Theor Appl Genet (1998) 97: 550–556

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Comparison of bread wheat lines selected by doubled haploid, single-seed descent and pedigree selection methods

Received: 23 December 1997 / Accepted: 12 March 1998

Abstract Yield performance of each group of ten spring bread wheat lines selected by doubled haploid (DH), single-seed descent (SSD) and pedigree selection (PS) methods from three F_1 crosses was compared with the aim of evaluating the DH method in breeding programs. Populations of 65–97 DH lines and 110 SSD lines per cross were used for selection. PS lines were developed by repeated selections from 1500 F_2 plants. Yield evaluation was performed at the F_6 generation of SSD and PS lines along with DH lines in a 2-year field experiment. It took only 2 years from the planting of wheat materials for DH production to the planting of selected DH lines for yield evaluation. There was no significant difference in grain yield between DH lines and PS lines selected from an F_1 cross whose parental varieties were closely related in their pedigrees. In two crosses with low coefficients of parentage and a large variation in their progenies, grain yield of selected DH lines was significantly lower than those of selected SSD and PS lines. These results confirm that the DH method can save time in obtaining recombinant inbred lines ready for yield evaluation. However, a larger DH population is required to achieve the same level of genetic advance with the PS method in crosses containing greater genetic variation.

Key words Bread wheat · Doubled haploids · Pedigree selection · Single-seed descent

Introduction

In self-pollinating crops, such as bread wheat (*Triticum aestivum* L.), breeding programs must include a three-step process for developing germplasm: (1) recombination of genes for enlarging variation, (2) identification and selection of recombinant genotypes according to their agronomic types and (3) fixation of genes in homozygous genotypes. Pedigree selection (PS) is the conventional method of accumulating genetic recombination in each generation. The heterozygosity in early generations makes the efficient identification and selection of recombinant genotypes more difficult. Repeated pedigree selection can increase homozygosity, but many generation cycles are required to reach homozygosity in loci associated with agronomic traits. The single-seed descent (SSD) method can be used to obtain homozygous inbreds by accelerating generation cycles, but its application is dependent on growth habit of the plant materials used.

The artificial production of haploid plants followed by chromosome doubling is the quickest method for obtaining homozygous recombinant genotypes from heterozygous parental genotypes in a single generation (Nei 1963). The doubled haploid (DH) method has the obvious advantage in that yield evaluation can begin earlier and varietal development is speeded up. This method also has the great advantage of increasing the efficiency of selection because DH lines do not express dominance variation and segregation within lines (Snape 1989).

During the last two decades, a technique for producing haploid plants has been developed in bread wheat using wide crosses followed by chromosome elimination (see Inagaki 1997 for a review). A high frequency of haploid production was first reported in intergeneric crosses with wild barley *Hordeum bulbosum* L. (Barclay 1975), but many wheat genotypes showed cross-incompatibility with *H. bulbosum* (Snape et al. 1979; Falk and

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Communicated by G. Wenzel

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Kasha 1981). The application of plant growth regulators, such as 2,4-dichlorophenoxyacetic acid (2,4-D), prior to pollination was found to be effective in increasing the frequency of haploid embryo formation but did not completely overcome cross-incompatibility (Inagaki 1986). Inter-subfamilial crossings with maize (*Zea mays* L.) showed successful hybridization followed by the rapid elimination of maize chromosomes, just as in the *H. bulbosum* crosses (Laurie and Bennett 1986). Maize crosses were used for wheat genotypes that were cross-incompatible with *H. bulbosum* and produced haploid plants of bread wheat (Laurie and Bennett 1988). 2,4-D treatment in combination with maize crosses drastically enhanced the development of haploid wheat embryos (Suenaga and Nakajima 1989). Efficient haploid production through crosses with maize and subsequent 2,4-D treatment was confirmed in diverse bread wheat genotypes (Inagaki and Tahir 1990). Since then, a technique for the production of DH lines has been improved to a level allowing its utilization in breeding programs.

This paper reports the results of a field evaluation of spring bread wheat lines selected by the doubled haploid (DH), single-seed descent (SSD) and pedigree selection (PS) methods from three F_1 crosses.

