

Genetic variability between and within progenies of sugarcane crosses developed by modified polycross method at the seedling selection stage

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ABSTRACT

Seedlings from a modified polycross involving seven male sterile and 11 male fertile clones were evaluated in the screen house of the Unilorin Sugar Research Institute, Ilorin, in plant and ratoon crops. The objective was to assess the utility of the breeding scheme. Between family variability was higher than within family variability for almost all the characters, indicating that higher number of progenies per maternal parent should be raised to maximize genetic variability among progenies of the same parent, and thus increase the probability of obtaining superior progenies. Narrow-sense heritability, correlation coefficients, and repeatability estimates also indicated that yield is a reliable indicator for selecting superior progenies at the seedling stage for further advancement.

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Introduction

The conventional hybridization procedure in sugarcane (*Saccharum officinarum* L.) entails immersing cut canes in sulphurous acid (H_2SO_3) solution to effect crosses. When used in conjunction with the breeding lantern and in a specialized glass house, it gives the breeder the opportunity of setting up planned crosses in an isolated situation which is devoid of contamination by wind-borne pollen. In Nigeria, these facilities are lacking. Sugarcane breeders, therefore, have resorted to either induced mutation, followed by

RÉSUMÉ

OLAJOYE, G. : *Variabilité génétique entre et dans les progénitures des croisements de canne à sucre développés par la méthode de croisement multiple modifiée au stade de la sélection de semis.* Les semis d'un croisement multiple modifié comprenant sept clones mâles stérile et onze mâles fertiles étaient évalués dans l'essai en cage de l'Institut de Recherche en Sucre d'Unilorin, à Ilorin dans les cultures de plante et de repousse. L'objectif était d'évaluer l'utilité du procédé de reproduction. La variabilité entre la famille était plus élevée que la variabilité dans la famille pour presque tous les caractères, indiquant qu'un nombre plus élevé des progénitures par parent maternel devrait être cultivé afin de maximiser la variabilité génétique parmi les progénitures de la même parent et augmenter ainsi la probabilité d'obtenir les progénitures supérieures. L'héritabilité au sens restreint, les coefficients de corrélation et les estimations de la capacité de répétition aussi indiquaient que le rendement est un indicateur fiable pour la sélection des progénitures supérieures au stade de semis pour plus d'avancement.

selecting useful variants (Obajimi, 1980), or raising fuzz (seeds) from open-pollinated arrows (flower) of male sterile clones. Since induced mutation acts by disrupting the coding sequence in a gene, it has the limitation of producing several undesirable traits along with the desired ones. On the other hand, the parentage of seedlings raised from open-pollinated arrows cannot be determined. Furthermore, the probability of obtaining superior progenies from such a method is very low.

In recognition of the limitations inherent in the two approaches, a breeding scheme was,

therefore, proposed with two distinct features. The first feature entails the grouping of all available male and female clones within the germplasm into early, mid, and late arrowing by period of arrowing. The second feature is the establishment of crossing blocks in the field between male and female clones whose arrowing period could be synchronized, while maintaining sufficient isolation distance from the nearest sugarcane plot. Since pollen is assumed to have come at random from any of the males used in the scheme, the genetic relationship which can be determined is that of the offspring of the same maternal parent (here that of half-sib).

Progeny testing is one method of evaluating the effectiveness of any breeding programme. Previous studies in sugarcane varietal development programme elsewhere (Thomas, 1978; 1983) have shown that certain characters, *viz* stalk weight and yield/stool, when measured at the seedling stage can be useful in increasing the proportion of superior progenies even when such measurements are taken on potted seedlings. Seedlings have been raised from fuzz collected from the arrows of male sterile clones used in this breeding scheme.

The study examines the performance of the progenies at the seedling stage as an index of the utility of the proposed breeding scheme.

Materials and methods

The materials and methods have been described elsewhere by Olaoye & Ogundipe (1997). Briefly, fuzz were collected after the 1994 arrowing season from arrows of seven male sterile clones, *viz* CO396, CO404, CO443, LS-026, LS-047, LS-098, and DB20/58 which were intermated with 11 male fertile clones, using a modified polycross method (Fig. 1). Equal number of stalks of the male fertile clones were cut into three-budded setts and used to plant the male rows. Each of the female rows was represented by two stalks placed in 3-m long rows and sandwiched in between the male rows. The fuzz, which were collected after ripening, were sown into wooden flats containing sterilized top

