

RESEARCH ARTICLE

GENETIC IMPROVEMENT AND DEVELOPMENT OF GENETIC STOCKS IN SUGARCANE THROUGH BACKCROSS BREEDING

S. Alarmelu*, Adhini. S. Pazhany and C. Jayabose

Abstract

Broadening genetic base through backcross breeding is an effective approach to generate diverse genetic stocks for important economic traits coupled with red rot resistance. Three hundred backcross hybrids developed by backcrossing elite F₁ clones (derived by utilizing improved clones (*S. officinarum*, *S. robustum* and *S. barberi*) with improved *S. officinarum* or improved *S. robustum* or Co canes showed variability for the traits studied. Heritability estimates varied over the different nobilising generations and study indicated that selection of parents for sucrose % during the nobilisation process will be effective. BC₁ and BC₂ hybrids developed from improved *S. robustum* x improved *S. officinarum* showed an improvement of 16.71% and 34.09%: 22.36% and 40.58% for sucrose % at 360 days in comparison with improved *S. officinarum* and *S. robustum* parents, respectively. Crosses with PIR 96-285 and PIR 001188 as one of backcross parents generated high quality recombinants. The introgression of new genes provided backcross progenies with redrot resistance. The prebred clones expressed wide variability for various traits like internode colour, cane thickness, leaf colour, canopy, hairiness, cane yield and quality characters. Among the pre bred materials, 26 clones recorded single cane weight > 1.25 kg, 45 clones with brix % above 20% and 41 clones with juice sucrose above 18.5% and most of them were derived from the crosses involving improved *S. officinarum* x improved *S. robustum* parents back crossed to improved *S. officinarum* or Co canes. Elite trait specific BC₁ hybrids viz., 13-69, 13-251, 13-103, 13-208, 13-253, 14-57 and 14-60 and BC₂ hybrids viz., 14-42, 14-66, 14-161 combining good quality and yield traits coupled with red rot resistance: 13-57, 13-114, 13-147 and 13-251, 14-76, 14-69, 14-163 and 14-174 with thickness and resistance to red rot can be utilized in sugarcane varietal improvement programmes.

Key words: sugarcane, interspecific hybrid, prebred, genetic stocks

Introduction

Sugarcane breeding has been a success all over the world by intercrossing few initially developed interspecific hybrids hence, a very few *Saccharum* species clones have been exploited in the breeding programme. The exploitation of the unutilized clones of cultivated and wild species of *Saccharum* in breeding program was suggested as early in 1960's (Berding and Roach, 1987) who indicated that the repeated use of few parents in hybridization have resulted in narrow genetic diversity in modern sugarcane cultivars.

In the present scenario, sugarcane cultivars under cultivation have genetic background of 20 *S. officinarum*, two *S. spontaneum* and a couple of *S. barberi* and *S. sinense* clones which may be a major threat to the sugarcane productivity and demands concerted efforts to utilize new genetic diversity (Walker, 1987; Nair, 2012). Wild species is the basic genetic material available in nature to breeders for continuous evaluation, characterization and exploitation in base broadening and for the development of better varieties. The paradigm shift in the climate has necessitated the need for

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continuous supply of potential valuable genes for crop yield, quality and biotic stresses. Base broadening (Kennedy, 2001) / Pre-breeding, aims at creating new base population by gene introgression that enhance genetic variability in the germplasm and to identify trait specific potential donors for direct use in breeding programmes and avoid genetic vulnerability due to uniformity. It offers the most important alternative to connect genetic resources and breeding programmes and will also assist in identifying heterotic groups for future programs and trait specific genetic stocks for genetic enhancement. Eberhart (1971) suggested that the base population developed from the first backcross to adapted germplasm is the best to start selection. Backcrossing is an efficient way to transfer characteristics controlled by one or two genes, although it can be used also for higher number of genes, including quantitative characters. The wild species used for introgression breeding in the present study was improved *S. officinarum*, improved *S. robustum* and *S. barberi* since it is far more genetically diverse than current commercial cultivars thereby making it useful for broadening the genetic base.

