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RESEARCH ARTICLE

Field evaluation of genotypes and identification of traits for selection of nitrogen use efficient stable sugarcane (*Saccharum Spp.* hybrids) clones

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Abstract

Sugarcane has high nitrogen (N) requirement that normally exceeds the inherent capacity of soil to supply via mineralization. Moreover, long term N fertilization dose also contribute to artificial soil acidification, and eutrophication of surface and ground water. So, in order to identify nitrogen use efficient (NUE) genotypes and its contributing traits, 36 sugarcane genotypes (28 advanced lines + 8 cultivars) were analyzed for cane yield, CCS %, CCS t/ha and their contributing traits by growing them under N0 and N150 environments over two years at Faridkot, India. Significant differences were observed due to genotypes, nitrogen environments and their interactions with major effects on agro-morphological traits along with minor effects on quality traits. Inverse and counteracting relationships were observed between cane yield t/ha and CCS % along with their independent contributing traits. The results of the 'Eberhart and Russell Model' and 'Genotype + Genotype x Environment (GGE) Biplot' analysis were in accordance to each other; but Eberhart and Russell's model was more appropriate for judging the genotype(s) to environment specificity/adaptation while GGE Biplot was observed as the best approach to evaluate the environments for their discriminating power to genotypes as well as to find the most stable /ideal genotype(s) for a wide range of concerned environment and as well specific to a particular environment. The genotype namely F 194/10 was identified as most stable as well as NUE clone.

Keywords: GGE Biplot; NUE; Regressions; Stability; Sugarcane

Introduction

Nitrogen (N) is a key factor for sugarcane growth and development, and its requirement normally exceeds the inherent capacity of the soil to supply it by mineralization of organic matter. Nitrogen fertilizer increases germination, tillering, shoot formation and ultimately results in high cane and sugar yield. Sufficient N supply at a critical time period has positive relation with crop performances and sugar production (Anas et al. 2021). Moreover, long term N fertilization also contributes to artificial soil acidification especially in topsoil, and high N applications are undesirable from an immediate environmental perspective (nitrate leaching to ground water and eutrophication of surface waters). While, insufficient or improper supply of N-fertilizer to sugarcane causes abnormal growth with different kind of ill effects on environment and phenological development of plant along with increment of production cast by reducing crop performance and sugar production (Anas et al. 2021; Bell et al. 2014; Chen et al. 2014; Muchow et al. 1996).

Nitrogen use efficiency (NUE) is a kind of complex phenomenal trait that defines the plant ability to uptake nitrogen (N) for its growth and productivity. Two tier approaches (i.e. agronomical with different doses of different fertilizers at different stages of crop as well as genetical with inherited uptake efficiency under different levels of N input) are always worked for NUE. However, it has been well established that genetic potential with variability for NUE and interactions with N supply play major role (Robinson et al. 2014). The importance of NUE is quite clear in India because of continuous planting, which causes low output-input ratio i.e. only 0.3. Brazil, which accounts for 4 42 % of global sugarcane production, uses only 25 % of global N-fertilizer in sugarcane cropping system. Contrarily, India and China together produce 31 % of global sugarcane but apply 50 % of N-fertilizers as demonstrating that N-applications are high (Robinson et al. 2011). The NUE of sugarcane varies greatly, and high NUE varieties are an important issue for farmers as well as for sugarcane industries. Hence, the identification of NUE genotypes for rational N fertilizer application and methods to identify a variety with high NUE under low N conditions are of great significance for reducing N pollution.

The understanding of genetic basis of NUE is further complicated by interactions with climate, changing soil water and nutrient availabilities and plant developmental stages (Hirel et al. 2011). For this, plant breeder performs multi-environment trials (METs) to evaluate the genotypes across different environments (having different nitrogen availability status over locations and years) just before its release for commercial production. For such experiments, genotype × environment (GE) interaction is ideally carried out. Different statistical models are adopted to describe GE interaction that facilitates genotype ecommendation in METs (Yan et al. 2007; Singh and Bhajan 2016). This study aimed to identify key traits affected by N stress, effect of nitrogen on sucrose content and cane yield, identify the growth response and efficient genotypes to N limitation, and to quantify the G×E interaction effects on cane and sugar yield.