Materials and methods

Production of plant materials

Three spring bread wheat F_1 crosses, 'Seri 82/Attila' (S/A), 'Bobwhite/Parula//Buckbuck/3/Luan' (B/L) and 'Kauz/Weaver' (K/W), were used for production of the DH, SSD and PS lines. The parental varieties were semi-dwarf genotypes that had been developed at CIMMYT for a target environment defined as Mega-environment 1. This is a temperate, low-rainfall and irrigated environment represented in the developing world by such locations as the Yaqui Valley in Mexico, the Indus Valley in Pakistan, the Gangetic Valley in India and the Nile Valley in Egypt (Rajaram et al. 1995). Table 1 shows the semi-dwarfing genes and coefficients of parentage of the six parental varieties used in this study. The type of semi-dwarfing gene present in two of the parental varieties is unknown. The coefficients of parentage were obtained from the International Wheat Information System (Fox et al. 1997).

Table 2 gives the production and selection process of DH, SSD and PS lines from each of the three bread wheat F_1 crosses. DH plants were produced from crosses of bread wheat F_1 plants with maize according to the method of Inagaki and Mujeeb-Kazi (1995). After a one-generation cycle for seed multiplication, there were 87, 65 and 97 DH lines from S/A, B/L and K/W crosses, respectively. Harvested seeds of the F_1 plants were used to develop the SSD and PS lines. Using 110 F_2 plants that had been randomly chosen for each cross, we self-pollinated SSD lines to reach the F_4 generation by growing 1 plant from each plant without selection. From approximately 1500 F_2 plants grown in the field for each cross, 150 plants were selected to produce F_3 families. The pedigree families (3 lines/family) were further selected at the F_3 and F_4 generations. Final visual selection for yield performance was conducted on populations of DH, SSD and PS lines grown in the field at CIMMYT, El Batan, Mexico State, Mexico, to produce 10 lines for each of the three breeding methods. Data on plant height were collected for all lines of the three populations to estimate the variation of the population as well as for 20 individual plants per line to estimate the uniformity of the line.

Field experiments

In the winter-spring seasons of 1995–1996 and 1996–1997, 32 genotypes including 10 DH, 10 SSD and 10 PS lines plus two parental varieties for each cross were grown in a three-replicate randomized block experiment at a CIMMYT station, Ciudad Obregon, Sonora State, Mexico. Seeding was done by using a plot drill on flat beds 80 cm wide with three rows 20 cm apart in plots consisting of six rows, 5 m long. The seeding rate was 72 g of seed per plot. Crop management included fertilizer application, optimal irrigation and chemical control of diseases, pests and weeds.

All plot materials were harvested after physiological maturity. Yield component traits were measured using a subsample of 50 wheat culms according to the method of Sayre et al. (1997). Data were collected on nine agronomic traits: grain yield, heading time, plant height, spike length, number of spikes, biomass, harvest index, 1000-grain weight and test weight. Analysis of variance was carried out on all traits collected with three replications for 2 years. Mean comparisons were performed using the least significant difference (LSD) calculated from the genotype × year interaction mean square.

Results

Populations of DH, SSD and PS lines

Parental varieties 'Seri 82' and 'Attila' were genealogically related, with a high coefficient of parentage of 0.383. On the other hand, coefficients as low as 0.133 and 0.156 were found between 'Bobwhite/Parula//

Table 1 Semi-dwarfing genes and coefficients of parentage of six parental varieties of spring bread wheat used in this study

Wheat variety		Semi-dwarfing gene[a]	Coefficient of parentage[b]					
			(S)	(A)	(B)	(L)	(K)	(W)
Seri 82	(S)	*Rht1*	1.000					
Attila	(A)	*Rht1*	0.383	1.000				
Bobwhite/Parula //Buckbuck	(B)	?	0.328	0.178	1.000			
Luan	(L)	?	0.208	0.222	0.133	1.000		
Kauz	(K)	*Rht1*	0.378	0.282	0.185	0.230	1.000	
Weaver	(W)	*Rht2*	0.170	0.148	0.213	0.112	0.156	1.000

[a] Sources: Singh et al. (1989); Sayre et al. (1997)

[b] Source: Fox et al. (1997)

Table 2 Production of spring bread wheat lines from three F_1 crosses by doubled haploid (DH), single-seed descent (SSD) and pedigree selection (PS) methods