Materials and Methods

Elite improved clones of *S. officinarum* and *S. robustum* produced through intra population improvement programme at ICAR-Sugarcane Breeding Institute (SBI Annual Report, 1991-1996) and commercial hybrids were utilized in introgression through backcross breeding. The breeding material comprised thirty five back crosses effected in 2012 involving eight improved *S. officinarum* (PIO 90-202, PIO 94-345, PIO 00548, PIO 00581, PIO 00760 ,PIO 00764, PIO 00845 and PIO 001100), ten improved *S. robustum* (PIR 89-369, PIR 96-258, PIR 96-475, PIR 03-107, PIR 001157, PIR 001054, PIR 001058, PIR 001062, PIR 001124 and PIR 00-1174) and six commercials (Co 775, Co 86011, Co 94008, Co 09014, CoA 7602 and CoC 671)

as backcross parents. The resulted introgressed progenies were evaluated in ground nursery (2013) and clonal trials (2014 - 2015) for HR brix, Number of millable canes (NMC) and cane thickness, cane height (cm) at 360 days and clones with cane thickness (above 2.5 cm), cane height (> 200 cm) ,NMC (> 40/6m row) and HR brix (>19.5%) and juice sucrose % (>18.0 %) were selected. A combined evaluation of 300 selected clones (from two backcross generation from different groups) was conducted in RBD of two replications and plot size of 6m x 1R for yield and quality traits during 2016 and 2017 at ICAR - Sugarcane Breeding Institute, Coimbatore. Normal agronomic and cultural practices were followed to get uniform phenotypic expression in hybrids. Observations were recorded on three canes selected randomly in each replication on agronomic characters such as NMC/row, cane thickness, cane height, Single cane weight (SCW), estimated cane yield/row and quality traits viz., brix % and sucrose % at 300 and 360 days after planting. Analysis of variance was performed on yield and yield related traits using standard RBD analysis. Elite BC₁, BC₂ hybrids were identified for each trait based on mean performance over the parents and genetic improvement in backcross populations was studied in comparison with base population.

Results and Discussion

The means for the F₁ and two backcross generations viz., BC₁, BC₂ generations showed progressive increase of yield traits and sucrose % with successive stages of nobilisation. Heritability estimates varied over the different nobilising generations and estimates indicated that selection of parents for sucrose % during the nobilisation process will be effective. The results indicate that gain from selection for yield traits like number of millable canes and single cane weight at the BC₁ and BC₂ stages was moderate to high.

Selection during nobilisation

The prebred clones developed (utilizing improved *S. officinarum*, improved *S. robustum* and commercial Co canes), from three nobilized generations and different mating groups exhibited extensive variability for both qualitative and quantitative traits. They expressed wide variability for the traits viz., cane population, cane height, cane diameter, canopy colour, internode shape, internode color, spines and flowering. The means for the F₁, BC₁, and BC₂ generations showed progressive increase of yield traits and sucrose % with successive stages of nobilisation. Heritability estimates varied from 31.20% - 62.14% over the different nobilising generations. The results indicate that gain from selection for yield traits like number of millable canes and single cane weight at the BC₁ and BC₂ stages was moderate to high. In general, sucrose % have shown moderate to moderately high heritability and clump yield of low to moderate heritability.

Genetic progress in the backcross generations

In the improved *S. robustum* x improved *S. officinarum* group, the first generation nobilized hybrids showed an improvement of 15.12%

for sucrose % at 360 days over the improved *S. officinarum* parents (Table 1). BC₁ hybrids showed an improvement of 16.71% and 34.09% for sucrose % at 360 days in comparison with improved *S. officinarum* and *S. robustum* parents, respectively. BC₂ hybrids showed an improvement of 22.36% and 40.58% for sucrose % at 360 days in comparison with improved *S. officinarum* and *S. robustum* parents respectively. Number of millable canes, cane height and single cane weight exhibited a substantial improvement compared to their improved *S. robustum* *S. officinarum* parents. BC₁ and BC₂ exhibited improvement for both yield and quality parameters suggesting further exploitation.