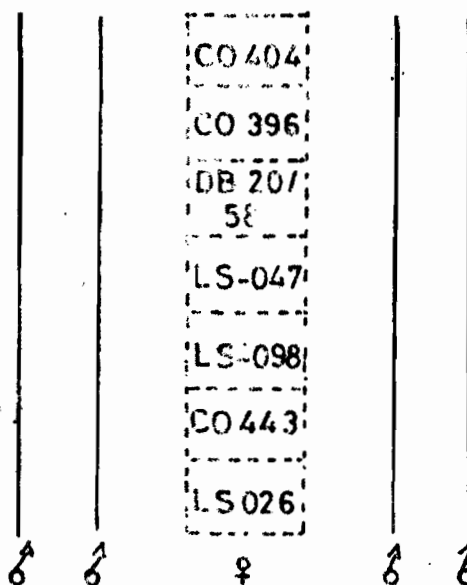


Fig. 1. Outline of the breeding plan showing the mating scheme between selected male clones as pollinator rows to the identified female clones.

List of male clones BJ6552, BR6223, CO440, CO453, CO6806, CO1001, Dacca, IAC 48/69, LS-054, LS-050, Mex 52/29

♂ = Male ♀ = Female

soil, filter mud, and bagasse of equal proportion in the screen house of the University of Ilorin Sugar Research Institute (USRI), Ilorin. All cultural operations (daily watering, weekly application of ferrous sulphate solution, and fertilizer application) to ensure good germination and optimal seedling performance were carried out.

At 28 days after sowing (DAS), 30 vigorous seedlings (by visual observation) were selected from each of the maternal parents and transplanted into black polythene bags containing the same medium. The seedlings were later transferred to 10-l plastic pots containing sterilized top soil at 109 DAS. The pots, which were perforated at the bottom to allow for proper drainage, were then arranged as a completely randomized block design in the screen house, but in such a way that the identity of each seedling could be traced to the maternal parent.

Data were collected from individual seedlings

at harvest (at 6 months old) on stalks/stool, stalk length, stalk diameter, number of internodes/stalk, number of green leaves/plant, leaf length, leaf width, stalk weight, yield/stool, and brix (an estimate of sucrose accumulation in the juice). All data except stalks/stool and yield/stool were collected from the primary stalk. The plants were allowed to regrow and the same data as for the plant cane were collected from the ratoon after 6 months. Leaf area was estimated from leaf length and leaf width as, $LA = \frac{3}{4} BW \times L$, where BW is the greatest width of leaf and L is leaf length.

The data were subjected to analyses of variance (ANOVA), first on individual crop basis before a combined ANOVA over harvest stage (HS) was performed. The combined data were analyzed as a nested design (Table 1) to partition the variance components into between and within

family variances. Additive (δ^2A) and dominance (δ^2D) genetic variances were estimated from the maternal parent and offspring components of variances, respectively. From the expectations in Table 1,

$$\delta^2f = \text{Cov}(\text{HS}) = \frac{1}{4} \delta^2A = 4 \delta^2f$$

$$\text{while, } \delta^2o/f = \text{Cov}(\text{FS}) - \text{Cov}(\text{HS}) = (\frac{1}{2} \delta^2A + \frac{1}{4} \delta^2D - \frac{1}{4} \delta^2A).$$

Narrow sense heritability (H^2N) was computed for each character as, $H^2N = \delta^2A / (\delta^2A + \delta^2D)$, while clonal repeatability (rc) estimate for each character was computed by using the phenotypic coefficient of a character in the plant cane with the same character in the ratoon crop.

Results

Results of the analyses of variance (ANOVA) showed significant differences ($P < 0.01$ or 0.001) between the two harvest stages (HS) for all the characters studied except for stalk length and leaf area (Table 2). The progenies also differed significantly ($P < 0.001$) for all the characters.

Between family means for the two crops (Tables 3 and 4) showed significant differences only for cane yield, brix and two of the yield compo-

TABLE 1

Combined Analysis of Variance (ANOVA) Format for the Harvest Stages (HS) Combined and the Expected Mean Squares

Source	df	MS	EMS
Harvest stages (HS)	(hs-1)	M5	
Females (F)	(f-1)	M4	$\delta^2_c + hs\delta^2o/f + o\delta^2hsf + hso\delta^2f$
HS x F	(hs-1)(f-1)	M3	$\delta^2_c + hs\delta^2o/f + o\delta^2hsf$
Offsprings/F	f(o-1)	M2	$\delta^2_c + hs\delta^2o/f$
Pooled error	f(o-1)(hs-1)	M1	δ^2_c

TABLE 2

Mean Squares from the Combined Analyses of Variance (ANOVA) for Yield and Related Traits in Progenies of Sugarcane