Year	Season (location)	Breeding method		
		DH	SSD	PS
1993	Summer (El Batan)	Wheat F_1 plants maize crossing		
1993	Winter/spring (Cd. Obregon)	Doubled haploid grains	110 F_2 plants	1500 F_2 plants *Selection*
1994	Summer (El Batan)	Seed multiplication	110 F_3 plants	150 F_3 families *Selection*
1994	Winter/spring (Cd. Obregon)		110 F_4 plants	60 F_4 families *Selection*
1995	Summer (El Batan)	65–97 lines *Selection*	110 F_5 lines *Selection*	20 F_5 families *Selection*
1995	Winter/spring (Cd. Obregon)	10 lines *Yield evaluation*	10 lines *Yield evaluation*	10 lines *Yield evaluation*
1996	Winter/spring (Cd. Obregon)	10 lines *Yield evaluation*	10 lines *Yield evaluation*	10 lines *Yield evaluation*

Buckbuck' and 'Luan', and between 'Kauz' and 'Weaver', respectively. 'Weaver' has a semi-dwarfing gene, *Rht2*, whose locus is different from that of the semi-dwarfing gene, *Rht1*, in 'Kauz'. Plant height of the DH, SSD and PS populations produced from three crosses is shown in Fig. 1. Across the three crosses, plant height variation of the DH population was similar to that of the SSD population. The PS population exhibited narrow variation due to repeated selection in previous generations. In particular, cross K/W, involving two different semi-dwarfing genes, expressed distinct segregation in plant height: dwarf, semi-dwarf and tall types. Coefficients of variance in plant height for the 10 lines selected by each of three breeding methods are given in Fig. 2. Plant height of DH lines showed relatively low coefficients of variance and was uniform within lines; however, unlike the DH lines, some SSD and PS lines had large variation within lines.

Grain yields of the best 10 lines selected by each of the three breeding methods

Means and ranges for nine agronomic traits of the 10 lines selected by each of the three breeding methods are shown in Table 3. Figure 3 gives individual grain yields of the selected 10 lines. In cross S/A, small differences were found in agronomic traits among lines selected by the three breeding methods. Grain yields of the 10 DH lines were similar to those of the 10 PS lines. Mean grain yield of the SSD lines was slightly higher than those of the DH and PS lines. In cross B/L, differences in agronomic traits were distinct among lines as well as among breeding methods. Mean grain yield of PS lines was significantly higher than that of SSD lines. DH lines expressed the lowest mean grain yield. The highest grain yield among DH lines was comparable to the mean grain yield of the 10 PS lines. Grain yield of SSD lines showed large variation that ranged between the highest value of PS lines and the lowest value of DH lines. In most agronomic traits of cross K/W, particularly heading time, plant height and number of spikes, large differences were found among lines as well as among breeding methods. Mean grain yield of DH lines was significantly lower than those of SSD and PS lines. However, grain yields of the top 6 DH lines were similar or higher than mean grain yield of the PS lines.

Discussion

Efficiency of DH production through maize crosses in bread wheat

In this study, DH populations were directly produced from different types of bread wheat F1 crosses and selected once before yield evaluation. The process took 2 years from the initial planting of the wheat materials for DH production to the final planting of selected DH lines for yield evaluation. The time-saving advantage of the DH method is great for wheat materials of the winter growth habit that require a relatively long growth period but small for spring wheat materials in which the breeding scheme can be accelerated to two generation cycles per year.

At CIMMYT, it is expected that the frequency of spring bread wheat DH production through maize

