Breeding Method of Hybrid Forage Sorghum by using Male-Sterile Lines

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Many investigators reported that heterosis is markedly expressed in F_1 hybrids of grain sorghum (Karper and Quinby 1937, Quinby 1963, Quinby et al. 1958). Already in 1964, when the author initiated his breeding of forage sorghum in Japan, everyone was familiar with the impact on production achieved by the commercial use of hybrid grain sorghum.

Hybrid forage sorghum, however, was unknown in commercial production and Japanese farmers pointed out the slower growing rate and lower forage yield, compared with corn, of their local sorgo and sudangrass cultivars (Duvick 1959, Poehlman 1959, Harada et al. 1966).

Early in his breeding program for improving growth rate and yield, the author found that the F_1 hybrids, even those produced by using dwarf grain sorghum male-sterile lines as seed parents, gave high forage yield (Tarumoto and Oizumi 1967a, 1968a and 1969a). In 1971, his new variety, "Sendachi" (Sorghum Norin 1) was officially licensed and two promising strains, Chugoku-ko 2 and Chugoku-ko 3, entered final regional (prefectural) trials (Tarumoto 1971).

In this paper the author would like to review this work and also to suggest an efficient breeding method for using male sterility.

Review

1) *Classification of sorghum cultivars to facilitate the breeding of forage sorghum*

Based on a literature survey of cytogenetical and morphological characteristics and on field studies of varietal performance, the author developed the following classification of sorghum cultivar groups into four major parental types (Tarumoto 1971). This classification will prove helpful in advancing forage sorghum breeding in Japan.

Table 1. Classification of sorghum spp.

Type	Group	Scientific name	Chromosome number (2n)
Grain sorghum	Feterita	S. vulgare Pers.	20
	Kafir (including male-sterile lines)	S. vulgare Pers.	20
	Hegari	S. vulgare Pers.	20
	Milo	S. vulgare Pers.	20
	Special purpose	S. vulgare Pers.	20
Sorgo	Sorgo	S. vulgare Pers.	20
Grass sorghum	Sudangrass	S. vulgare Pers. (var.) sudanenses	20
	Colombus grass	S. almum Parodi	40
	Johnsongrass	S. halepense (L.) Pers.	40
Broomcorn	Broomcorn	S. vulgare Pers.	20

2) Charatceristics of male-sterile lines

Twenty-five male-sterile strains of grain sorghum, each consisting of an A-line (male-sterile) and a B-line (fertile and non-restorer) and two sudangrass male-sterile strains were introduced, mainly from the United States in 1964 and in 1968, respectively. This section describes the investigations that were conducted to clarify the fertility and forage characteristics in those strains.

(1) Male-sterile lines of grain sorghum

1) All A-lines except Reliance and all B-lines except 4692 showed stable sterility and fertility, respectively, in the self-pollination test conducted by bagging heads under various environmental conditions. Male-sterile strains were easily maintained by cross-pollination between the corresponding A- and B-lines. It was not difficult to produce fertile F_1 hybrids by cross-pollination between grain sorghum A-line and other diploid sorghum cultivars with restorer genes (Tarumoto and Oizumi 1967b). Furthermore, the mass-production of F_1 seed by wind-fertilization using grain sorghum A-lines was easy, using either forage sorghum or grain sorghum cultivars as pollen parents (Tarumoto 1971).

2) It is generally recognized that the male-sterile lines of grain sorghum have almost the same characteristics as cultivars of the Kafier group because of their breeding history. This was also confirmed by this study, and among these male-sterile lines many morphological variations were found (Tarumoto and Oizumi 1967b). The lines were classified for forage yield, stem length at maturity (Table 2), days to 50% heading and tillering number at the early regrowth stage.