Materials and methods

Plant materials and crop culture

Fifteen sugarcane crosses with chosen parental stocks (Table 4) were attempted at National Hybridization Garden, ICAR-Sugarcane Breeding Institute, Coimbatore (Tropical region of India, Peninsular Zone, 11°00′58″N/76°58′16″E), India during October-November, 2009. The seedlings from the attempted crosses were raised at Punjab Agricultural University, Regional Research Station, Faridkot (South Western Zone, 30^0 40'

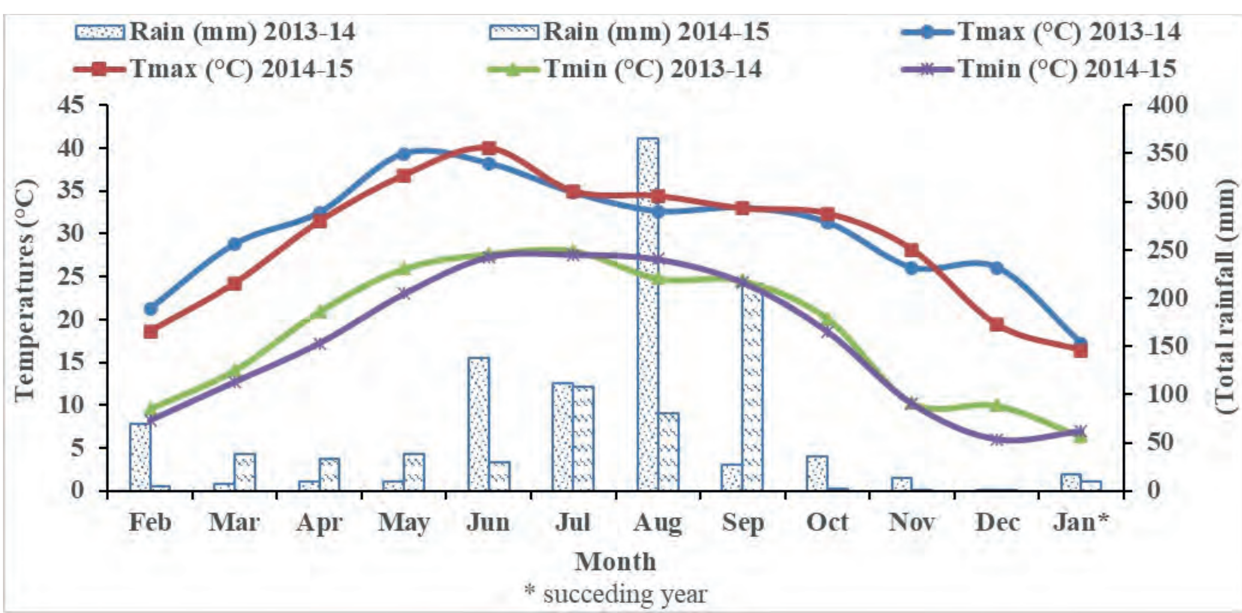

Figure 1. Monthly temperature and rainfall during 2013-14 to 2014-15

00"N/74^{0} 45' 00"E), Punjab, India during 2010-11. Clonal selections were performed based on growth and quality parameters to reduce population size at Stage I (2011-12) and Stage II (2012-13). During spring season, by following recommended agronomic practices (Anonymous 2017) under well irrigated condition, a total 36 genotypes (i.e. 8 popular varieties covering North-West Zone of India and 28 advanced clones, Table 4) were grown under N0 (i.e. no application of Nitrogen) and N150 (i.e. applied recommended dose of Nitrogen @ 150 kg/ha) environments for their evaluation to Nitrogen Use Efficiency (NUE) for two years i.e. 2013 and 2014. All the field experiments were laid out in a Randomized Block Design with two replications of two rows of five meters length at 75 cm row to row spacing.

Weather and soil environments

Weather data were recorded from Agrometeorology Observatory of PAU Regional Research Station. Weather during the years (2013 & 2014) was normal as experienced by North-West Zone Indian environment with annual variations between years (Fig. 1, Singh et al. 2019). Since, experiments were conducted under well management conditions; only differences in soil-nitrogen availability were the major consideration. Soil analyses of both environments were performed as suggested by Van Reeuwijk 2002. The soil status was normal (Table 1). The soil texture of experimental sites is sandy loam with slight alkaline pH, normal EC, high K and medium N & P content.

Agro-morphological and biochemical traits

Data on germination percentage (Gm %), tillers (000/ha) and number of millable canes (NMC 0000/ha) were recorded under field conditions. Data on cane length (cm), cane girth (cm), single cane weight (kg), cane yield (t/ha), extraction per cent (Extn %), total soluble solids (Brix %), sucrose % (Pol %), Purity %, commercial cane sugar per cent (CCS %) and commercial cane sugar tonnage per hectare (CCS t/ha) were recorded at crop harvest. Agro-morphological trait data were taken on plot basis and converted into per hectare basis; while biochemical trait's data i.e. Brix %, Pol %, Purity % and CCS % including cane yield attributes i.e. cane length (CL), cane girth (CG) and single cane weight (SCW) were recorded from randomly selected competitive five millable canes from each plot of each replication. For cane juice analyses, "Biquartz Sodium Lamp Polarimeter" was used and standard protocols were followed (Meade and Chen 1977, Gupta 1977).