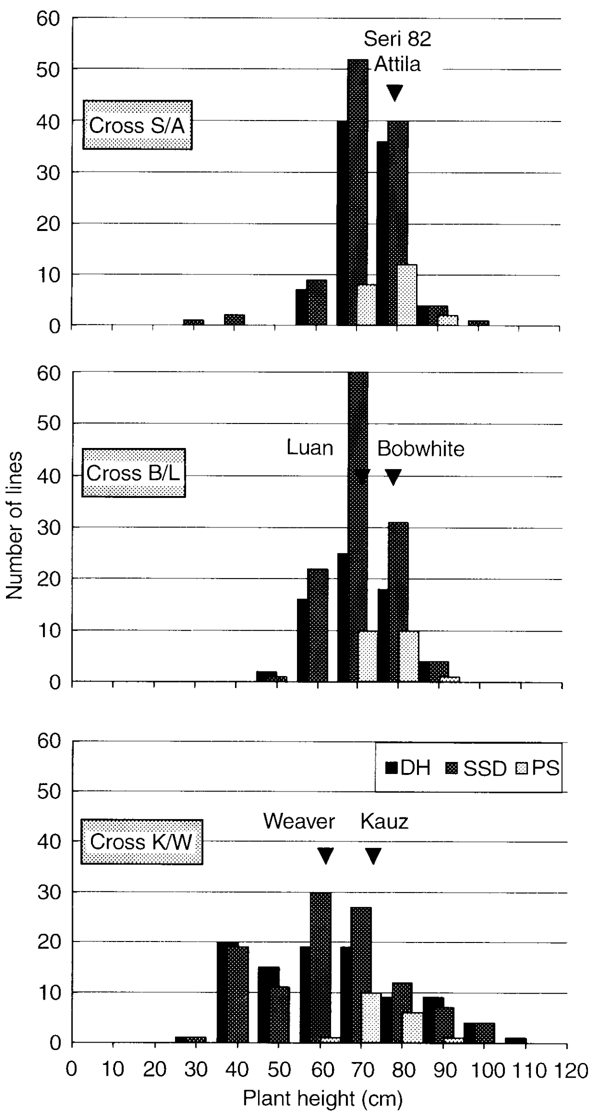

Fig. 1 Plant height of bread wheat populations produced by the doubled haploid (DH), single-seed descent (SSD) and pedigree selection (PS) methods from three F_1 crosses

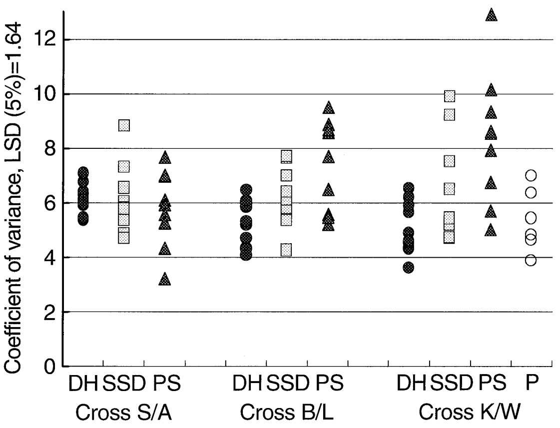

Fig. 2 Coefficients of variance (20 plants per line) for plant height of each group of ten bread wheat lines selected by the doubled haploid (DH), Single-seed descent (SSD) and pedigree selection (PS) methods from three F_1 crosses

crosses is approximately 1.5 DH lines per wheat spike (Inagaki 1997). Efficient production techniques have been recently developed using hot-water emasculation, pollen storage and detached-tiller culture which can give a considerable savings in terms of labor and space required for wheat wide crosses (Inagaki et al. 1997). The frequency of DH production in bread wheat has been significantly increased. Therefore, to obtain a completely random sample from gametes of starting wheat materials is more crucial for the purposes of genetic analysis and breeding than to improve the frequency of DH production. A previous report indicated that there is no significant distortion of segregation ratios of some genetic markers in a DH population produced from an F_1 cross through maize crosses (Suenaga and Nakajima 1993b). Negligible variation in agronomic traits was also found in DH populations derived from bread wheat varieties (Laurie and Snape 1990; Suenaga and Nakajima 1993 a). In spring barley crosses, the means of the three sample populations developed by the DH, SSD and PS methods did not differ significantly (Park et al. 1976).

The variation in plant height of the DH populations produced from the three F_1 crosses in this study was similar to that of SSD populations produced from the same crosses, suggesting no serious distortion of genetic segregation in plant height. The DH plants were also uniform in plant height within lines. This uniformity became significant when DH lines were produced from crosses with low coefficients of parentage and large variation in their progenies. The uniformity of breeding lines has the effect of increasing selection efficiency. These results suggest that DH populations produced from F_1 crosses can be used for genetic analyses and breeding selection.