3) As forage plants, the male-sterile lines of grain sorghum have no defective characters

Table 2. Statistical relationships and classification of male-sterile lines for forage yield and stem length

Total green forage yield			Stem length at the mature time		
Male-sterile line	kg/a	Statistical significance[1)]	Male-sterile line	cm	Statistical significance[1)]
4692	540.5	a	390	156.7	a
390	484.9	a b	4692	130.3	b
Redlan	424.3	b c	Redlan	108.9	c
Martin-1	411.0	b c d	Redbine 58	100.9	c d
Redbine 58	389.6	c d	3197	94.5	d e
Wheatland	373.9	c d	Reliance	94.1	d e
3128	362.8	c d	Martin-2	93.7	d e
Martin-2	361.0	c d	385	93.4	d e
385	353.0	c d	Combine Kafir 60	93.0	d e
3127	350.8	c d	Martin-1	90.3	d e f
3197	349.4	c d	Westland	82.7	e f
Westland	342.2	c d	Wheatland	77.8	f g
3121	330.5	c d	3121	69.4	g h
Combine Kafir 60	318.1	d	406	63.1	h
Reliance	215.2	e	3128	59.6	h
			3127	59.2	h

(Classification)			(Classification)		
High yield	>450 kg/a	4692, 390	2-dwarf type>120 cm		390, 4692
Medium yield	250-450 kg/a	Redlan CK. 60	3-dwarf type	70-120 cm	Redlan～Wheatland
Low yield	<250 kg/a	Reliance	4-dwarf type>70 cm		3121, 406, 3128, 3127

1) Measured by Duncan's multiple range test (5% level). Means belonging to the same subgroup (same letter) are not significantly different

except short plant height and low forage yield. It was planned to use these lines as seed parents for breeding F_1 hybrids of forage sorghum.

(2) Male-sterile lines of sudangrass

The B-line of tall Rhodesian sudangrass showed stable fertility, but the sterility of the A-line was not stable under various temperatures. Both the A-line and B-line of dwarf Rhodesian were fertile in Japan. Seeds of these sudangrasses frequently shattered before harvesting.

3) Heterosis and combining ability by a diallel cross in sorghum

Diallel crossings among the cultivars representing each type of sorghum classified in Table 1 were made to evaluate heterosis, inbreeding depression and combing ability in F_1 and F_2 hybrids.

Heterosis was strongly expressed in the F_1's in forage yield and its components, especially plant height. This was particularly so in the crosses which involved cultivars of far distant relationships, for example in the F_1's of grain sorghum × sudangrass. Conversely, inbreeding depression in F_2's appeared greatest in the same characters which showed high heterosis in F_1's. The diallel analysis showed that both additive and non-additive gene effects are important in F_1, but only additive in F_2. These results suggest the following conclusions:

(1) F_1 hybrid forage sorghums will prove as advantageous for producing forage as F_1 grain sorghums are for prdoucing grain. It is possible to develop F_1 hybrids with high forage yield even when using the dwarf male-sterile lines of grain sorghum as seed parents.

(2) General combining ability as well as specific combining ability should be emphasized in selecting superior F_1 crosses between cultivars of distant relationships.

4) Heterosis and combining ability in F_1 *hybrids between grain sorghum A-lines and cultivars of other morphological groups.*

In the foregoing two sections, the "dwarf" grain sorghum A-lines were found to be very useful seed parents for developing hybrid forage sorghum. At the third step, two sets of top crosses between grain sorghum A-lines, representing 2-, 3- and 4-dwarf types, and representative cultivars of four groups of sorghum (Table 1) were made to clarify the genetical and physiological basis of heterosis and combining ability in F_1 hybrids.

F_1 hybrids between grain sorghum A-lines and hegari cultivars (designated MS-HE hybrid hereafter) and F_1's between grain sorghum A-lines and sudangrass cultivars (MU-SU hybrid) showed remarkably high heterosis in forage yield and its components (Fig. 1). There were highly significant differences in general combining ability among both seed parents and pollen parents. Cultivars of hegari and sudangrass groups were found to be good general combiners with grain sorghum A-lines.