Statistical analyses

The experimental design was RBD (randomized block design) in 2013 (Experiment 1) and 2014 (Experiment 2) for the nitrogen (N0: No nitrogen applied; N150: Recommended dose applied) response studies. There were two replications in both years. Analysis of variance was performed on genotypes using the PROC MIXED procedure in SAS for all traits. The probability threshold level was kept 0.05. The mixed effect model was followed. Genotypes were treated as a fixed effect, and replication nested within year were treated as a random effect. Genotype, replication, nitrogen doses and year were used as class variables. Separation of means was done using the LSD test

Table 1. The soil properties of experimental sites

Environments (Years)	**Soil texture**	**pH**	**EC** (m mhos cm^{-1})	**N** (kg ha^{-1})	**P** (kg ha^{-1})	**K** (kg ha^{-1})
2013-14	Sandy Loam	8.4	0.51	346	13.25	405
2014-15	Sandy Loam	8.5	0.50	340	13.46	400

Table 4: Regression coefficient, deviation from regression, and mean value of cane yield and sucrose % over the environments

S.No./ Code	Genotypes	Parentage	Cane Yield t/ha (CY)			Commercial Cane Sugar % (CCS %)			Commercial Cane Sugar t/ha (CCS t/ha)		
			m	S_d^2	i	m	S_d^2	β_i	m	S_d^2	β_i
1	F 3/10	Co 1158 X Co 62198	88.41	40.55	1.02*	10.01@	0.05	-2.01	8.87	-0.06	1.65*
2	F 20/10	Co 1158 X BO 91	113.02$	-52.14	0.12*	9.15	-0.01	1.41*	10.33@	-0.51	-0.16
3	F55/10	BO 91 X CoJ 46	111.38@	66.64	0.56	9.41	-0.04	2.92*	10.46@	0.99	0.03
4	F79/10	-do-	123.31	312.23*	0.80	8.76	0.04	-0.77	10.84	3.64*	1.05
5	F90/10	CoJ 83 X CoS 510	120.06#	-22.25	1.78*	9.59@	0.12	2.05	11.47#	0.67	1.73*
6	F115/10	Co 98008 X CoSe 92423	104.19@	47.24	0.50	9.22	-0.04	1.78*	9.63	0.21	0.18
7	F128/10	-do-	103.93	-42.63	0.62*	8.80	0.02	1.22	9.14	-0.22	0.45
8	F130/10	-do-	112.65#	76.72	2.02*	7.63	0.27*	-0.27	8.58	-0.22	2.00*
9	F133/10	-do-	103.06	-21.36	0.78*	10.60	0.16*	0.51	10.92@	0.34	0.99
10	F144/10	-do-	135.59#	-30.80	1.12*	8.92	-0.06	-0.43*	12.13#	-0.28	1.37*
11	F152/10	-do-	95.11	-43.88	0.16	8.43	0.07	1.82	8.01	-0.24	-0.19
12	F186/10	-do-	120.61@	3.54	0.56	7.64	0.04	-0.002	9.23	0.01	0.52
13	F194/10	Co 98010 X CoSe 92423	114.51$	-25.55	0.81*	9.56@	0.005	-0.41	10.94#	-0.22	1.09*
14	F210/10	-do-	106.45$	-11.77	0.91*	9.66	0.13*	-0.73	10.28#	-0.52	1.24*
15	F221/10	-do-	98.33	-36.32	0.74*	8.72	-0.06	2.16*	8.54	-0.43	0.37
16	F268/10	Co 98008 X Co 1148	91.79	-43.32	0.97*	9.55#	-0.05	2.29*	8.71	-0.42	0.76*
17	F316/10	Co 98010 X CoS 8436	99.25	-49.82	1.34*	9.39	0.45*	1.57	9.27	0.27	1.22*
18	F335/10	MS 6847 X CoV 92102	87.59	-13.18	1.12*	9.97@	-0.04	0.50	8.73	-0.10	1.31*
19	F390/10	Co 86032 X CoSe 92423	97.63	33.22	1.85*	10.17#	-0.001	1.52*	9.88#	-0.03	2.09*
20	F490/10	CoSe 95422 PC	96.42	276.15*	0.18	9.90	0.65*	2.93	9.46	0.35	-0.28
21	F519/10	CoSe 92423 PC	78.44	77.47	0.71	9.68@	-0.03	0.96	7.59	0.55	0.71
22	F 527/10	-do-	129.42#	22.26	2.19*	7.87	-0.05	-3.73*	10.34#	0.05	3.15*
23	F562/10	CoJ 86 GC	118.34#	-6.77	2.31*	9.46	-0.04	1.45*	11.15#	-0.20	2.44*
24	F568/10	-do-	111.37#	-30.35	1.65*	9.39	0.08	-0.16	10.45#	-0.51	1.95*
25	F674/10	CoH 119 GC	96.09	-16.64	0.89*	8.72	-0.02	-0.44	8.38	-0.41	1.07*
26	F711/10	CoH 70 GC	100.29	-32.88	0.31	8.66	0.03	0.66	8.69	-0.15	0.17
27	F714/10	-do-	119.03$	21.53	0.97*	8.76	0.17*	2.76	10.36@	0.11	0.40
28	F751/10	-do-	114.17#	8.30	1.07*	9.69#	0.05	2.34*	11.02@	0.26	0.73
29	CoJ 64	Co 976 X Co 617	85.28	70.73	0.76	11.44#	-0.01	1.99*	9.74	1.21*	0.76
30	CoJ 88	CoJ 82315 X Co 1148	109.07#	-42.91	1.04*	11.22@	0.08	-0.30	12.23#	-0.48	1.53*
31	CoPant 84211	Co 9806 X Co 6812	94.69	-46.26	0.41*	11.07	0.13*	1.31	10.49@	-0.03	0.30
32	CoJ85	Q 63 X CoJ 70	81.26	58.91	0.88	10.85#	0.08	2.37*	8.76	0.23	0.85
33	CoJ 767	Co 419 X Co 313	94.39	181.36*	1.04	10.17#	0.03	2.84*	9.56	2.66*	0.78
34	CoJ 89	LG 72115 X CoJ 82315	91.90	-7.36	0.94*	10.00#	-0.04	2.72*	9.15	-0.04	0.68
35	CoH119	Co 7704 GC	112.68#	61.54	1.69*	10.20#	-0.001	2.16*	11.44#	0.82	1.70*
36	CoS 8436	MS 6847 X Co 1148	88.90	-35.23	1.19*	10.21@	0.04	1.04	9.06	-0.39	1.37*