The selected hybrids (123) from the improved *S. officinarum* x improved *S. robustum* group also exhibited improvement for both yield and quality traits. There was an improvement of 23.57% for sucrose in F₁, 30.37% in BC₁ and 33.35% in BC₂ in comparison with improved *S. officinarum* parents (Fig. 1). Similarly an improvement of 41.97%, 49.78% and 53.21% was observed in F₁, BC₁ and BC₂ respectively in comparison with improved *S. robustum* parents. The BC₃ hybrids from these two mating groups also exhibited improvement for

Table 1. Genetic improvement in backcross population of Improved *S. robustum* x Improved *S. officinarum*

Improved <i>S. robustum</i> x <i>S. officinarum</i>	Mean performance at 360 days			
	NMC / row	C. ht (cm)	SCW (kg)	Sucrose % (360 d)
F ₁ (N = 43)	91.00	270.00	0.99	18.12
% imp over <i>S. o</i> parents	82.00	17.39	-7.48	15.12
% imp over <i>S. r</i> parents	37.88	8.00	16.47	32.26
BC ₁ (N = 30)	85.00	280.00	1.13	18.37
% imp over <i>S.o</i> parents	70.00	21.74	5.61	16.71
% imp over <i>S.r</i> parents	28.79	12.00	32.94	34.09
BC ₂ (N = 30)	74.00	300.00	1.34	19.26
% imp over <i>S.o</i> parents	48.00	30.43	25.23	22.36
% imp over <i>S.r</i> parents	12.12	20.00	57.65	40.58
Improved <i>S.officinarum</i> parents	50.00	230.00	1.07	15.74
Improved <i>S.robustum</i> parents	66.00	250.00	0.85	13.70

yield and quality traits suggesting further studies on cytogenetical behaviour and breeding potential in further backcross generations.

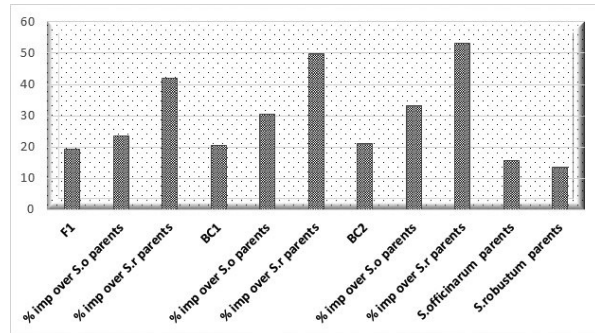


Fig.1. Genetic improvement for sucrose % in backcross population of Improved *S.officinarum* x Improved *S.robustum*

Evaluation of introgressed hybrids

Nine families viz., PIR 001157 x PIO 00845, PIO 001057 x PIR 001062, PIR 03-107 x PIO 96-475, PIO 94-345 x PIR 96-258, PIR03-107 x PIR 96-475, PIO 001100 x PIR 001174, PIO 001057 x PIR 001100, PIR 96-285 x Co 09014 and PIO 94-345 x PIR 96-258 yielded more selections combining yield and quality traits and the elite clones from these families based on synchrony in flowering were subsequently backcrossed with recurrent parent or commercial canes. Crosses with PIR 96-285 and PIR 001100 as one of the back cross parents gave high quality recombinants. The prebred clones expressed wide variability for various qualitative traits like internode colour, cane thickness, leaf colour, canopy and hairiness. Significant variation was recorded for cane yield and quality characters in the prebreeding population. Among the prebred materials, 26 (F₁:6, BC₁:11 and BC₂:9) clones recorded single cane weight >1.25 kg, 45 clones (F₁:7, BC₁:17, BC₂: 21 with brix % above 20%) and 41 clones (F₁:5, BC₁:16: BC₂: 20) with juice sucrose above 18.5% and most of them were derived from the crosses involving improved *S. officinarum* x improved *S. robustum* hybrids

either back crossed to improved *S. officinarum* or Co canes. The trait-based selection approach in the hybrid population combined high yield potential, quality and red rot resistance.

Introgressed hybrids for cane parameters

Nineteen backcross hybrids viz., 13-69, 13-114, 13-253, 14-171, 14-59, 14-179a, 13-36, 13-38, 13-39, 13-44, 13-78, 13-80, 13-125, 13-150, 13-151, 13-198, 13-272, 13-266 and 13-250 were found promising for number of millable canes in comparison with improved parents and commercial check Co 86032. The clone 13-147 recorded the maximum single cane weight of 1.45 kg and nine clones viz., 13-69, 13-114, 13-253, 14-171, 14-59, 14-179a, 14-57, 14-160, 14-161 were identified with higher single cane weight than the checks. Five clones viz., 13-57, 13-69, 13-114, 13-147 and 13-251 were thick types and recorded cane diameter above 2.8 cm. (Table 2). Thirteen other clones viz., 13-36, 13-38, 13-39, 13-44, 13-78, 13-80, 13-125, 13-150, 13-151, 13-198, 13-272, 13-266 and 13-250 expressed good field stand with good NMC, cane height and cane thickness and were identified as genetic stocks for further base broadening programmes. Among *S. barberi* backcross hybrids 14-60 recorded 2.87 cm and 3.07 cm cane thickness and 1.07 kg and 1.25 kg single cane weight at 300 and 360 days respectively.