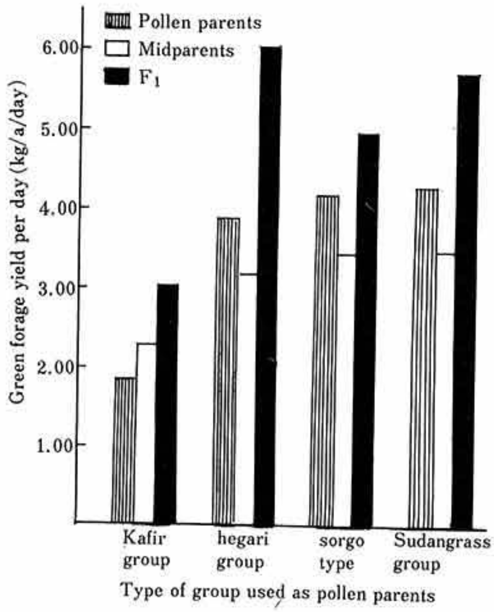

Fig. 1. Average green forage yield per day for pollen parents, mid-parents and F_1's

However, general conclusions were not drawn about the effects of dwarf genes because the dwarf genes had only a small effect on forage yield and its components in the forage sorghum hybrids. This contrasts with the results in grain sorghum hybrids reported by Hadley et al. (1965), Graham and Lessman (1966), Casady (1965, 1967) and others.

I consider that the vigorous growth in MS-HE hybrids depends largely on the combination effects of multiple gene action on plant height and of complementary gene action on maturity. Therefore, the high forage yield of MS-HE hybrids results from tall growth and late maturity. On the other hand, the high yielding ability of MS-SU hybrids is presumably attributed to: (1) the rapid elongation of leaves and stems at the initial and regrowing stages, and (2) the higher percentage of effective tillers.

5) Combining ability in F_1 *hybrids between grain sorghum A-lines and hegari cultivars*

Studies were conducted to find the best way of selecting high forage yield in F_1 hybrids of the MS-HE combination, which is one of the most promising. The 4×4 top crosses were examined in a droughty field in 1968 and in both a droughty field and a field with optimum soil moisture in 1969. In 1968 the plots were harvested using the "Successive Cutting Method" (each plot cut when 50% headed) and data were obtained on heading

Table 3. Analysis of variance adapted from Design II (Comstock and Robinson, 1948) for analyzing quantitative genetic traits in forage sorghum populations. The appropriate F-test should be performed by repeated use of Error mean square for both G. C. A.'s and S. C. A.'s

1) Trials in a single year and locations

Source of variations	D. F.	Expectation of mean squares
Replications	$r-1$	
Hybrids	$mf-1$	
G_i's (G. C. A. of R-lines)[1]	$m-1$	$\sigma^2+rf\sigma^2_{g_i}$
G_j's (G. C. A. of A-lines)	$f-1$	$\sigma^2+rm\sigma^2_{g_j}$
S_{ij}'s (S. C. A.)[2]	$(m-1)(f-1)$	$\sigma^2+r\sigma^2_{S_{ij}}$
Error	$(r-1)(mf-1)$	σ^2

1) General combining ability
2) Specific combining ability

2) Trials over several years or locaotins

Source of variations	D. F.	Expectations of mean squares
Replicates	$k(r-1)$	
Experimental Field[1]	$k-1$	$\sigma^2+rmf\sigma^2_1$
Hybrids	$mf-1$	
G_i's (G. C. A. of R-lines)[2]	$m-1$	$\sigma^2+rfk\sigma^2_{g_i}$
G_j's (G. C. A. of A-lines)	$f-1$	$\sigma^2+rmk\sigma^2_{g_j}$
S_{ij}'s (S. C. A.)[3]	$(m-1)(f-1)$	$\sigma^2+rk\sigma^2_S$
Hybrid×Field	$(mf-1)(k-1)$	
G_i's×Field	$(m-1)(k-1)$	$\sigma^2+rf\sigma^2_{g_i1}$
G_j's×Field	$(f-1)(k-1)$	$\sigma^2+rm\sigma^2_{g_j1}$
S_{ij}'s×Field	$(m-1)(f-1)(k-1)$	$\sigma^2+r\sigma^2_{S1}$
Error	$k(mf-1)(r-1)$	σ^2

1) Field: (1) "Tenjin" field (Fukuyama, Hiroshima) of granite, residual soil which becomes dry in summer. (2) "Higashi-Fukatsu" field (Fukuyama, Hiroshima) of gray, clayed soil which is wet in summer
2) General combining ability
3) Specific combining ability

date and other traits at the 50% heading stage.