Source	df	Stalk weight 10 ⁴	Yield/stool 10 ⁴	Brix	No. of inter-nodes/stalk 10 ²	Stalks/stool	Stalk length 10 ⁴	Stalk diameter	No. of green leaves/ stalk	Leaf area 10 ⁴
HS	1	173.41***	260.71***	83.53*	15.10***	951.91***	221.91	1.50**	401.74***	14.64***
F	6	1.33	2.45*	32.13	0.28*	2.44	0.02	0.18	3.53	0.27
HS x f	6	0.96	1.68	33.96	0.24	4.77	0.08*	0.46	4.41	0.40
O/f	168	0.78***	1.49***	14.74***	0.12***	2.55***	0.04***	0.21***	11.22***	0.22***
Pooled error	168	0.69	1.29	14.70	0.11	3.19	0.03	0.20	8.01	0.25

*; **, ***; significant at 0.05, 0.01 and 0.001 levels of probability, respectively. HS = harvest stage; f = female parent; o = offsprings.

TABLE 3

Means for Cane Yield and Related Traits of Progenies of Sugarcane Crosses in the Plant Crop

Female parent	Stalk weight (g)	Yield/stool (g)	Brix	No. of internodes/stalk	Stalks/stool	Stalk length (cm)	Stalk diameter (cm)	No. of green leaves/stalk	Leaf area (cm ²)
CO396	125.30 ^{ab}	195.2 ^b	10.8 ^a	2.7 ^a	8.5 ^a	109.9 ^b	1.94 ^a	9.53 ^a	162.1 ^a
DB20/58	107.10 ^{ab}	176.8 ^b	7.3 ^b	2.9 ^a	12.1 ^a	123.9 ^a	1.97 ^a	9.47 ^a	167.0 ^a
LSI-047	124.90 ^{ab}	189.4 ^b	9.3 ^{ab}	2.5 ^a	11.3 ^a	116.0 ^{ab}	1.98 ^a	10.10 ^a	182.9 ^a
CO443	144.60 ^{ab}	194.7 ^b	8.0 ^{ab}	2.8 ^a	9.7 ^a	120.9 ^a	2.02 ^a	9.57 ^a	178.2 ^a
LSI-098	132.60 ^{ab}	183.1 ^b	6.9 ^b	2.6 ^a	11.4 ^a	120.1 ^a	1.95 ^a	9.20 ^a	164.9 ^a
LSI-026	143.30 ^{ab}	193.7 ^b	7.9 ^{ab}	2.9 ^a	11.0 ^a	120.4 ^{ab}	1.98 ^a	8.67 ^a	172.1 ^a
CO404	156.70 ^a	236.1 ^a	8.9 ^a	2.7 ^a	10.8 ^a	118.3 ^a	2.05 ^a	8.93 ^a	172.9 ^a
SE ±	14.83	17.02	1.27	1.12	0.29	4.52	0.09	1.06	11.11

Means followed by the same alphabet(s) in a column are not significantly different.

TABLE 4

Means for Cane Yield and Related Traits of Progenies of Sugarcane Crosses in the Ratoon Crop

Female parent	Stalk weight (g)	Yield/stool (g)	Brix	No. of internodes/stalk	Stalks/stool	Stalk length (cm)	Stalk diameter (cm)	No. of green leaves/stalk	Leaf area (cm ²)
CO396	253.04 ^a	337.4 ^b	7.6 ^a	6.3 ^a	5.1 ^a	63.3 ^{bc}	2.34 ^a	7.80 ^a	153.5 ^a
DB20/58	277.16 ^a	369.7 ^{ab}	7.3 ^a	6.1 ^a	4.8 ^a	57.7 ^{bc}	2.04 ^a	7.16 ^a	144.2 ^a
LSI-047	284.03 ^a	378.7 ^{ab}	7.9 ^{ab}	6.8 ^a	5.5 ^b	72.7 ^a	1.95 ^{bc}	6.96 ^a	155.4 ^a
CO443	314.69 ^a	419.6 ^a	7.3 ^{ab}	6.7 ^a	6.0 ^a	63.0 ^{bc}	2.02 ^{ab}	6.20 ^{ab}	128.2 ^a
LSI-098	233.31 ^a	311.1 ^b	7.9 ^a	6.1 ^a	6.0 ^a	63.8 ^{bc}	2.17 ^{abc}	7.12 ^a	131.9 ^a
LSI-026	257.64 ^a	343.5 ^{ab}	7.0 ^a	5.2 ^a	5.2 ^b	65.2 ^{ab}	2.06 ^{abc}	6.64 ^a	129.1 ^a
CO404	291.63 ^a	388.8 ^a	7.8 ^a	6.2 ^a	6.0 ^a	66.6 ^{ab}	1.91 ^{bc}	6.48 ^a	116.4 ^b
SE ±	14.83	17.02	0.44	0.38	0.51	3.83	0.13	0.46	12.50