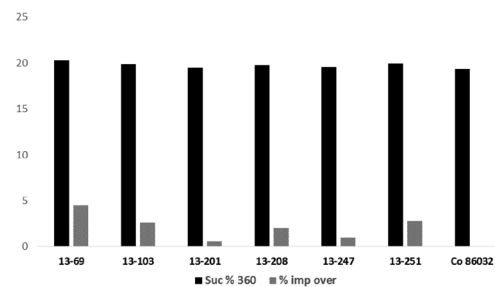
Introgressed hybrids for juice quality (sucrose %)

Eleven BC₁ hybrids viz., 13-57, 13-69, 13-76, 13-103, 13-114, 13-186, 13-201, 13-208, 13-147, 13-251 and 13-253 (Table 2) recorded juice sucrose in the range of 18.00 - 20.26% and performed better than the improved parents and Co 86032. These elite clones identified for high sucrose % at 300 and 360 days were from improved *S. officinarum* x improved *S. robustum* crosses. The clone 13-69 with

Table 2. Performance of BC₁ hybrids

S. No	Clone	Brix (%) 300d	Suc (%) 300d	SCW (kg)	Brix (%) 360d	Suc (%) 360d	C. dia (cm)
1	13-57	20.12	18.03	1.08	20.58	18.20	3.04
2	13-69	20.12	18.65	1.38	22.10	20.26	2.80
3	13-76	20.93	18.73	1.00	21.15	19.14	2.50
4	13-103	18.35	16.30	1.04	21.85	19.89	2.50
5	13-114	18.07	15.02	1.28	20.46	18.00	2.89
6	13-186	19.22	17.98	1.06	21.32	19.38	2.50
7	13-201	19.47	17.81	0.81	21.27	19.50	2.11
8	13-208	19.99	18.39	1.21	21.26	19.77	2.71
9	13-147	21.33	19.25	1.45	21.45	19.57	3.04
10	13-251	21.25	19.04	1.15	21.27	19.92	2.81
11	13-253	20.97	18.77	1.20	21.84	19.34	2.72
	Co 86032	19.75	18.13	1.18	21.32	19.40	2.80
	SE	0.22	0.26	0.03	0.28	0.31	0.04
	CD	1.48	1.60	0.28	2.01	2.24	0.39

the highest sucrose of 20.26% at 12 months of age is from the cross combination of (PIO 03-107 x PIR 96-475) x PIO 03-107. The BC₁ hybrids 13-69, 13-103 and 13-251 showed an improvement of 4.54, 2.63 and 2.79% for quality: juice sucrose % at 360 days (Fig. 2) and more recombinants with high mean brix was obtained with improved *S. officinarum* as one of the parent in backcrosses. Among the BC₂ clones 14 - 42 recorded the highest sucrose of 22.71% followed by 14-66 (21.86%), and 14-161 (21.06%) at 360 days in comparison with Co 86032 (19.03%). The clones, 14-76, 14-69, 14-163 and 14-174 recorded more NMC and estimated cane yield above 78.53 kg/ row. Two BC₁ hybrids viz., 14-57 and 14-60 (Co 8371 x Pathri (*S. barberi*)) x Co 0209 recorded juice sucrose of 18.22 and 18.17% respectively at 300 days in comparison with Co 86032.

**Fig. 2.** Elite clones and % improvement for Sucrose % at 360 days

The pre-bred clones were tested for red rot through CCT and for natural incidence of smut. The clones involving PIR 001057, PIR 001058, PIR 96-285, PIR 001022 as one of the parent in the back crosses were moderately resistant and moderately susceptible to red rot. Among the tested hybrids, 16 were moderately resistant, 10 were resistant to red rot and 24 were moderately

susceptible. The back cross hybrids viz., 14-57 and 14-60 from Co 8371 x *S. barberi* cross were MS and MR to red rot respectively.

