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Identification of maintainer and restorer lines for development of wheat hybrids

Aruna Devi Ahirwar, Niharika Shukla, RS Shukla and SK Singh[1*]

Jawahar Lal Nehru Krishi Vishavidhalaya
Jabalpur 482 004, India

Abstract

Hybrid wheat development is an attractive approach for enhancing productivity of wheat. For an efficient hybrid development programme through CMS approach, a stable CMS line, its suitable maintainer line and restorer lines for high pollen and spikelet fertility is needed. An experiment was conducted to identify suitable maintainer and restorer line among 13 high yielding wheat genotype based on spikelet and pollen fertility in the hybrid combinations using 10 CMS lines. Range of complete sterility to complete fertility was observed among 130 hybrid combinations. All the genotypes DBW 17, GW 273, GW 322, GW 366, HI 1531, HI 1544, JW 3020, JW 3173, JW 3211, JW 3269, Lok 1, PBW 343 and WH 147 were good fertility restorers for either of the CMS lines. On the other hand, only seven genotypes, HI 1531, JW 3020, JW 3211, JW 3269, PBW 343, GW 366 and GW 273 were identified as maintainers for any of the six CMS lines, namely JWH 4, JWH 8, JWH 14, JWH 16, JWH 17 and JWH 20. Other wheat genotypes were not able to maintain male sterility in the CMS lines.

Keywords: Wheat hybrid, maintainers, restorers, cytoplasmic genetic male sterility

Introduction

Wheat is a major staple food of world population and occupies about 21.8 per cent to total cultivated area accounting for 35.5 percent of total food grain production at global level. India is maintaining its second position in total wheat production since 2000 AD and recorded wheat production of 94.9 m tons during 2011-12, whereas the wheat production during 2012-13 was 92.46m tons from 29.65 million hectare area with productivity of 3.12 t ha^{-1} (Anonymous, 2013). Similarly Madhya Pradesh has produced 13.13 million ton wheat from 5.30 mha area with average productivity of 2.48 t ha^{-1}. To keep pace with increasing population, the estimated wheat requirement in India is about 109 million tons by 2020. To meet this target, annual increase in productivity level of ~2 per cent is required. Since area under cultivation is gradually decreasing or stable, the projected demand of wheat production can be met only with increased in productivity level at >3.8 t ha^{-1} within a stipulated period. In recent years, the enhanced production is due to enhanced wheat area under cultivation and no quantum jump in productivity of wheat genotypes was observed. Further, global climatic changes are also major oncern for enhancing productivity levels. Among some promising wheat improvement approaches, hybrid wheat is an attractive option for realizing hybrid vigour and, therefore, enhanced yield levels.

Wheat is a self pollinated species, therefore, hybrids can be produced manually but this is commercially unviable option. Therefore, making wheat as cross pollinated species, there is a need to change in the floral biology *i.e.* to allow cross fertilization between two parental lines.

[1] Current address: Directorate of Wheat Research, Karnal-132001
[*] Corresponding author's email: *sksingh.dwr@gmail.com*

Success in hybrid wheat has already been reported in USA, France, Australia, South Africa, UK, China and India. A number of male sterility or pollination control systems are available in wheat, i.e., cytoplasmic genetic male sterility, nuclear genetic male sterility, transgenic male sterility, photoperiod sensitive-genetic male sterility, temperature sensitive-genetic male sterility and male sterility induced through chemical hybridizing agents (Virmani and Edwards, 1983; Singh *et al.*, 2010). Each of the pollination system in wheat has merits and demerits depending on the practical feasibility but the Triticum timopheevii based cytoplasmic male sterility system (Wilson & Ross, 1962) has been widely utilized. Some of the basic and fundamental components for a successful development of hybrid wheat are stable male sterile line (A line with widely opened glumes/ angle), a good agronomic base of maintainer (B line with large anther size, long style) and perfect fertility restorer (R line with large anthers having high pollen number).