*Significant at 0.05 probability level; +Significantly deviate from unity; @, # and $ Average, High and Low responsive genotypes to soil nitrogen availability, respectively, with high mean value for CY, CCS% and CCS t/ha; β_i Regression Coefficient, mGeneral Mean for concerned traits, (P, 0.05). Pearson's correlation coefficient and simple regression analyses of concerned traits environment wise separately for N0 and N150 were done to find out best selection index for cane yield and sucrose content. In the present study; correlation coefficient (Pearson 1920), regression coefficient (for stability, Eberhart & Russell 1966) and genotype plus GE interaction biplot (GGE) (Zoble et al. 1988; Yan et al. 2007) analyses were performed. Pearson correlation procedure in "SPSS Statistics 22.0" (IBM Corp. 2013) was used to find out the correlation coefficients among all thirteen traits. Simple regression procedure in MS Excel 2007 was used to regress Cane Yield (CY) on NMC and SCW; CCS % on Brix %, Pol % and Purity %; CCS t/ha on CY and CCS %. For genotype × environment interaction analyses, a total four environments were considered here by two fertility levels i.e. N0 & N150 over two years i.e. 2013-14 & 2014-15. Eberhart & Russell's regression coefficient analysis using "SPAR 2.0" (IASRI 2012) and GGE Biplot analyses using "PBTools 1.4" (http://bbi.irri.org/products) were carried out for the trait CY t/ha, CCS % and CCS t/ha.

Results and discussion

All the soil and aerial environments were normal as per the North-West Zone (30^0 40' 00"N/74^0 45' 00"E) of India (Singh et al. 2019) during the years 2013-14 & 2014-15 (Table 1, Fig. 1). However, due to changes in Soil-Nitrogen application, the plant establishment and cane yield was affected too much with minor effects on quality traits (Table 2). The Gm %, tiller count (000/ha), NMC (000/ha), CY (t/ha), CL (cm), CG (cm), SCW (kg), Extn % and CCS t/ha was increased by 5.90 %, 9.55 %, 11.87 %, 17.86 %, 7.30 %, 4.37 %, 10.24 %, 3.04 % and 15.25 % under N150 environment in comparison to N0 environment, respectively. While the biochemical trait expression were reduced under N150 environment over N0 environment i.e. -0.07%, -2.30%, -2.15% and -3.39% for traits Brix %, Pol %, Purity % and CCS %, respectively. The traits NMC, SCW, CY and CCS t/ha were highly affected i.e. > 10% deviations; while biochemical traits were comparatively less affected due to differences in N fertility level. In the opinion of authors, this yield reduction was not so pronounced as in other crops; it might be that sugarcane utilizes the entire resource as it is a long duration crop. However, Han et al. (2015) pointed the significance of both Nitrogen Uptake Efficiency (NUpE) and Nitrogen Utilization/ Usage Efficiency (NUtE) for Nitrogen Use Efficiency (NUE) in crop plants. NUE varies according to the genotypes being evaluated, the amount of nitrogen administered, and how the relevant features are quantified or generated. It is undoubtedly noteworthy that the hereditary and environmental characteristics play key role in addition to the comprehend interplay between absorption and application of nutrients.

