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Research Note

Development of downy mildew resistant maintainer inbreds (B lines) of Pearl millet for Arid environment

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Abstract
Developing early maturing downy mildew resistant male sterile lines of pearl millet, which are also well adapted to the harsh climatic conditions of western Rajasthan, forms an important aspect of the pearl millet breeding programme. To achieve this goal crosses were made among 18 maintainer lines (B lines) suitable for this region. Eighteen crosses found resistant to downy mildew were selfed to obtain F_2 populations. These F_2 populations were evaluated for downy mildew disease at the Central Arid Zone Research Institute, Jodhpur. Disease incidence among populations ranged from 0 to 82% with mean downy mildew incidence of 41% and coefficient of variability 61%. F_2 population resulting form the cross ICMB 95444 x ICMB 93333 was free from downy mildew followed by ICMB 95111 x ICMB 93333, which showed only 6% downy mildew incidence. Five hundred and ninety three F_3 progenies were selected from these F_2 populations. Further evaluation of these F_3 progenies against Jodhpur pathotype of downy mildew revealed that selections from segregating populations of crosses ICMB 95444 x ICMB 94555 gave maximum number (35) of resistant progenies. It was observed that F_2 populations showing higher disease incidence produced less number of resistant F_3 progenies, also disease resistant selections made in the F_2 populations continued to show downy mildew resistance in the F_3 generation. This showed that selection for disease resistance was effective in the F_2 populations. The resistant F_3 progenies were carried forward by pedigree breeding to F_6 and are being utilized in backcrossing for developing new male sterile lines of pearl millet for this region.

Key words
Pearl millet, *Pennisetum glaucum*, ms lines, arid region

Downy mildew[*Sclerospora graminicola* (Sacc.) J. Schrot] is the major disease that limits pearl millet production in the state of Rajasthan and in other millet-cultivating tracts in India. For successful cultivation of pearl millet hybrids in the arid western Rajasthan, it is important that the hybrids are developed using parents that are DM resistant and adapted to the harsh climatic conditions of this region. Development of hybrids of pearl millet in the last forty years has led to its increased productivity and stability, largely in the regions with relatively better environments, while regions like western Rajasthan with poor environments still suffer from low productivity of about 470 kg ha^{-1}. This is because most of the hybrids recommended for this region were developed elsewhere and lacked the desired adaptability and characteristics required for this region (Kelley *et al.,* 1996). Being genetically uniform, single cross hybrids become susceptible to downy mildew within three to five years of their release due to evolution of new strains of the pathogen, specifically adapted to the new hybrid (Thakur *et al.*, 1999). International Pearl Millet Downy Mildew Virulence Nursery (Anonymous, 1998) conducted at the Central Arid Zone Research Institute, Jodhpur during 1995-97, as one of the sites for understanding variability in the pathogen (*S. graminicola*) populations revealed the existence of variable pathotypes, which was supported by Thakur *et al*. (2001) and now this is known as a new virulent (Jodhpur) pathotype (Thakur *et al*., 1998). Thus, for successful cultivation of hybrids for longer period in this area, it is imperative that hybrid and its parents posses resistance to the pathotypes of this region, failing which it would be risky to introduce a hybrid. To check rapid multiplication, evolution and spread of a virulent pathotype, it is essential to broaden the genetic base of the hybrids in cultivation. This can be achieved through genetic diversification of both the parents, seed parent (ms line) as well as restorer parent.Diversification of the genetic base of male sterile (ms) lines has been done by transferring

A_1 cytoplasm to another genetic background, and also by developing new ms lines based on different sources of male-sterility-inducing cytoplasm. However majority of the available pearl millet ms lines still carry A_1 cytoplasm. Several hybrids and their parental lines found to be resistant to DM in other states have shown susceptibility to DM in western Rajasthan, probably due to a different and highly virulent pathotype that occurs in this region (Thakur *et al*., 1998). Significant differences in DM incidence due to differences among pathotypes of Indian origin have been reported (Thakur *et al*., 2001). Hence need was felt to initiate a separate and specific programme to develop DM resistant male sterile lines that are well adapted to the harsh climatic conditions of the western Rajasthan. Such hybrids would lead to better adaptation, beside providing cultivar diversity, giving more choice to farmers. The present study was therefore conducted to evaluate and identify DM resistant B x B F_2 populations and selection of DM resistant progenies for developing DM resistant maintainer inbreds to be utilized for developing new male sterile lines.