There are several problems in hybrid wheat development and among them, lack of genetic diversity among parents for harnessing heterosis, out-crossing/natural crossing potential, complete fertility restoration and efficient and cost effective hybrid seed production technology are notable. To achieve hybrid vigour, it is necessary to identify, suitable parents for conversion into either cytolasmic male sterile or fertility restorer lines. Keeping in view these facts, an experiment was conducted to identify the maintainers and restorers for F1 hybrids of wheat.

Materials and methods

The experimental material consisted of 130 F_1 wheat hybrids from 10 cytoplasmic genetic male sterile (CMS) lines namely,

JWH 1, JWH 4, JWH 5, JWH 8, JWH 10, JWH 14, JWH 16, JWH 17, JWH 20, JWH 23 and 13 wheat genotypes namely, DBW 17, GW 273, GW 322, GW 366, HI 1531, HI 1544, JW 3020, JW 3173, JW 3211, JW 3269, Lok1, PBW 343 and WH 147. These hybrids were made during 2008-09 crop season in line x tester design in which CMS lines were used as female and wheat genotypes were used as tester male lines. The CMS lines used were received from the Directorate of Wheat Research, Karnal as support to hybrid wheat development programme of the university. The investigation was carried out during *Rabi,* 2009-10 under Wheat Improvement Project, Department of Plant Breeding and Genetics at Seed Breeding Farm, College of Agriculture, JNKVV, Jabalpur (MP). This farm is located at 23.90^0N and 79.58^0 E at 411.87 m above msl and represents subtropical, semi-arid climate with hot and dry summer and cold winter with occasional showers. All the 130 F_1's were planted in November, 2009 in a Randomized Complete Block Design with two replications in single row plot of 1.5 m. The crop was raised under high fertility condition following recommended package of practices.

Data were recorded on five randomly selected plants of all the hybrids in each replication. Identification of maintainers and restorers was done on the basis of pollen fertility percentage through pollen viability test and spikelet fertility and sterility percentage. Pollen viability (%) was estimated using 1 per cent solution of aceto-carmine in 45% acetic acid in which well filled, stained and round pollen grains were recorded as viable. The pollen fertility (%) was calculated as ratio of stained and round pollen grains to total pollen grains examined (Singh, 2006; Singh *et al*., 2007). For estimation of spikelet fertility and sterility, five randomly selected emerging ears from each F_1 hybrid were bagged (to avoid outcrossing) before flowering. Spikelet fertility was calculated as a percentage of ratio of number of grains in a spike to the total number of spikelets in a spike. For calculation of number of fertile/sterile spikelets per spike, one spikelet /ear from middle of the spike of each plant was selected randomly and the total number of fertile/sterile spikelets was counted. The pollen fertility and spikelet fertility was categorized as per Virmani *et al.* (1997) for identification of maintainer and restorer lines.

Results and discussion

The perusal of data revealed that spikelet fertility percentage was zero in nine crosses JWH 4/HI 1531, JWH 8/HI 1531, JWH 8/JW 3269, JWH 14/PBW 343, JWH 16/GW 366, JWH 17/JW 3211, JWH 17/GW 273, JWH 17/PBW 343 and JWH 20/JW 3020. These crosses also showed zero per cent pollen fertility except in JWH 14/PBW 343 (0.80%). Sixty crosses showed more than 75 per cent spikelet fertility and among these, JWH 1/HI 1531, JWH 1/ Lok1, JWH1/GW 322, JWH 4/HI 1544, JWH 10/GW 273, JWH 10/DBW 17, JWH 10/JW 3173, JWH 14/GW 366, JWH 16/HI 1531, JWH 20/DBW 17, JWH 23/GW 366, JWH 23/PBW 343 showed 100 per cent spikelet fertility. These 60 crosses also showed more than 80 per cent pollen fertility and among these, JWH 1/HI 1531, JWH 1/GW 322, JWH 5/GW 273, JWH 8/JW 3173, JWH 8/GW 366, JWH 8/DBW 17, JWH 10/GW 273, JWH 10/JW 3269, JWH 16/HI 1531, JWH 17/HI 1531, JWH 20/HI 1544 and JWH 23/PBW 343 showed 100 per cent pollen fertility.