The "Simultaneous Cutting Method" in which all the plots in a test are harvested simultaneously at each harvesting date (which were determined by the 1968 test) was used in 1969 to obtain more critical information. The method shown in Table 3 was used for analyzing the data obtained in this manner.

(1) Almost all the MS-HE hybrids showed similar morphological and ecological characteristics. They were taller, had relatively more tillers, greater stalk diameter and longer and wider leaf blades in comparison with the familiar cultivars of the sorgo group. The MS-HE hybrids showed high sensitivity to short days due to maturity genes derived from hegari cultivars, so that their heading date was delayed substantially by long days. In MS-HE hybrids, it is necessary to make a preliminary screening of types for height and maturity before selecting the cross parents, since the combinations of both height and maturity genes affect heterosis and combining ability, as was described in Section 4.

(2) General combining ability was found to be an important selection criterion for forage yield and its components in both years. The comparative size of variance components and the correlation coefficients between the two fields in 1969 showed that it would be more effective to test the MS-HE hybrids in the droughty field, where differences in combining abiilty were better expressed.

(3) The following scheme is recommended as the most effective way to develop the MS-HE hybrids with high forage yield. The first step is to test many top crosses in relatively small plots using the "Successive Cutting Methods", to identify suitable height and maturity genotypes and roughly select parents for high general combining ability. This will be followed by a large plot test using the "Simultaneous Cutting Method" in a relatively droughty field for selecting the best crosses.

6) Combining ability in F_1 *hybrids between grain sorghum A-lines and sudangrasse cultivars*

Studies similar to those on MS-HE hybrids were conducted on MS-SU hybrids, another very promising combination, to find the best way to select high forage yield. The results were as follows;

(1) All the MS-SU hybrids were similar to each other in both morphological and ecological types in the test using the "Successive Cutting Method", in which various characters could be observed at the optimum harvest time for each hybrids. They were taller, had slightly fewer but more effective tillers, greater stalk diameter and longer and wider leaf blades in comparison with the common cultivars of the sudangrass group. All the MS-SU hybrids also showed rapid elongation in leaves and stems in the initial and regrowing stages. This uniformity would be because the sudangrass cultivars are not too dissimilar from each other especially in tillering, maturity and height compared with hegari cultivars.

(2) Because 1968 had normal rainfall and 1969 was very dry it is suggested that high-yielding MS-SU hybrids will be developed by emphasizing specific combining ability in the "Successive Cutting Method" in 1968 and general combining ability in the "Simultaneous Cutting Method" in 1969. Reasonably large differences also were shown in the results of combining ability tests between the two fields in 1969. These differences are attributed to the fact that inheritance in the MS-SU hybrids is easily affected by the artificial and natural external-conditions. Therefore, it is important to select parents with high general combining ability, based on the average performance in two environments differing in soil moisture.

(3) It is concluded that the following scheme would be one of the most convenient ways to develop high-yielding MS-SU hybrids. A top-cross test with at least two locations and/or years should be conducted by using the "Simultaneous Cutting Method", for

selecting parents with high general combining ability. An evaluation test follows to screen the best hybrids for particular environmental conditions.

7) Growing responses to soil and meteorological conditions of MS-HE and MS-SU hybrids

The adaptability tests using several superior MS-HE and MS-SU hybrids were conducted in order to determine their growth responses to soil moisture and meteorological conditions.

(1) The genotype-environmental interaction was studied in two fields differing substantially in soil moisture. Differences due to the soil moisture were significant in plant height and forage yield, but no appreciable interaction was detected. The rankings of entries were the same in both fields.

These results, however, largely depend on the materials used here, which had already been selected in relatively droughty fields. Therefore, it would be necessary, in general, to conduct the drought test prior to the adaptability test.