Means followed by the same alphabet(s) in a column are not significantly different.

nents (stalk length and dia-meter) in the plant cane (Table 3). In the ratoon crop, between family means were different for almost all the characters except stalk weight, number of internodes/stalk, and stalks/stool (Table 4). Mean stalk weight was higher for all families in the ratoon crop but brix was lower. Progenies of var. CO404 had significantly higher stalk weight in the plant cane while in the ratoon crop, progenies of vars. CO404 and CO443 were superior to the others for the same character. For brix, vars. CO404 and CO396 were superior to others in the plant cane, but

between family means were the same for this character in the ratoon.

The range in the means for the seedling population (Table 5) indicated a lot of variability amongst the progenies for all the characters that were studied. In consonance with the estimates for between family (δ^2_f) and within family (δ^2_o/f) components of variances, estimates of additive genetic variance (δ^2_A) were larger in magnitude than the dominance variance (δ^2_D) for all the characters. Narrow sense heritability (H^2_N) estimates were high for stalk weight, yield/stool,

TABLE 5

Variance Components (δ^2), Narrow-sense Heritability (H^2_N), and Repeatability (rc) for Cane Yield and Related Traits in Progenies of Sugarcane Crosses

Parameter	Stalk weight (g)	Yield/stool (g)	Brix	No. of internodes/stalk	Stalks/stool	Stalk length (cm)	Stalk diameter (cm)	No. of green leaves/stalk	Leaf area (cm ²)
Mean	202.68	277.79	8.01	8.26	4.05	90.12	2.09	8.07	157.36
SE \pm	(111.59)	(141.37)	(3.88)	(4.04)	(2.23)	(31.71)	(0.46)	(3.25)	(53.18)
Range	90-388	172-578	4.5-15.5	5-12	6-12	57-118	1.5-2.4	5-17	97.3-239.5
δ^2_e	0.59	1.29	14.7	0.11	3.10	0.03	0.20	8.01	0.25
$\delta^2_{o/f}$	2.73	4.10	51.59	0.42	8.93	0.14	0.74	39.92	0.77
δ^2_f	4.57	8.36	109.80	0.78	8.06	0.06	0.59	0.81	0.90
δ^2_A	18.08	33.44	439.20	3.12	32.24	0.24	2.36	43.24	3.60
δ^2_D	6.40	8.04	96.56	0.90	27.66	0.50	2.37	146.27	2.18
δ^2_{ph}	7.69	13.75	176.39	1.31	20.09	0.23	1.53	58.74	1.29
H^2_N	0.74	0.81	0.82	0.77	0.54	0.32	0.50	0.33	0.62
rc	0.279	0.353*	0.121	0.471**	0.209	0.420**	0.580**	0.300*	0.742**

*, **; Significant rc values at 0.05 and 0.01 levels of probability, respectively.

number of internodes/stalk and brix, while estimates for stalk length and number of green leaves/plant were low.

Simple linear correlation coefficients between cane yield (stalk weight and yield/stool) and yield components (number of internodes/stalk, stalk

diameter, and stalks/stool) were positive either in the plant cane, the ratoon or both crops combined (Table 6). Surprisingly, the correlation of yield/stool and brix was positive in the plant cane ($r = 0.305^{**}$), but negative (though non-significant) in the ratoon crop. There was no association

TABLE 6

Simple Linear Correlations Between Cane Yield and Related Traits Among Progenies of Sugarcane Crosses in the Plant Crop (on diagonal) and Ratoon Crop (off diagonal)

Character	Stalk weight	Yield/stool	Brix	Internodes/stalk	Stalks/stool	Stalk length	Stalk diameter	No. of green leaves	Leaf area
Stalk weight		0.789**	0.006	-0.373**	-0.026	-0.026	0.711**	-0.543**	0.299*
Yield/stool	0.705**		0.305*	-0.414**	-0.101	-0.226	0.762**	-0.391**	0.176
Brix	-0.146	-0.152		-0.745**	-0.330**	-0.929**	-0.115	0.356*	-0.000
Internodes/stalk	0.510*	-0.509**	-0.594**		0.064	0.741**	-0.067	-0.028	0.172
Stalks/stool	0.258	0.245	0.351**	0.352**		0.505**	0.119	-0.536**	-0.258
Stalk length	0.107	0.105	0.341**	0.310**	0.377**		0.311**	-0.292*	0.172
Stalk diameter	-0.335**	-0.337**	-0.215	-0.038	-0.127	-0.422**		-0.226	0.574**
No. of green leaves/plant	-0.653**	-0.639**	0.314*	0.033	-0.569**	-0.195	-0.251		0.374**
Leaf area	-0.216	-0.215	0.239	0.382**	-0.593**	0.127	0.291*	0.742**	