The hybrid combinations based on different CMS lines showed variable spikelet fertility and pollen fertility. The highest spikelet fertility was 100 per cent for JWH 1 (JWH 1/HI 1531, JWH 1/Lok 1, JWH 1/GW 322), JWH 4 (JWH 4/HI 1544), JWH 10 (JWH 10/JW 3173, JWH 10/DBW 17, JWH 10/GW 273), JWH 14 (JWH 14/GW 366), JWH 16 (JWH 16/HI 1531), JWH 20 (JWH 20/DBW 17) and JWH 23 (JWH 23/PBW 343, JWH 23/GW 366) whereas it was 99.15 per cent for JWH 5 (JWH 5/JW 3269), 99.17 per cent for JWH 8 (JWH 8/JW 3173) and 99.08 per cent for JWH 17 (JWH 17/HI 1531). Contrary to this, the least spikelet fertility was zero per cent for JWH 4 (JWH 4/HI 1531), JWH 8 (JWH 8/HI 1531, JWH 8/JW 3269), JWH 14 (JWH 14/PBW 343), JWH 16 (JWH 16/GW 366), JWH 17 (JWH 17/JW 3211, JWH 17/GW 273, JWH 17/PBW 343) and JWH 20 (JWH 20/JW 3020), 0.99 per cent for JWH 23 (JWH 23/JW 3211), 1.60 per cent for JWH 5 (JWH 5/GW 366), 1.65 per cent for JWH 10 (JWH 10/Lok 1) and 2.75 per cent for JWH 1 (JWH 1/JW 3173).

The highest pollen fertility was 100 per cent for JWH 1 (JWH 1/ HI 1531, JWH 1/GW 322), JWH 5 (JWH 5/GW 273), JWH 8 (JWH 8/ JW 3173, JWH 8/ GW 366, JWH 8 /DBW 17), JWH 10 (JWH 10/ JW 3269, JWH 10/ GW 273), JWH 16 (JWH 16/ HI 1531), JWH 17 (JWH 17 / HI 1531), JWH 20 (JWH 20 / HI 1544), JWH 23 (JWH 23 / PBW 343), 99.00 per cent for JWH 4 (JWH 4/ HI 1544) and 98.94 per cent for JWH 14 (JWH 14 /WH 147). On the other hand , the least pollen fertility was zero per cent for JWH 4 (JWH 4 / HI 1531), JWH 8 (JWH 8/ HI 1531, JWH 8/ JW 3269), JWH 16 (JWH 16 /GW 366), JWH 17 (JWH 17 /JW 3211, JWH 17/ GW 273 ,JWH 17/ PBW 343) and JWH 20 (JWH 20/ JW 3020), 0.8 per cent for JWH 14 (JWH 14/PBW 343), 1.19 per cent for JWH 23 (JWH 23/ JW 3211), 1.50 per cent for JWH 5 (JWH 5/ JW 3211), 1.90 per cent for JWH 10 (JWH 10 /GW 322) and 8.13 per cent for JWH 1 (JWH 1 /JW 3173).