In variance analyses for all concerned traits (Table 2), significant ($p \leq 0.05$) differences were observed due to genotypes, nitrogen doses i.e. N150 and N0 and their interactions over both the years 2013-14 and 2014-15. Significance ($p \leq 0.05$) differences due to varieties and N environments interaction for concerned traits justify the further statistical analyses *viz*., stability and GGE biplot. Similar error MS values were observed for both ANOVA i.e. "ANOVA having separate year & nitrogen effect" and "ANOVA having combined year & nitrogen effects". A wide range of variations were observed among the genotypes for all concerned traits under testing which is being justified by the MS, mean and range value (Table 2).

Table 2. Analyses of variance (mean squares) results for various agro-morphological and biochemical traits in sugarcane

Source of variations	df	Gm %	Tillers	NMC	CY	CL	CD	SCW	EXTN%	Brix%	Pol%	Purity %	CCS %	CCS t/ha
On effects of years (Y), replications (I), nitrogen doses (N), varieties (V) and their respective interactions														
Years	1	11532.62*	22944.92*	1091.19*	37.96	2300.68*	0.01	0.60*	1301.14*	0.42	0.07	5.87	0.01	0.21
Replication	1	26.53	277.80	927.69*	1.22	61.42	0.01	0.01	31.03	0.85	0.01	33.10*	0.12	0.26
Nitrogen	1	503.35*	19360.14*	12152.6*	30038.97*	28560.5*	1.15*	1.03*	168.99*	0.01	7.09*	251.17*	7.26*	189.62*
Y*I	1	236.06*	426.52*	324.65	237.34	1.39	0.01	0.01*	18.98	0.16	0.21	27.18	0.35	0.72
Varieties	35	123.34*	4639.51*	738.58*	1511.44*	2737.11*	0.13*	0.07*	58.26*	15.46*	12.78*	68.21*	6.85*	10.77*
Y*V	35	125.19*	3522.89*	640.64*	260.78*	1629.85*	0.12*	0.06*	34.54	0.54*	0.34*	22.25*	0.33*	2.37*
N*V	35	43.88*	478.91	126.01	252.32*	327.04*	0.02*	0.01*	37.11	0.63*	0.50*	28.31*	0.46*	3.11*
Y*N	1	190.25*	4586.4*	730.94*	23.71	1667.53*	0.26*	0.03*	0.49	0.09	0.01	1.8	0.04	0.02
N*R	1	60.14	440.7	36.92	2.49	105.13	0.01	0.01	56.74	0.62	0.01	20.72	0.08	0.21
Y*N*V	35	21.04	710.7*	78.58	48.21	362.03*	0.03*	0.02*	38.87	0.28	0.19	9.98	0.17	0.74
Error	141	19.07	306.51	100.93	107.14	52.72	0.01	0.01	28.88	0.33	0.16	8.26	0.12	1.12
On effects of varieties, environments (four environments with two years i.e. 2014 & 2015 and two nitrogen doses i.e. N0 & N15 0) and their interactions														
Varieties (V)	35	123.34*	4639.51*	738.58*	1511.44*	2737.11*	0.13*	0.07*	58.26*	15.46*	12.78*	68.21*	6.85*	10.77*
EnvironmentI	3	4075.40*	15630.48*	4658.24*	10033.55*	10842.90*	0.47*	0.55*	490.21*	0.17	2.39*	86.28*	2.44*	63.28*
V×E	105	63.37*	1570.83*	281.74*	187.10*	772.98*	0.06*	0.03*	36.84	0.48*	0.34*	20.18*	0.32*	2.07*
Error	144	20.92	308.07	107.78	106.58	52.79	0.01	0.01	29.02	0.33	0.15	8.65	0.12	1.11
Summary of statistics														
R-Square		0.90	0.90	0.83	0.87	0.97	0.95	0.96	0.65	0.93	0.96	0.81	0.94	0.83
CV %		10.05	10.71	9.76	9.94	2.93	2.64	4.30	10.82	3.63	2.85	3.27	3.61	10.78
Root MSE		4.37	17.51	10.05	10.35	7.26	0.07	0.05	5.37	0.57	0.39	2.87	0.34	1.06
Mean		43.46	163.50	102.98	104.13	247.41	2.55	1.13	49.68	15.71	13.80	87.94	9.51	9.83
Range		32.62-51.59	120.62-231.87	78.76-121.51	78.44-135.59	205.25-276.81	2.36-2.87	1.01-1.33	42.63-55.88	13.11-18.88	11.21-16.37	79.84-92.36	7.63-11.44	7.59-12.23
Per cent deviations under N150 Environment over No Environment		5.90	9.55	11.87	17.86	7.30	4.37	10.24	3.04	-0.07	-2.30	-2.15	-3.39	15.25