*, **; Significant rc values at 0.05 and 0.01 levels of probability, respectively.

whatsoever between brix and stalk weight. The association between number of green leaves/plant and cane yield were negative in both crops while that between number of green leaves/plant and leaf area were high ($r = 0.374^{**}$ and 0.742^{**}) in both crops.

Estimates of clonal repeatability (rc) were high for leaf parameters, but moderate for yield, some of the yield components (number of internodes/stalk, stalk length, and stalk diameter), and number of green leaves/plant (Table 5). Estimates for other characters were negligible.

Discussion

Sugarcane is a polyploid and highly heterozygous with chromosome number in the range of $2N = 40$ to $2N = 226$ (Daniels & Roach, 1987). Maximum variability is expected from the F_1 of a cross involving two parents of contrasting characters. Thus, the resulting progenies show an array of genotypes which permits the breeder to select desirable ones for advancement and further selection such as observed from the significant o/f components of variance (Table 2), and the wide range in the means for all the characters studied when seedling population was considered (Table 5).

The between family component of variance (δ^2_f) which was larger in magnitude than the within family ($\delta^2_{o/f}$) for yield, brix, and leaf parameters indicates the variability between progenies of a different maternal parent and the other, but variation among progenies of the same maternal parent was lower. This result implies that larger number of seedlings than used in this study should be raised from a female parent to maximize progress from selection in the F_1 . Although this was anticipated such that higher number of seedlings per maternal parent was actually intended for the breeding scheme, the many female parents used and the limited space in the screen house necessitated the number selected. Wu *et al.* (1978) also suggested that a minimum of 40 seedlings per cross would be adequate to recover the desired progenies from a given cross.

Narrow sense heritability indicates the additive genetic variance in relation to the observed variability. The preponderance of δ^2_A , from which high H^2N for yield was derived (Table 5), suggests a high genetic variance for yield among the progenies and indicates that many of the females combined very well with the males that were used in the scheme, or that there was a low environmental influence. The second hypothesis appears more valid, since this study was conducted in the screen house where the seedlings had been provided with a near ideal environment. Stalk length and number of green leaves/plant recorded very low values which indicate that selecting for these characters may be very ineffective at the seedling stage. The high heritability ($H^2N = 0.82$) but negligible repeatability ($rc = 0.121$) for brix (Table 5) indicates that the seedling character cannot be used for adult plant selection. The high correlation of the character with yield in the plant cane, but negligible association with stalk weight (from which it was measured) is a further proof.

In conjunction with heritability, associations between characters and repeatability of characters between crops are very important tools in sugarcane varietal development and testing. For example, Thomas (1978) noted that yield/stool at the potted stage may provide a useful indication of the likelihood of promising clones emerging from the progeny of a cross without necessarily lengthening the period of the breeding programme. Thomas (1983) further observed that yield/stool measurements of potted sugarcane seedlings at 6-7 months old closely correlated with yield/stool at later stages of selection.

The significant association of some yield components with yield as well as the significant repeatability estimates recorded for these characters in this study, therefore, indicate their usefulness in improving selection for yield even at the seedling stage. The negative association of number of green leaves/plant with cane yield in this study tends to suggest that it may be unimportant at the seedling selection stage, since

field studies (Olaoye, 1987, 1995; Singh, Singh & Singh, 1995) have shown that it contributes significantly to observed variation in cane yield. The similar values for correlation of cane yield (stalk weight and yield/stool) with other characters in the ratoon crop is also likely because most of the tillers had negligible contribution to yield/stool. This may be one of the limitations of seedling selection in pots, especially if carried beyond the plant cane.

Conclusion

The effectiveness of any breeding programme partly depends on the choice of parents and on how well such parents combine in crosses. Evaluating the progenies involving such parents can test this. Between family means indicate that although all the female parents combined well with the males that were used in the scheme, the CO-series probably combined better with the males than the others, judging by their progeny means. The fact that they are more recent than the adapted (LS-series) may be responsible for this difference.

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