On the basis of three years study conducted at the Central Arid Zone Research Institute Jodhpur, a number of male sterile lines were found to be promising for their performance and ability to generate hybrids for this region (Manga *et al*., 2004). These were ICMA 91444, ICMA 92111, ICMA 92444, ICMA 93111, ICMA 93333, ICMA 94111, ICMA 94555, ICMA 95111, ICMA 95444, ICMA 95555, ICMA 96111, ICMA 97555, ICMA 98004, ICMA 98111, ICMA 98222, 841A, and ICMA 98333. Maintainers (B-lines) of these A lines and other male sterile lines CZMS 44A, CZMS 47A developed at Jodhpur (Manga and Yadav 1997) and HMS 9A from HAU Hissar, were crossed during summer of 2002 in a diallel fashion at ICRISAT and following evaluation of these crosses against DM in the seedling screening nursery, 18 crosses were selected and later selfed to obtain segregating (F_2) populations. These F_2 populations of inter-maintainer inbred crosses (B x B) were planted on 11th July 2003 in larger plots of twenty rows each of 4 m length, at the Central Arid Zone Research Institute, Jodhpur during the rainy season (*kharif*) of 2003. The row-to-row distance was 60 cm while plant to plant distance was maintained at 20 cm to allow full expression of traits. The crop was fertilized with 40 kg N ha^{-1} and 20 kg P ha^{-1}. After planting, crop received a well distributed rainfall of 283 mm. This provided sufficient moisture and humidity for development of DM disease (infector rows on the average had DM incidence of 95%). Data on days to 50% flowering and DM incidence at dough stage were also recorded. Agronomic score was recorded on 1 to 5 scale (1=poor, 5=best). Most of the plants in each of the segregating population were selfed. At maturity plants with desirable traits like DM resistance, good tillering, dwarf to medium dwarf plant height, medium to long panicles, compact panicles and having complete panicle exertion were selected for further pedigree breeding of B lines. The selected progenies (F_3) were further evaluated during 2004 in the green house at ICRISAT, for identification of DM resistant lines. Resistant lines were carried forward by selfing and advance generations (F_6) of these lines are being utilized for the development of new male sterile lines for which backcrossing was initiated in the year 2006.

Results and Discussion

As is evident from the table 1, there was lot of variability among the populations for DM incidence. DM incidence ranged from 0 to 82% with mean disease incidence of 41% and coefficient of variability 61%. This variability among the F_2 populations provided ample opportunity to select desirable plants. F2 population resulting form a cross ICMB 95444 x ICMB 93333 was free from DM symptoms and it was followed by ICMB 95111 x ICMB 93333, which showed only 6% DM incidence. Two populations ICMB 94555 x ICMB 96222 and ICMB 97555 x ICMB 95555 showed $\leq$ 20% DM incidence. Maximum DM incidence (82%) was found in population ICMB 93111 x ICMB 97555 and it was followed by ICMB 93111x ICMB 91444 (78%), ICMB 94111 x ICMB 93333 and ICMB 93111 x ICMB 98004 (74% each). Interestingly all the three populations were from crosses involving ICMB 93111 as one of the parents. This line was also found to have second highest DM incidence (26%) at Jodhpur (Manga *et al.,* 2004). Thus ICMB 93111 could be the one imparting DM susceptibility to these populations. Using this opportunity of high downy mildew incidence, selections were made in each of the eighteen populations and selfed seed obtained. Number of plants selected from each F_2 population, are given in table 2. Overall 593 selections were made. ICMB 94555 x ICMB 96333, ICMB 94555 x ICMB 91444, ICMB 94555 x ICMB 96222, ICMB 95333 x ICMB 96333, ICMB 94111 x ICMB 93333, ICMB