Correlation and regressions coefficients among the traits

Under both the environments i.e. N150 and N0, cane yield was observed to be significantly positively ($p \leq 0.01$, $r > 0.50$) correlated with Gm %, tillers and NMC while it was negatively ($p \leq 0.01$, $r > -0.20$) correlated with quality traits i.e. Brix %, Pol %, purity % and CCS %. Under N0 environment in contrast to N150 environment, cane yield, there was negative correlation with SCW and Extn %; although this association was observed non-significant (Table 3). In sugarcane, cane yield (t/ha) is decided by NMC and SCW; while these both traits are directly linked to a number of other traits i.e. Gm %, tiller count, shoot count, CL and CG. In our present study, NMC was observed to be main yield contributing trait with $R^2 \geq 0.50$ while role of SCW was not so pronounced especially under low nitrogen environment (Fig. 2). CCS t/ha was observed to be positively ($p \leq 0.01$, $r > 0.35$) correlated with its quality contributing traits i.e. Brix %, Pol % and CCS % while negative (r from -0.10 to -0.60) association was observed with other cane yield (t/ha) contributing traits under both environments. The main quality traits deciding the CCS% are Brix %, Pol % and purity % i.e. all having major role on CCS % with R^2 value ≥0.60, ≥0.95 and ≥0.12, respectively (Fig. 3). However, pol % was observed to be the main trait determining the CCS % under both environments. CCS t/ha was also observed having positive association with its contributing traits i.e. cane yield t/ha ($p \leq 0.01$, $r > 0.70$, $R^2 \geq 0.45$) and CCS% ($r > 0.28$, $R^2 \geq 0.08$) under both environments (Table 2, Fig. 4). Here in this study, cane yield (t/ha) was observed to be more pronounced trait having comparatively more role in determining CCS t/ha compared to CCS% under both the environments. The quality traits have been observed to have high correlation coefficient value and strong correlation with its contributing traits under N0 environment than N150 environment. Such kind of trend was not observed for cane yield and its contributing traits (Table 3; Fig. 2, 3 & 4). For quality traits, it may be the reason that enough/ sufficient N generally leads to increased moisture in stalks and consequently increased amounts of the reducing sugars (glucose and fructose) in juice in place of non-reducing sugars (especially sucrose) (Das 1936; Muchow and Robertson 1994).

Eberhart & Russell regression coefficient analyses

Based on Eberhart and Russell's regression coefficient parameters (1966), all the 36 sugarcane genotypes were evaluated for cane yield t/ha, CCS % and CCS t/ha (Table 4). The significant ($p \leq 0.01$) value of environment (linear) components and G × E linear component were observed. So, the observed higher magnitude of environmental (linear) effect in comparison to G × E (linear) suggested that lower magnitude of environmental interaction effect might be responsible for high adaptation of the genotypes in relation to quality and cane yield. Pooled deviations differed significantly. Sixteen sugarcane genotypes including checks out of 36 were observed stable, deviated non-significantly from zero (S~0) for cane yield along with high per se mean performance (>104.13 t/ha). Among these genotypes, three average responsive (β_i~1) genotypes i.e. F 55/10, F 115/10, F 186/10, and three low responsive (β_i <1) genotypes i.e. F 20/10, F 194/10, F 210/10 to soil nitrogen level were identified. Nine genotypes namely F 90/10, F 130/10, F 144/10, F 527/10, F 562/10, F 568/10, F 751/10, CoJ 88 and CoH 119 were highly responsive (β_i > 1) to soil nitrogen level. For CCS %, seven average responsive (β_i~1) genotypes i.e. F 3/10, F 90/10, F 194/10, F335/10, F 519/10, CoJ

Table 3: Pearson Correlation Coefficient among Agro-morphological and biochemical traits in sugarcane under N 0 (upper diagonal) and N 150 (lower diagonal) environments.