(2) Three locations with different meteorological conditons were chosen for the adaptability test. The MS-SU hybrids had more tillers and greater plant height than the MS-HE hybrids in each location, especially in locations and seasons with relatively lower temperatures. On the other hand, MS-HE hybrids were superior to MS-SU hybrids in stalk diameter and leaf, especially in relatively hot locations and seasons. Reflecting these growth responses, MS-HE hybrids yielded much forage in the low altitude location, with a relatively high temperature throughout the growing season. MS-SU hybdids performed best in the high altitude location, with a relatively low temperature during the growing season. The interaction between meteorological conditions and hybrids, therefore, was significant for forage yield.

The adaptability test should be conducted in locatinos with different meteorological conditions, especially in temperature.

8) New forage sorghum hybrids, "Sendachi" and Chugoku-ko 2"

The MS-SU hybrid of 605A × Sweet Sudan and the MS-HE hybrid of 399 × Regs. Hegari gave high forage yields not only in the preliminary yield tests at our station but also in the adaptability test at several locations and years. They were named "Chugoku-ko" and "Chugoku-ko 2" in February 1968.

Since then, other evaluation tests were conconducted to determine which varieties to recommend as forage crops. Shugoku-ko 1" was recognized to be superior to the commercial F_1 hybrids such as "Sweet Sorgo" and "Sudax SX-11" especially in forage yield, drought tolerance and adaptation ability in the southwestern part of Japan. It was licensed officially by the Ministry of Agriculture and Forestry in Japan as "Sorghum Norin-ko 1" with the varietal name "Sendachi" in 1971

Fig. 2. Sendachi and its parents. Leff to right: 605A (Female), Sendachi and Sweet Sudan (Mate)

(Fig. 2). The other MS-HE hybrid of 390A × Regs. Hegari through the same steps was named "Chugoku-ko 3" in 1970. This strain instead of "Chugoku-ko 2" is now being evaluated in the final test. "Chugoku-ko 3" is expected to be licensed in 1975.

Incidentally, the characteristics of the parents of these crosses have been slightly changed from those of the original introductions from the United States, because we slected heads by the pure-line selection method in order to improve adaptability to the southwestern parts of Japan.

Proposal for breeding hybrids by using male-sterile lines

Fig. 3 shows not only the author's forage sorghum breeding program described in Part 1 but also one of the breeding schemes which the author should like to propose for breeding hybrids of other crops by using male-sterile lines. In this program there is one premise, that we shall be able to obtain the male-sterile lines from other sources. Developing male-sterile lines requires at major program of