94555 x ICMB 92111, ICMB 94555 x ICMB 94111 were late in flowering and took more than 50 days to flower. Rest of the populations flowered around 45 days. Number of progenies selected from an F_2 population depended upon the DM incidence in the population, time taken to flower and agronomic score. More plants were selected from populations that had less disease, early in flowering and high agronomic score. Populations showing less DM, earliness and good agronomic score were ICMB 95444 x ICMB 93333 (0% DM, 4 Ag score and 42 days to flowering), ICMB 95111 x ICMB 93333 (6% DM, 4 Ag score and 44 days to flowering). Consequently maximum progenies were selected from these populations. Both of these crosses had ICMB 93333 as common Parent. This line was found to be the most resistant to DM at Jodhpur (Manga *et al*., 2004). Total of 593 plants were selected on the basis of disease resistance, plant height, compactness of earhead, earliness, tillering and agronomic score to form F_3 progenies. To confirm the DM resistance behaviour of these F_3 progenies and to further use these progenies in breeding programme, these progenies were further evaluated in seedling screening nursery against Jodhpur pathotype. Table 2, showed number of progenies showing $\leq$10% and $\geq$10% DM incidence. Out of 593 F_3 progenies, 273 progenies showed less than 10% disease incidence. Thus 46% of the selections showed less than 10% DM. Selections from segregating populations of crosses ICMB 95444 x ICMB 94555 gave maximum number (35) of resistant progenies, and it was followed by ICMB 94555 x ICMB 96222 (27), ICMB 95111 x ICMB 93333 (27) and ICMB97555 x ICMB 94555 (23). A correlation study between DM incidence in the F_2 populations and number of F_3 progenies showing less than 10% DM in the green house, revealed that a significant negative correlation (-0.48 P 0.05=0.46) existed between these two. Thus F_2 populations showing higher disease incidence produced less number of resistant progenies. Correlation between number of resistant plants selected in F_2 populations and number of resistant F3 progenies was positive. This showed that resistant selections made in the F_2 population continued to show DM resistance in the F_3 generation also, hence selection was effective in F_2. The disease resistant material so identified, was carried forward using pedigree breeding procedure to F_6, and promising B lines (maintainer inbreds) are being utilized for the development of new male sterile lines for this region, for which backcrossing was initiated in the year 2006 and presently it is in the BC6 stage,

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Table 1. Days to flowering, agronomic score and downy mildew incidence (%) in B x B F_2 populations of pearl millet

S.No.	B X B F_2 Populations	Days to 50 % bloom	Agronomic Score	Downy Mildew (%)	No. of plants selected in F_2
1	ICMB 93111 X ICMB 91444	49	3	78	27
2	ICMB 94555 X ICMB 96333	55	2	54	19
3	ICMB 94555 X ICMB 91444	51	3	36	23
4	ICMB 94555 X ICMB 96222	52	3	14	29
5	ICMB 95333 X ICMB 96333	51	3	48	23
6	ICMB 94111 X ICMB 93333	53	2	74	27
7	ICMB 94555 X ICMB 92111	50	2	50	29
8	ICMB 94555 X ICMB 94111	50	2	28	29
9	ICMB 95444 X ICMB 92111	43	2	28	51
10	ICMB 95444 X ICMB 93333	42	4	0	50
11	ICMB 95444 X ICMB 94555	44	2	24	51
12	ICMB 97555 X ICMB 94555	45	2	30	23
13	ICMB 97555 X ICMB 95555	43	2	20	31
14	ICMB 93111 X ICMB 94111	45	3	42	38
15	ICMB 93111 X ICMB 97555	44	3	82	25
16	ICMB 95111 X ICMB 93333	44	4	6	58
17	ICMB 93111 X ICMB 98004	45	2	74	25
18	ICMB 841 X ICMB 93333	44	2	50	35
	Control 843A	44	2	100	-
	Mean			41	
	CV%			61	

Table 2. Downy mildew incidence in F3 progenies derived from B x B F_2 populations

	B X B F_2 Populations	No. of F3 Progenies	Number of F3 progenies showing DM incidence (%)	
			<10%DM	>10% DM
1	ICMB 93111 X ICMB 91444	27	8	19
2	ICMB 94555 X ICMB 96333	19	18	1
3	ICMB 94555 X ICMB 91444	23	10	13
4	ICMB 94555 X ICMB 96222	29	27	2
5	ICMB 95333 X ICMB 96333	23	13	10
6	ICMB 94111 X ICMB 93333	27	11	16
7	ICMB 94555 X ICMB 92111	29	10	19
8	ICMB 94555 X ICMB 94111	29	19	10
9	ICMB 95444 X ICMB 92111	51	12	39
10	ICMB 95444 X ICMB 93333	50	8	42
11	ICMB 95444 X ICMB 94555	51	35	16
12	ICMB 97555 X ICMB 94555	23	23	0
13	ICMB 97555 X ICMB 95555	31	10	21
14	ICMB 93111 X ICMB 94111	38	10	28
15	ICMB 93111 X ICMB 97555	25	8	17
16	ICMB 95111 X ICMB 93333	58	27	31
17	ICMB 93111 X ICMB 98004	25	12	13
18	ICMB 841 X ICMB 93333	35	12	23
	Total	593	273	320