The elite BC₁ hybrids viz., 13-69, 13-251, 13-103, 13-208, 13-253, 14-57 and 14-60 and BC₂ hybrids viz., 14-42, 14-66, 14-161, 14-125 and 14-102

combining good quality and yield traits coupled with red rot resistance and 13-57, 13-114, 13-147 and 13-251, 14-76, 14-69, 14-163 and 14-174 (Table 3) with thick canes and resistance to red rot can be utilized as parental donors for further use in sugarcane varietal improvement programmes.

Table 3. List of trait specific genetic stocks

Trait	Genetic stocks
Sucrose %	13-57, 13-69, 13-76, 13-103, 13-114, 13-186, 13-201, 13-208, 13-147, 13-251, 13-253, 14-57, 14-42, 14-66, 14-161, 14-125, 14-102
Cane diameter (cm)	13-251, 14-76, 14-69, 14-60, 14-163, 14-174
Number of millable canes	14-76, 14-69, 14-163, 14-174
Single cane weight (kg)	13-69, 13-114, 13-253, 14-171, 14-59, 14-179a, 14-57, 14-160, 14-161, 14-102, 14-125

Though introgression of wild germplasm is time consuming and requires considerable efforts and resources, it is very important to improve the commercial breeding populations and pre-breeding is a promising alternative to link genetic resources and breeding programs. The products of base broadening programme have reached several stages of selection in many countries and varieties of commercial use from *S. spontaneum* have been released for cultivation in Barbados (Kennedy, 2001). Horsley and Zhou (2014) have suggested for improvement of BC₁ populations for the coastal hinterland breeding programmes of South Africa using high sucrose-yielding parents as sucrose levels progressively increased in subsequent backcrosses. Jackson *et al.* (2014) indicated in his studies that the overall challenge linked with introgression breeding of basic germplasm into commercially adapted germplasm is that the basic germplasm carries along with it many undesirable traits which need to be selected through back crossing to the commercially superior parental material, while at the same time desirable traits and genes from the wild

donor may be diluted or lost with successive generations. The variability created through backcrossing in our study serves as new base population for further utilization. Introgression through backcross-breeding strengthens the breeding pool with genes for desirable economic characters and resistance to diseases as evidenced in this study and serves as an efficient source of genetic diversity. The backcross population of *S. robustum* may be an alternate source for quality and yield traits and superior back cross families and trait specific stocks for yield, quality and resistance to red rot and with better adaptability to the prevailing changing climatic scenario were identified. Utilization of these clones by the breeders will help in the development of trait specific populations to work upon and on identifying clones with stable resistance to red rot, smut and tolerance to drought and salinity stresses. The study suggest further research on cytogenetical behaviour and genomic-assisted pre-breeding which will help to overcome the linkage drag and aid in transfer of useful genes for sugarcane genetic improvement.

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References

- Berding N, Roach BT (1987) Germplasm collection, maintenance and use. In: Heinz, D.J. (Eds.) *Sugar Cane Improvement through Breeding*. Elsevier Scientific Publishers, Amsterdam, pp. 143–210.
- Eberhart SA (1971) Regional maize diallels with U.S. and semi-exotic varieties. *Crop Science*, 11: 911-914.
- Horsley TN, Zhou MM (2014) Potential gains from introgression breeding based on analysis of three breeding populations. *Proceedings of the South African Sugar Technologies Association*, 87: 438 - 446
- Jackson P, Hale AL, Bonnett G, Lakshamanan P (2014) Sugarcane. In: Pratap A and Kumar J, (Eds) *Alien Gene Transfer in Crop Plants, Volume 2. Achievements and Impacts*. New York: Springer. pp. 317-345.
- Kennedy AJ (2001) Genetic base-broadening in the West Indies sugar cane breeding programme by the incorporation of wild species. In: Cooper HD, Spillane C and Hodgkin P (Eds) *Broadening the genetic base of crop production*. pp. 283-294.
- Nair NV (2012) Sugarcane genetic resources - status, potential and role in sugarcane improvement. *Journal of Sugarcane Research*, 2(2): 1-8.
- Walker DIT (1987) Manipulating the genetic base of sugarcane. *Proceedings of the Copersucar's International Sugarcane Breeding Workshop*. Sao Paulo, Brazil. pp. 321–334.