Particulars	Gm	Tiller	NMC	CY	CL	CD	SCW	Extn %	Brix %	Pol %	Purity %	CCS %	CCST
Gm	1	0.53**	0.55**	0.65**	0.11	0.29	0.02	-0.18	-0.20	-0.26	-0.23	-0.28	0.49**
Tiller	0.36*	1	0.59**	0.52**	0.08	0.06	-0.15	0.01	-0.12	-0.17	-0.16	-0.18	0.43**
NMC	0.49**	0.66**	1	0.75**	-0.12	-0.17	-0.38*	-0.24	-0.04	-0.18	-0.40*	-0.23	0.63**
CY	0.54**	0.59**	0.70**	1	0.29	0.15	-0.01	-0.35*	-0.33*	-0.48**	-0.46**	-0.52**	0.67**
CL	0.40*	0.11	0.13	0.25	1	0.01	0.51**	-0.06	-0.56**	-0.60**	-0.23	-0.59**	-0.17
CD	0.06	-0.14	-0.36*	0.06	0.16	1	0.48**	0.12	-0.19	-0.16	0.01	-0.15	0.03
SCW	-0.01	-0.03	-0.16	0.21	0.41*	0.58**	1	0.12	-0.41*	-0.34*	0.08	-0.30	-0.28
Extn %	0.05	-0.07	-0.19	0.08	0.43**	0.23	0.33*	1	0.09	0.19	0.29	0.23	-0.21
Brix %	-0.36*	-0.02	-0.12	-0.23	-0.41*	-0.08	-0.22	-0.37*	1	0.93**	0.03	0.87**	0.36*
Pol %	-0.34*	-0.07	-0.30	-0.35*	-0.43**	-0.06	-0.12	-0.32	0.89**	1	0.40*	0.99**	0.31
Purity %	0.08	-0.11	-0.37*	-0.21	0.01	0.06	0.23	0.16	-0.32	0.14	1	0.53**	-0.04
CCS %	-0.30	-0.09	-0.37*	-0.37*	-0.41*	-0.04	-0.07	-0.27	0.77**	0.98**	0.35*	1	0.28
CCST	0.35*	0.53**	0.48**	0.78**	0.01	0.04	0.18	-0.08	0.27	0.30	0.03	0.28	1

*. Correlation is significant at the 0.05 level (2-tailed), **. Correlation is significant at the 0.01 level (2-tailed).

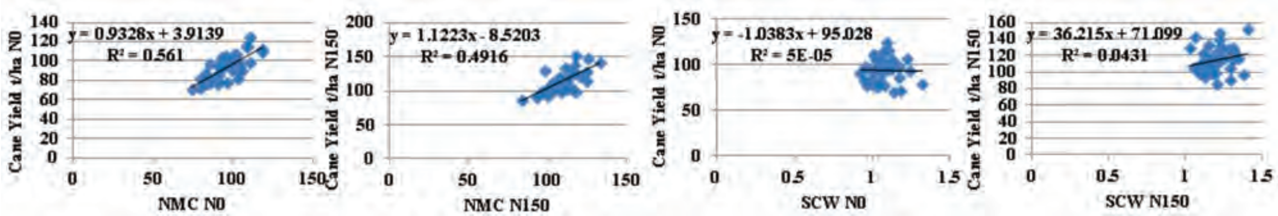

Figure 2 Relationship of caneyield with NMC and SCW under N0 and N150 environment within the set of 36 sugarcane genotypes.

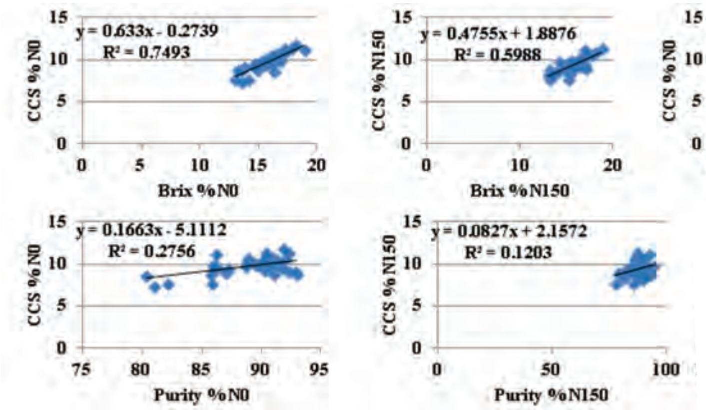

Figure 3 Relationship of CCS% with Brix %, Pol% and Purity % under N0 and N150 environment within the set of 36 sugarcane genotypes.