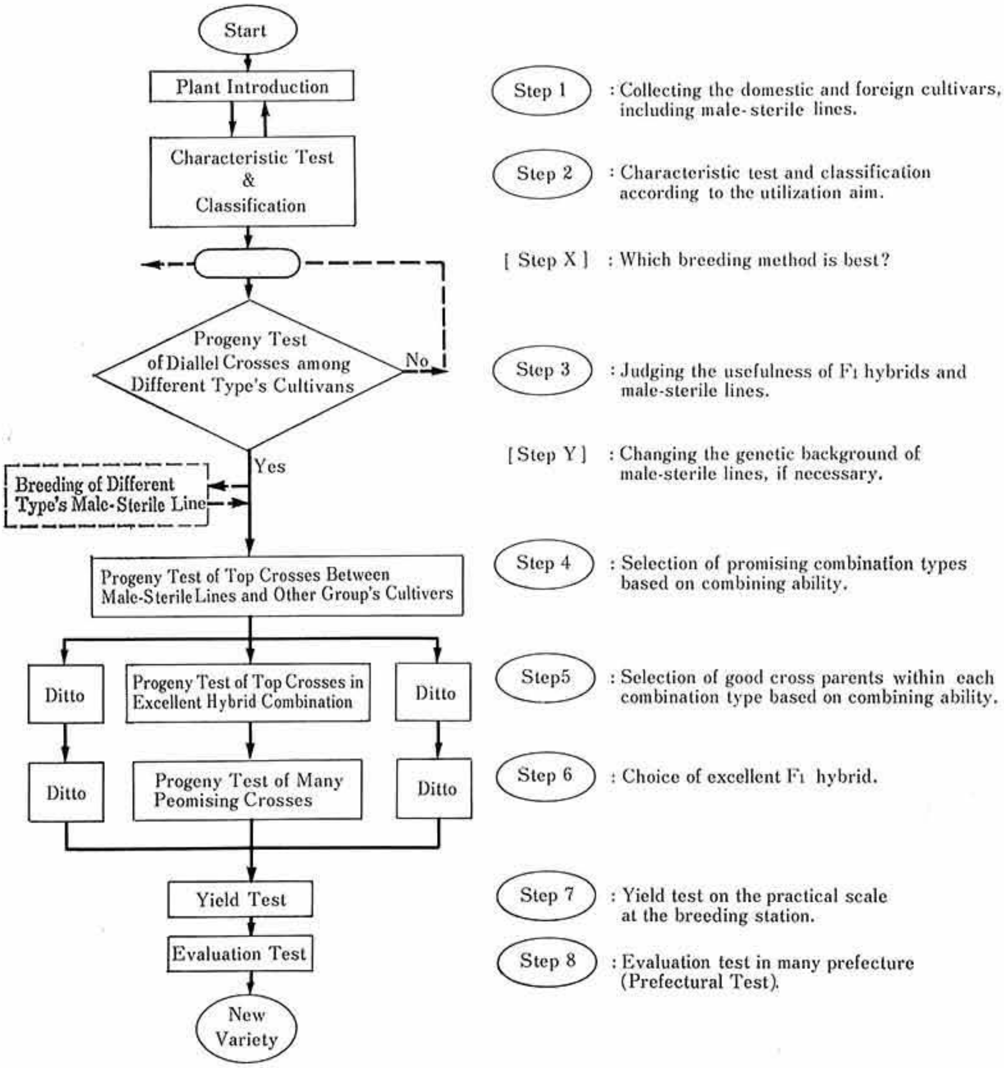

Fig. 3. One breeding procedure for using male sterility

its own.

Since Steps 2 to 6 are important in his proposal, some explanations are in order. In Step 2 the characteristic test should be conducted according to the utilization aim and the classification shown in Table 1 will be helpful in furthering the breeding program, especially in Steps 3 and 4.

In Step 3 we determine: (1) if heterosis expressed in F_1 hybrids is large enough to recover the increased seed price, (2) if male-sterile lines obtained from other persons are suitable for developing excellent F_1 hybrids, and (3) which procedure is the more effective for advancing the breeding. When (1) and (2) are "yes", we can go to Step 4. When (1) is "yes" but (2) is "no", we have to go through Step Y and change the gentic background of the male-sterile lines. When (1) is "no" despite (2), we have to go back to Step X and seek another breeding scheme.

In Step 4 we select some groups that combine well with the male-sterile lines mainly based on general combining ability, as described in 4 of Part 1. In Step 5 we obtain more critical information on a particular hybrid combination type and can choose good parents based on both general and specific combining ability. In Step 6 the promising F_1 hybrids in each combination type will be selected, based on their performance of yield, quality, disease resistance, etc.

As the breeding program shown in Fig. 3 is for the case in which everything is unknown, we can change or skip over some steps according to the objective crop and/or proceeding stage of the breeding in that crop. For example, where the phenomena described in Step 3 are already known, we can skip over Step 3. After having gone through all steps once, we can return to Step 4 and/or Step 5.

Acknowledgment

This work was carried out mainly at the Crop Science Division, Chugoku National Agricultural Experiment Station, Hiroshima, Japan and partly at the Pasture Plant Division, National Grassland Research Institute, Chiba, Japan. The author wishes to thank the breeding staffs of forage sorghum, Hiroshima Prefectural Agricultural Experiment Station, for increasing the hybrid seed and conducting the final prefectural test of the licensed variety "Sendachi", and Dr. D. R. Sampson, Cereal Crops Section, Ottawa Research Station, Central Experimental Farm, Ottawa, Canada, for reading the manuscrip and making valuable suggestions.

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