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Identification, characterization and geographic distribution of male-sterility restorer and maintainer lines from diverse pearl millet germplasm

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Summary

To determine the distribution and geographic specificity of sterility maintainers in pearl millet, *Pennisetum americanum* (L.) Leeke, 428 diverse pearl millet germplasm accessions representing variation from 12 countries were crossed with a male-sterile line 5141A. The F_1 hybrids were classified as male-fertile or male-sterile based on the seed set on bagged ear heads and an other morphology. Among these, 87 (20.3%) were classified as male-fertile, 32 (7.5%) as male-sterile, 282 (65.9%) as segregating for male-fertile/male-sterile and 27 (6.3%) behaved as male-fertile in the rainy and male-sterile in the postrainy season. Restorer lines were distributed in all the countries studied except Cameroon and USSR. Maintainer lines were observed from six countries but were concentrated in India. These maintainer lines differ from one another in several morphological and agronomic characters such as flowering, plant height, spike length and grain size. They may prove to be useful sources of material for generating new male-sterile lines. The restorers can be used to produce commercial hybrids.

Introduction

The discovery of cytoplasmic male-sterility in pearl millet (Burton, 1958), led to the production of high-yielding commercial hybrids using the male-sterile Tift $23A_1$. Subsequently, two additional sources of cytoplasmic male-sterility designated 'L $66A_2$' and 'L $67A_3$' were identified (Burton & Athwal, 1967; Burton & Powell, 1968). It is desirable to have an array of desirable male-sterile lines which improve the probability of identifying hybrids with high yield and wide adaptability. In fact, a number of male-sterile lines were developed by genome substitution of Tift $23A_1$ by repeated back-crossing to several maintainers (Burton, 1965; Pokhriyal et al., 1974; Athwal et al., 1976; Appadurai & Sambathkumar, 1976; Sharma, 1980). Downy mildew resistant versions of Tift $23A_1$ were developed by back-crossing with downy mildew resistant mutants which were induced in the maintainer line Tift 23B (Murty, 1974; Anand Kumar & Andrews, 1984). However, it is desirable to have male-sterile lines with varying maturity levels to increase the scope for selecting restorers with good combining ability and adaptation to diverse agroclimatic conditions. The distribution of pollen fertility alleles in some representative accessions of the world collection was reported after crossing to two cytoplasmic

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male sterile lines (Upadhyay, 1971).

Characters and therefore genes do not occur at random in populations (Qualset, 1976). For example, barleys of high elevations in Central and North-Central Ethiopia, which were known for their genetic diversity (Mengesha, 1976), showed much more disease resistance than those from lower altitude areas (Qualset, 1976). The present investigation was aimed at identifying the distribution and geographic specificity of maintainer lines for the A_1 cytoplasm in the pearl millet germplasm. For the purpose of identifying the maintainer lines, diverse germplasm accessions representing variation from different geographic regions were selected to study their sterility maintenance/fertility restoration ability, when crossed onto a female parent used in several commercial pearl millet hybrids in India.

Materials and methods

The male-sterile line utilized as a tester in this study was 5141A, was developed from a downy mildew resistant donor 1587 through back-crossing to Tift $23A_1$ (Pokhriyal et al., 1976). Diverse landrace accessions of pearl millet maintained at ICRISAT Center were used as male parents after one rejuvenation and seed increase. After preliminary evaluation of over 12,000 accessions of the world collection, 428 accessions were selected, representing variation from 12 countries and some of the material generated at ICRISAT (Table 1). Each test entry was planted in an area of $6\,m^2$ accommodating about 100 plants. The female parent was planted thrice at fortnightly intervals. For crossing, both the male and female parents were bagged 3 days before stigma emergence. Pollination was done in the morning hours between 8.00 to 10.00 a.m. In each case, bulk pollen of 2–4 individual plants were used for crossing 5–10 heads of the female parent (5141A). Care was taken to avoid pollen shedders in the female parent. Bulked seed of every cross (F_1) was planted in a row of 4-m length, accommodating about 50 plants. About 5–10 heads from each row were bagged 3 days before stigma emergence. Seed set was observed 3 weeks after flowering. For confirmation, anthers of the open-polli-