Evaluation of the DH method

In comparison with the PS method, the DH method has some disadvantages (Snape 1989). A larger DH population is required to have a reasonable chance of selecting desirable recombinant genotypes as the number of genes that control agronomic traits becomes larger. The DH method with homozygosity has no chance of generating genetic recombination between parental genotypes after DH production, whereas the PS method can accumulate and select genetic advances occurring in subsequent generations. The results obtained in this study indicate that genetic advances in grain yield of selected DH lines were comparable to that of PS lines when the parents involved in the F_1 cross were genealogically related. On the contrary, in F_1 crosses of parents that were genealogically and

Table 3 Means (ranges) for nine agronomic traits of each group of 10 bread wheat lines selected by doubled haploid (DH), single-seed descent (SSD) and pedigree selection (PS) methods from three F_1 crosses

Cross	Breeding method	Grain yield (t/ha)	Heading time (days)	Plant height (cm)	Spike length (cm)	Number of spikes (/m^2)	Biomass (t/ha)	Harvest index (%)	1000-grain weight (g)	Test weight (g/l)
S/A	DH	6.95a[a]	92.9a	99.4a	9.18a	427a	15.84a	44.4a	37.6a	80.7a
		(7.37–6.70)	(97.7–88.5)	(105.6–94.7)	(9.55–8.86)	(494–328)	(17.01–14.40)	(49.5–41.8)	(41.8–33.9)	(82.6–78.4)
	SSD	7.13b	88.2c	96.8b	9.28a	418a	15.69a	45.7a	38.4b	81.1a
		(7.50–6.63)	(94.3–82.0)	(101.0–93.5)	(9.88–8.29)	(492–367)	(16.58–14.46)	(48.7–43.5)	(39.5–36.2)	(82.5–79.3)
	PS	6.99a	91.0b	97.2b	9.24a	423a	15.72a	44.7a	38.4b	80.9a
		(7.46–6.69)	(95.0–87.2)	(103.2–93.5)	(9.61–8.77)	(488–391)	(16.69–14.82)	(47.2–42.5)	(40.7–35.9)	(81.7–79.6)
	Seri 82	6.61	88.0	96.4	9.78	370	14.21	46.8	39.5	78.7
	Attila	6.33	91.5	96.9	8.70	450	14.69	43.6	43.0	80.8
	c.v. (%)	4.27	1.23	2.95	4.94	10.8	7.64	6.20	3.85	0.39
B/L	DH	6.56a	88.6a	93.0a	9.21a	395a	14.95a	44.1a	37.6a	79.0a
		(6.93–6.12)	(94.2–79.8)	(101.4–86.8)	(9.47–8.89)	(430–372)	(15.72–14.05)	(45.7–41.9)	(42.3–32.8)	(80.4–76.6)
	SSD	**6.73**b	88.3a	95.2b	9.09a	380a	15.39ab	44.0a	39.5b	79.1ab
		(7.48–6.09)	(92.2–79.7)	(101.8–88.3)	(10.04–7.64)	(423–352)	(16.03–14.52)	(48.1–40.2)	(45.3–34.8)	(81.3–75.6)
	PS	6.99c	85.8b	99.2c	9.27a	381a	15.73b	44.8a	40.3b	79.4b
		(7.37–6.73)	(90.7–80.5)	(102.9–92.9)	(10.04–8.56)	(425–347)	(16.82–15.22)	(46.6–42.7)	(43.3–35.5)	(81.1–78.0)
	Bobwhite	6.13	84.5	99.3	11.59	342	14.12	45.1	38.9	80.9
	Luan	6.69	92.2	87.3	9.00	392	14.20	47.5	31.5	79.0
	c.v. (%)	3.45	1.18	2.98	5.96	12.7	8.57	7.36	3.92	0.53
K/W	DH	6.60a	95.9a	94.6a	9.00a	465a	16.22a	41.0a	37.3a	80.8a
		(7.69–5.53)	(103.5–85.0)	(132.1–78.0)	(10.55–7.95)	(552–371)	(17.74–14.69)	(46.6–32.7)	(44.5–28.9)	(82.5–75.7)
	SSD	**6.85**b	91.2b	93.1a	9.04a	456a	15.58b	44.1b	38.6b	80.4a
		(7.22–6.02)	(102.5–83.0)	(115.3–83.2)	(9.78–8.21)	(577–380)	(16.27–14.18)	(48.0–42.5)	(44.0–32.5)	(82.1–77.3)
	PS	6.82b	81.5c	101.3b	8.40b	420b	15.10b	45.3b	40.6c	81.6b
		(7.33–5.61)	(88.3–77.0)	(118.0–93.4)	(9.19–6.75)	(512–353)	(16.25–14.19)	(47.9–39.0)	(45.5–37.0)	(83.1–80.3)
	Kauz	7.70	85.8	90.2	8.87	431	16.02	48.2	36.9	81.6
	Weaver	6.99	90.7	79.7	8.65	539	15.78	44.0	35.1	78.7
	c.v. (%)	5.97	2.17	3.15	5.92	11.0	9.83	6.08	3.70	0.70