All 130 F_1 wheat hybrids under study were evaluated for pollen/spikelet fertility/sterility for identification of maintainers/partial maintainers/partial restorers and restorer lines (Virmani *et al.*, 1997). Pollen viability was observed from 0 -100 per cent indicating complete pollen sterility to complete pollen fertility in the hybrid combinations attempted using CMS lines. Similarly, spikelet fertility was observed from 0 -100 per cent indicating complete spikelet sterility to complete spikelet fertility in these hybrid combinations. The spikelet fertility analysis indicated that nine crosses, JWH 4/HI 1531, JWH 8/HI 1531, JWH 8/JW 3269, JWH 14/PBW 343, JWH 16/GW 366, JWH 17/JW 3211, JWH 17/GW 273, JWH 17/PBW 343 and JWH 20/JW 3020 showed complete spikelet sterility whereas twelve combinations viz., JWH 1/HI 1531, JWH 1/Lok 1, JWH 1/GW 322, JWH 4/HI 1544, JWH 10/GW 273, JWH 10/DBW 17, JWH 10/GW 3173, JWH 14/GW 366, JWH 16/HI 1531, JWH 20/DBW 17, JWH 23/GW 366, JWH 23/PBW 343 showed complete spikelet fertility. Besides these, 46 crosses showed spikelet fertility of 0.86 to 46.72 per cent and four crosses showed 55.78 to 65.6 per cent spikelet fertility.

The pollen fertility analysis indicated that eight crosses, JWH 4/HI 1531, JWH 8/HI 1531, JWH 8/JW 3269, JWH 16/GW 366, JWH 17/JW 3211, JWH 17/GW 273, JWH 17/PBW 343 and JWH 20/JW 3020 showed complete pollen sterility (0%) whereas twelve combinations viz., JWH 1/HI 1531, JWH 1/GW 322, JWH 5/GW 273, JWH 8/JW 3173, JWH 8/GW 366, JWH 8/DBW 17, JWH 10/GW 273, JWH 10/JW 3269, JWH 16/HI 1531, JWH 17/HI 1531, JWH 20/HI 1544 and JWH 23/PBW 343 showed 100 per cent pollen fertility. In addition, 46 crosses showed pollen fertility of 1.10 to 42.66 per cent and six crosses showed 51.42 to 78.03 per cent pollen fertility (Singh and Singh, 2001; Sabar *et al.*, 2007). Hybrid combinations showing complete spikelet sterility and/or pollen sterility are indicative of respective male parents as maintainer lines (Table 1). Similarly, hybrid combinations showing complete spikelet fertility and/or pollen fertility are indicative of respective male parents as restorer lines (Table 2).

Table 1. Complete spikelet / pollen sterility in hybrid combinations

Hybrid	Spikelets fertility (%)	Pollen fertility (%)
JWH 4/HI 1531	0	0
JWH 8/HI 1531	0	0
JWH 8/JW 3269	0	0
JWH 14/PBW 343	0	0.80
JWH 16/GW366	0	0
JWH 17/JW 3211	0	0
JWH 17/GW 273	0	0
JWH 17/PBW 343	0	0
JWH 20/JW 3020	0	0

Table 2. Complete spikelet / pollen fertility in hybrid combinations

Hybrid combination	Spikelet fertility (%)	Pollen fertility (%)
JWH 1/HI 1531	100	100
JWH 1/Lok 1	100	93.70
JWH 1/GW 322	100	100
JWH 4/HI 1544	100	99
JWH 5/GW 273	97.4	100
JWH 8/GW 366	99.04	100
JWH 8/DBW 17	85.71	100
JWH 8/JW 3173	99.17	100
JWH 10/GW 273	100	100
JWH 10/DBW 17	100	94.73
JWH 10/JW 3173	100	99.02
JWH 10/JW 3269	99.14	100
JWH 14/GW 366	100	94.28
JWH 16/HI 1531	100	100
JWH 17/HI 1531	99.08	100
JWH 20/HI 1544	97.19	100
JWH 20/DBW 17	100	83.51
JWH 23/GW 366	100	87.5
JWH 23/PBW 343	100	100