Table 1. Distribution of sterility maintainers/fertility restorers in diverse pearl millet germplasm

Origin	Number of accessions				
	Evaluated	R	M	S	R/M
Burkina Faso	4	1	0	3	0
Cameroon	8	0	0	8	0
Chad	7	2	0	5	0
India	95	8	22	56	9
Lebanon	14	3	2	6	3
Malawi	1	1	0	0	0
Mali	36	6	1	27	2
Niger	137	35	0	93	9
Nigeria	31	3	1	26	1
Senegal	48	22	0	26	0
Uganda	14	3	2	9	0
USSR	2	0	0	2	0
ICRISAT	31	3	4	21	3
Total	428	87	32	282	27
Per cent		20.3	7.5	65.9	6.3

R = Restorer; M = Maintainer; S = Segregating as restorer/maintainer; R/M = Classified as restorer in one season and maintainer in the other season.

nated heads of the F_1 hybrids were observed for pollen shedding. The F_1 hybrids were evaluated during the rainy and postrainy seasons, which differ in day length and temperature.

The new maintainer lines identified were evaluated for agronomic characters during rainy and postrainy seasons. Each entry was planted in four rows of 4-m length, spaced 75 cm apart with plants at 10 cm spacing within a row. Flowering (DFL) was recorded as the number of days from sowing to when inflorescences on 50% of the main tillers showed emergence. Plant height (PHT) was measured in centimetres from ground level to the tip of the spike. The number of mature spikes per plant were considered as productive tillers (PT). Spike length (SPL) was measured in centimetres from the base to the tip of the spike. The diameter of the spike measured in millimetres was considered as spike thickness (STH). The mass in grams of 200 randomly selected grains with around 12% moisture content multiplied by five was taken as 1000 grain mass (GM).

Results and discussion

The sterility maintaining/fertility restoring ability of the 428 diverse landrace accessions showed grades of fertility, ranging from complete sterility maintenance to complete fertility restoration. If there was no seed set in bagged ear heads of the hybrid, the accession was classified as a maintainer. If there were seeds on the ear heads, it was classified as a restorer, and a combination of these two was classified as being heterozygous and heterogeneous for this trait, because bulk pollen from different plants was used in producing the hybrids.

Of the 428 diverse pearl millet germplasm lines evaluated, 87 (20.3%) were classified as restorers, 32 (7.5%) as maintainers, and 282 (65.9%) were classified as segregating for restoring/maintaining ability (Table 1). The behaviour of 27 accessions (6.3%) differed in the two test seasons viz., kharif (rainy) and rabi (postrainy). The hybrids that differed for fertility in the two seasons revealed that they behaved as male-fertile in the rainy season while the same hybrids behaved as male-sterile in the postrainy season. This might be because of variation in temperature and/or day length.

It is interesting to note that restorers were present among accessions from 10 out of the 12 countries included in the test (Table 1). However, restorers were concentrated in West Africa, mainly from Niger, Senegal and Mali. All accessions tested from Cameroon, Malawi, Burkina Faso and the USSR were observed to be either restorers or seg-

Table 2. Geographic specificity of sterility maintainers in pearl millet germplasm from India

State of origin	Number of accessions				
	Evaluated	R	M	S	R/M
Andhra Pradesh	15	2	4	9	0
Gujarat	16	1	3	10	2
Madhya Pradesh	20	0	4	12	4
Maharasbtra	14	1	8	4	1
Punjab	1	0	0	1	0
Rajasthan	7	0	1	6	0
Tamil Nadu	1	0	0	1	0
Uttar Pradesh	21	4	2	13	2
Total	95	8	22	56	9
Per cent		8.4	23.2	58.9	9.5

R = Restorer; M = Maintainer; S = segregating as restorer/maintainer; R/M = Classified as restorer in one season and maintainer in the other season.

regating for fertility restoration.