[a] Means followed by the same letter within each cross in the same column are not significantly different at the 5% probability level

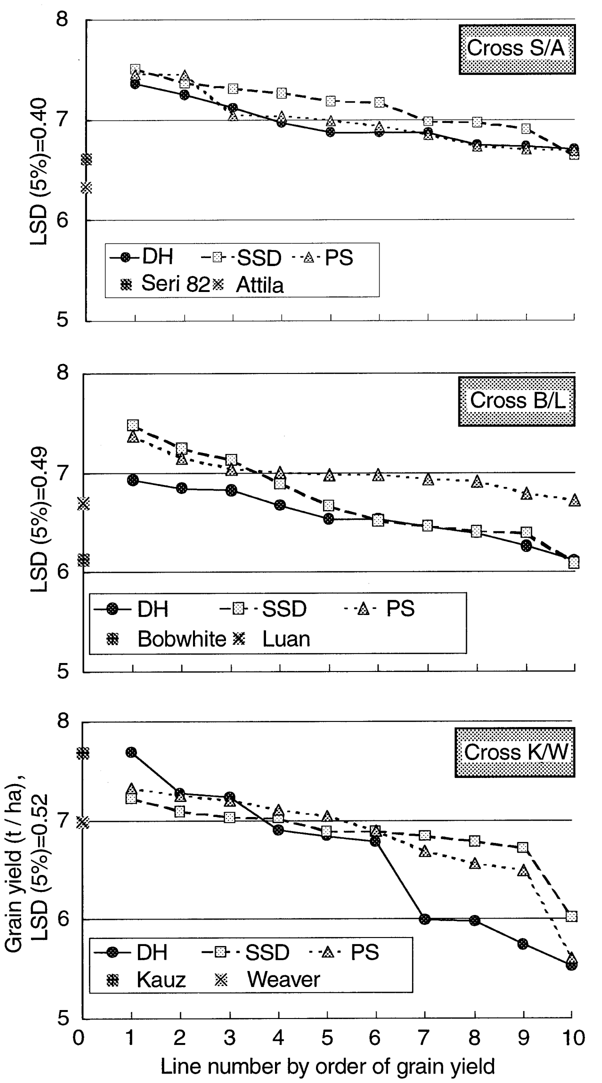

Fig. 3 Grain yields of each group of 10 bread wheat lines selected by the doubled haploid (DH), single-seed descent (SSD) and pedigree selection (PS) methods in three F_1 crosses

morphologically different from each other, genetic advance of the DH lines was significantly lower than that of the PS lines. Thus, effectiveness of the DH method greatly depends on the genetic variation contained in breeding materials. As one of the conditions favoring the DH method over conventional breeding methods, Yonezawa et al. (1987) pointed out that a relatively small number of genes is involved in the breeding objective concerned.

The size of DH population produced in cross S/A may be enough to reach the genetic advance in grain yield obtained by the PS method. However, grain yield of the best DH line reached the mean grain yield of 10 PS lines in cross B/L. The large genetic variation found in this cross requires a population at least 10 times the size of the DH population produced in this study to achieve the same level of genetic advance in the PS method. It can be roughly estimated that the size of the DH population derived from F_1 crosses should be 100–700 lines per cross, depending on the genetic variation involved in breeding materials. A scheme for reducing the number of DH lines is to give a chance of selection in the process of developing haploid plants from heterozygous materials. Haploid plants can express desirable recessive alleles as well as dominant alleles, whereas diploid plants cannot due to their heterozygosity. Another option is to delay DH production until the F_2 or F_3 generation and to select desirable recombinants before DH production, although this decreases the time-saving advantage. These schemes can increase the frequency of homozygous plants with desirable traits, and the size of the DH population to be produced for the given genetic advance becomes smaller.

In conclusion, the DH method has the advantages of reducing the time required to obtain recombinant inbred genotypes ready for yield evaluation and of increasing selection efficiency; however, a larger DH population is required to increase the probability of selecting desirable genotypes in crosses containing larger genetic variation.