The male parents of hybrids having pollen fertility percentage 0-1.0, 1.1-50.0, 50.1-80.0 and >80.0 per cent were categorized as maintainers, partial maintainers, partial restorers and restorers, respectively. Similarly, the male parents were also categorized based on spikelet fertility as maintainers (0.0%), partial maintainers (0.1-50.0%), partial restorers (50.1-75.0%) and restorers (>75.0%). Out of 13 wheat genotypes under study, HI 1531, JW 3020, JW 3211, JW 3269, PBW 343, GW 366 and GW 273 were identified as maintainers for either of the six CMS lines, namely JWH 4 (HI 1531), JWH 8 (HI 1531, JW 3269), JWH 14 (PBW343), JWH 16 (GW 366), JWH 17 (JW 3211, GW 273, PBW 343) and JWH 20 (JW 3020). However, none of the genotypes were found suitable maintainer for CMS lines JWH 1, JWH 5, JWH 10 and JWH 23. Other wheat genotypes were also not able to maintain male sterility in the CMS lines. On the other hand, all the wheat genotypes were identified as restorers for either of the CMS lines (Table 3). Each CMS line has three or more restorer sources except for JWH 4 where HI 1544 was the only restorer source.

Table 3. CMS lines and their respective maintainer and restorer lines

CMS Line	Wheat variety			
	Maintainer	Partial maintainer	Partial restorer	Restorer
JWH 1	-	HI 1544, JW 3211, JW 3173	-	HI 1531, Lok 1, GW 366, GW 273, WH 147, JW 3020, DBW 17, GW 322, PBW 343, JW 3269
JWH 4	HI 1531,	Lok 1, GW 366, JW 3211, WH 147, JW 3020, DBW 17, GW 322, JW 3173, JW 3269	-	HI 1544
JWH 5	-	HI 1531, Lok 1, GW 366, JW 3211, JW 3020, JW 3173	HI 1544	GW 273, GW 322, JW 3269
JWH 8	HI 1531, JW 3269	Lok 1, JW 3211, GW 273, WH 147, JW 3020, GW 322	-	GW 366, HI 1544, DBW 17, JW 3173, PBW 343
JWH 10	-	HI 1531, Lok 1, GW 366, GW 322	WH 147	HI 1544, JW 3211, GW 273, JW 3020, DBW 17, JW 3173, PBW 343, JW 3269
JWH 14	PBW 343	JW 3211, GW 273, JW 3173, JW 3269	-	HI 1531, Lok 1, GW 366, HI 1544, WH 147, JW 3020, GW 322
JWH 16	GW 366	HI 1544, JW 3211, GW 273	PBW 343	HI 1531, Lok 1, WH 147, JW 3020, GW 322, JW 3173, JW 3269
JWH 17	JW 3211, GW 273, PBW 343	GW 366, JW 3020, JW3173, JW 3269	-	HI 1531, Lok 1, HI 1544, WH 147, DBW 17, GW 322
JWH 20	JW 3020,	JW 3211, GW 273, GW 322, PBW 343	JW 3173	HI 1531, Lok 1, GW 366, HI 1544, WH 147, DBW 17, JW 3269
JWH 23	-	JW 3211, GW 322, JW 3269	Lok 1, GW 273	GW 366, HI 1544, WH 147, JW 3020, JW 3173, PBW 343

From the above results, it was observed that eight hybrid combinations JWH 4/HI 1531, JWH 8/HI 1531, JWH 8/JW 3269, JWH 16/GW 366, JWH 17/JW 3211, JWH 17/GW 273, JWH 17/PBW 343, JWH 20/JW 3020 indicated 100 per cent spikelet as well as pollen sterility and the male parents involved in these combinations may be used for diversification programme for maintainer lines for the respective CMS lines.

Similarly, five hybrid combinations, viz., JWH 1/HI 1531, JWH 1/GW 322, JWH 10/GW 273, JWH 16/HI 1531 and JWH 23/PBW 343 showed 100 per cent spikelet as well as pollen fertility.

The male parents involved in these combinations may be used for restorer line development programme for getting better hybrid combinations.

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