this becomes a source for multiple homoeologous transfers.

Agrotricum (2n = 8x = 56): potential for barley yellow dwarf virus resistance and its cytogenetics.

M. Henry, A. Cortes, V. Rosas, R. Delgado, and A. Mujeeb-Kazi.

Testing of various germ plasms for resistance to BYDV has been done over the past few years in CIMMYT both in the field and in controlled greenhouse conditions. The germ plasm was comprised of elite cultivars, Triticeae species of the three gene pools, amphiploids from some intergeneric hybrids, BC_1 self-fertile derivatives of the intergeneric hybrids, and some partial amphiploids. One such partial amphiploid is *Agrotricum* (2n = 8x = 56), which was identified in Canada as being resistant to BYDV. In Mexico, we have obtained similar data to support the resistance and, after studying the cytogenetics of this genetic stock, have initiated a program to produce addition lines, identify addition lines that show resistance, and transfer the resistance genes to our spring wheat cultivars. We report here the cytogenetic progress and BYDV resistance data on the original 56-chromosome stock and its initial backcross derivatives. Appropriate controls were included in the study.

Cytology of *Agrotricum*. Somatic cytology of *Agrotricum* indicated some aneuploidy in the seedlings analyzed where 56-chromosome normal derivatives were present, but plants with 54, 55, and 57 chromosomes and some with telocentrics also were present. Plants with 56 chromosomes were Giemsa C-banded, and the stable lines were analyzed further by FISH. The 56-chromosome, partial amphiploid possessed 14 *Thinopyrum* chromosomes, 40 normal wheat chromosomes, and a pair of wheat chromosomes with a translocation between chromosome 3D of wheat and *Thinopyrum*. The exchange is at the terminal end of 3DL. Such plants were analyzed at meiosis, and all were normal with 29 bivalents at metaphase I and a normal anaphase separation. These plants were seed increased and tested for BYDV resistance. They were all resistant.

The euploid stock then was backcrossed to the susceptible wheat cultivars Prinia and Bagula, which resulted in BC_1 progeny with 2n = 7x = 49 chromosomes. These progenies also tested positive for BYDV resistance. The BC_1 progeny was advanced further by backcrossing to each of the two parental wheat cultivars and also by selfing to eventually identify plants with 43 chromosomes (21 bivalents +1 univalent). From these, 44 chromosome (22 bivalent) derivatives were obtained by selfing of the 43-chromosome plants or by producing 22-chromosome haploids after crosses with maize and then doubling these haploids.

The 44-chromosome progeny was C-banded, and five disomic additions were identified. One of these addition derivatives has a very low BYDV concentration when tested by ELISA, and further work is in progress to introgress the resistance into wheat. Backcross derivatives with 42 chromosomes (possessing the translocated pair) did not possess BYDV resistance.

BYDV screening–virus isolates and BYDV inoculation. The BYDV-PAV isolate used was collected in Mexico and maintained in CIMMYT's greenhouse through transmission by aphids. Inoculation was by infesting 7-day-old seedlings from the *Agrotricum* germ plasm, parental wheat cultivars, and a resistant check (TC14) with 10 viruliferous aphids (*Rhopalosiphum padi*) that had acquired BYDV by feeding on infected plants for 48 hours. The seedlings were isolated from each other by transparent plastic tubes. After a 2- to 5-day period, aphids were killed with Metasystox (Bayer). In each entry, two plants were kept free of aphids to serve as the uninoculated controls.

Enzyme-linked immunosorbent assay (ELISA). The flag-1 leaf was collected at different dates after inoculation for the evaluation of the virus titer by ELISA. Double antibody sandwich ELISA (DAS ELISA) was used with a few modifications. Polystyrene microtiter plates (NUNC) were incubated at 37°C for 3 hours with coating polyclonal antibodies directed against the U.S. isolates provided by K. Perry (Purdue University, W. Lafayette, IN). Plant sap (1:10, in 0.1M phosphate buffer pH 7.0) was incubated for 3 hours at 37°C. Alkaline phosphatase-labeled, polyclonal, anti-PAV antibodies (1:1000) were incubated overnight at 4°C. P-nitrophenyl phosphate substrate (Sigma) was added at a concentration of 1 mg/ml, and the mixture was incubated for 1 to 2 hours at room temperature. Optical density (OD) was measured at 405 nm. A plant was considered infected when the OD was higher than twice that obtained for the