Acknowledgements The authors thank Ms. A. McNab, Wheat Program editor, CIMMYT, for her editorial review of the manuscript. This contribution results from a collaborative research project with the Japan International Research Center for Agricultural Sciences (JIRCAS), Tsukuba, Ibaraki, Japan.

References

Barclay IR (1975) High frequencies of haploid production in wheat (*Triticum aestivum* L.) by chromosome elimination. Nature 256:410–411

Falk DE, Kasha KJ (1981) Comparison of the crossability of rye (*Secale cereale*) and *Hordeum bulbosum* onto wheat (*Triticum aestivum*). Can J Genet Cytol 23:81–88

Fox PN, Magaña RI, Lopez C, Sanchez H, Herrera R, Vicarte V, White JW, Skovmand B, Mackay MC (1997) International Wheat Information System (IWIS), version 2. CIMMYT, Mexico, D.F. (on compact disk)

Inagaki MN (1986) Crossability of Japanese wheat cultivars with *Hordeum bulbosum* L. Jpn J Breed 36:363–370

Inagaki MN (1997) Technical advances in wheat haploid production using ultra-wide crosses. JIRCAS J 4:51–62

Inagaki MN, Mujeeb-Kazi A (1995) Comparison of polyhaploid production frequencies in crosses of hexaploid wheat with maize, pearl millet and sorghum. Breed Sci 45:157–161

Inagaki MN, Tahir M (1990) Comparison of haploid production frequencies in wheat varieties crossed with *Hordeum bulbosum* L. and maize. Jpn J Breed 40:209–216

Inagaki MN, Nagamine T, Mujeeb-Kazi A (1997) Use of pollen storage and detached-tiller culture in wheat polyhaploid production through wide crosses. Cereal Res Commun 25:7–13

Laurie DA, Bennett MD (1986) Wheat × maize hybridization. Can J Genet Cytol 28:313–316

Laurie DA, Bennett MD (1988) The production of haploid wheat plants from wheat × maize crosses. Theor Appl Genet 76: 393–397

Laurie DA, Snape JW (1990) The agronomic performance of wheat doubled haploid lines derived from wheat × maize crosses. Theor Appl Genet 79 : 813–816

Nei M (1963) The efficiency of haploid method of plant breeding. Heredity 18 : 95–100

Park SJ, Walsh EJ, Reibergs E, Song SP, Kasha KJ (1976) Field performance of doubled haploid barley lines in comparison with lines developed by the pedigree and single seed descent methods. Can J Plant Sci 56 : 467–474

Rajaram S, van Ginkel M, Fischer RA (1995) CIMMYT's wheat breeding mega-environments (ME). In: Li ZS, Xin ZY (eds) Proc 8th Int Wheat Genet Symp. China Agricultural Scientech Press, Beijing, China, pp 1101–1106

Sayre KD, Rajaram S, Fischer RA (1997) Yield potential progress in short bread wheats in northwest Mexico. Crop Sci 37 : 36–42

Singh RP, Villareal RL, Rajaram S, Del Toro E (1989) Cataloguing dwarfing genes *Rht1* and *Rht2* in germplasm used by the bread wheat breeding program at CIMMYT. Cereal Res Commun 17 : 273–279

Snape JW (1989) Doubled haploid breeding: theoretical basis and practical applications. In: Mujeeb-Kazi A, Sitch LA (eds) Review of advances in plant biotechnology, 1985–88. CIMMYT/IRRI, Mexico D.F., Mexico, Manila, Philippines, pp 19–30

Snape JW, Chapman V, Moss J, Blanchard CE, MIller TE (1979) The crossability of wheat varieties with *Hordeum bulbosum*. Heredity 42 : 291–298

Suenaga K, Nakajima K (1989) Efficient production of haploid wheat (*Triticum aestivum*) through crosses between Japanese wheat and maize (*Zea mays*). Plant Cell Rep 8 : 263–266

Suenaga K, Nakajima K (1993 a) Variation in doubled haploid plants of wheat obtained through wheat (*Triticum aestivum*) × maize (*Zea mays*) crosses. Plant Breed 111 : 120–124

Suenaga K, Nakajima K (1993 b) Segregation of genetic markers among wheat doubled haploid lines derived from wheat × maize crosses. Euphytica 65 : 145–152

Yonezawa K, Nomura T, Sasaki Y (1987) Conditions favouring doubled haploid breeding over conventional breeding of self-fertilizing crops. Euphytica 36 : 441–453