The frequency of maintainers in the germplasm was low, only 7.5%. No maintainers were identified from Burkina Faso, Cameroon, Chad, Malawi, Niger, Senegal, and the USSR. A large percentage (68.7%) of maintainers have their origin in India (Table 2). Of the 22 maintainers from India, eight had their origin in Maharashtra. These eight landrace accessions were collected from farmers' fields near Sangamner, Maunchar, Kokangaon and Nasik in Maharashtra during 1978 (Appa Rao, 1978a). In their original habitat, these landraces were locally known as 'Gaorani' and were usually grown mixed with pulses. These millets grow 100 to

Table 3. Characteristics of the 32 landrace accessions that maintained male-sterility of 5141A

IP No.	Origin	DFL (days)		PT (No.)	(PHT) (cm)		SPL (cm)	GM (g)
		K	R		K	R		
4803	India	71	50	1.8	244	145	21.3	8.90
7499	India	57	66	2.6	218	233	15.5	6.95
7500	India	58	69	4.4	182	249	14.9	4.25
7503	India	-	-	1.8	158	121	18.0	3.70
7507	India	58	75	3.4	132	225	22.2	6.00
7518	India	71	61	2.0	230	84	15.7	6.85
7520	India	57	55	2.4	176	90	13.4	9.40
7521	India	53	56	3.2	163	99	14.8	8.50
7522	India	58	55	3.0	203	123	19.8	9.25
7523	India	52	56	2.0	161	107	14.8	9.50
7525	India	50	47	2.4	170	110	13.8	9.15
7526	India	58	59	4.2	150	123	17.2	10.45
7527	India	48	56	2.4	136	133	14.4	8.80
7532	India	58	56	3.4	177	96	18.0	7.20
7533	India	71	65	2.6	222	125	21.4	7.70
7535	India	53	66	2.0	176	98	18.9	6.90
7538	India	58	62	2.0	196	115	18.9	8.30
7543	India	74	66	1.8	232	118	26.4	7.25
7552	India	71	80	1.2	217	136	23.2	6.70
7554	India	57	69	1.6	163	212	25.0	6.90
7562	India	52	57	2.4	149	92	25.6	9.75
7604	India	68	69	4.2	192	160	14.2	5.75
4856	Lebanon	55	66	2.2	100	107	18.4	8.10
4880	Lebanon	55	62	2.8	122	102	19.0	8.85
7926	Mali	66	63	1.0	174	122	17.7	11.50
5078	Nigeria	68	80	1.0	192	112	38.0	11.00
4948	Uganda	68	65	1.6	151	127	21.8	5.35
4960	Uganda	58	75	1.8	105	146	15.0	8.75
318	ICRISAT	66	67	1.0	147	98	20.4	5.25
4969	ICRISAT	59	75	3.4	176	160	21.0	7.70
7576	ICRISAT	53	82	2.4	153	106	20.6	9.65
7580	ICRISAT	52	58	4.8	134	87	16.7	5.30

DFL = Days to 50% flowering; PT = No. of productive tillers; PHT = Plant height; SPL = Spike length; GM = Mass of 1000 grain; K = Kharif (rainy); R = Rabi (postrainy).

200 cm tall, and produce 2–3 small ear heads (Appa Rao et al., in press). When the 32 maintainer accessions were evaluated under uniform conditions at ICRISAT Center, plant height varied from 136 to 230 cm, days to 50% flowering from 48 to 71, and spike length between 13 and 20 cm (Table 3). The maintainer lines from Andhra Pradesh were collected from the hilly areas of Eastern Ghats and vary enormously for morphological and agronomic characters (Appa Rao, 1978b).

The male-sterile line used in this study has sterile cytoplasm from Tift $23A_1$ and the genome from donor 1587 from Baroda, India (Pokhriyal et al., 1976). The frequency of fertility restoration genes for Tift $23A_1$ in Indian breeding stocks was found to be low, thus restricting the choice of pollinators for heterosis breeding to a relatively small portion of the best adapted breeding lines (Rachie & Majmudar, 1980; Upadhyay, 1971).

The high frequency (65.9%) of segregating lines indicates that the non-restoring genes are often maintained in the populations in heterozygous form. This is expected because pearl millet is a cross-pollinated crop. The identification of geographic areas where the highest frequency of maintainers are found, such as around Nasik in Maharashtra, India, suggest priority areas for collection/screening of pearl millet germplasm for identifying sterility maintainers that can be used to develop new male-sterile lines by repeated backcrossing. The identification of maintainers from seven different countries makes further diversification of male-sterility in different genetic backgrounds possible. The restorers identified can be used to produce commercial hybrids.

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