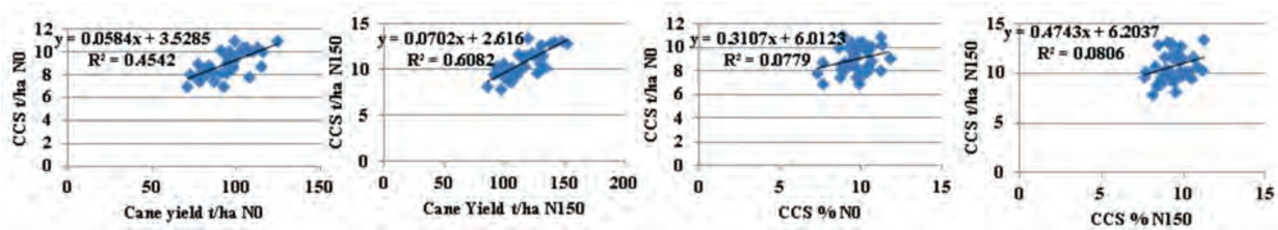

Figure 4 Relationship of CCS t/ha with cane yield and CCS% under N0 and N150 environment within the set of 36 sugarcane genotypes

88, CoS 8436, and eight high responsive ($\beta_i > 1$) genotypes i.e. F 268/10, F 390/10, F 751/10, CoJ 64, CoJ 85, CoS 767, CoJ 89, CoH 119 were identified with high per se stable performance (>9.51 CCS%, S~0). For the trait CCS t/ha, six average responsive ($\beta_i \sim 1$) genotypes i.e. F 20/10, F 55/10, F 133/10, F 714/10, F 751/10, CoPant 84211, and ten high responsive ($\beta_i > 1$) genotypes i.e. F 90/10, F 144/10, F 194/10, F210/10, F 390/10, F 527/10, F 562/10, F 568/10, CoJ 88, CoH 119 were identified. However based on above studies, four genotypes i.e. F 90/10, F 751/10, CoJ 88, CoH 119 were identified as nitrogen responsive genotypes because of high/& average responsiveness and high mean value for the traits cane yield t/ha, CCS % and CCS t/ha to soil available N environment. Only one genotype i.e. F 194/10 was identified to be suitable for low soil available nitrogen environment because of its low/& average responsiveness and high mean performances for the traits namely cane and sugar yield to soil available nitrogen environment.

GGE Biplot Analyses

Three major aspects i.e. mega environment analyses, test environment evaluation and genotype evaluation (Yan and Kang 2003) were studied under genotype by environment data (GED) analyses for three major traits (cane yield t/ha, CCS% and CCS t/ha) of sugarcane. The genotype main effect (G) + genotype bye environment (GE) interaction (GGE) biplots were constructed by plotting the first principal component (PC1) scores of genotypes and environments against their respective scores for the second principal component (PC2) that result from the singular value decomposition (SVD) of environment centered GED (Yan et al. 2007).

Based on mean value of each genotypes in each environments over replications; the four environments fell into two sectors (two mega environments) i.e. one with ENV1 & ENV2 (N 0 environment) and another with ENV3 & ENV4 (N 150 environment) for all three concerned traits i.e. cane yield t/ha with six and ten genotypes, CCS % with seven and eleven genotypes, CCS t/ha with seven and five genotypes, respectively, as winning genotypes in the 'what won where' view (Fig 5A, 5B, 5C) of mega-environment analysis (Yan et al. 2000). This indicated that these genotype(s) had the highest cane yield &/or sugar yield potential in that concerned environments. Specifically, the clones namely F 527/10 (22) under N0 & F 144/10 (10) under N150 for the traits cane yield t/ha, CoJ 88 (30) under N0 & CoJ 64 (29) under N150 for CCS %, and F 144/10, CoJ 88 (10, 30) under N0 & F 55/10 (3) under N150 for CCS t/ha were the better performing genotypes than others. Crossover GE suggests that the target environments i.e. ENV1, ENV2, ENV3 & ENV4 may be grouped into two mega-environments as N0 & N150 with no significant differences over years (Yan et al. 2007). The corner genotypes (most responsive to soil nitrogen availability) can be visually determined as most favourable & lowest yielding for the traits cane yield (10 genotypes, Fig 5A); CCS % (10 genotypes, Fig 5B); CCS t/ha (10 genotypes, Fig 5C).

The assessment of genotype becomes meaningful after identifying the mega environment. The ideal genotype should have both high stability as well as high mean performance. Fig 6A, 6B, 6C are the average environment coordination (AEC) view of the GGE biplots with four environments in the niche of genotypes namely 10 (F 144/10), 29 (CoJ 64), 10 & 30 (F 144/10 & F CoJ 88) as identified in Fig 5A, 5B, 5C, respectively. The arrow lines on AEC axis represent the mean value of genotypes; consequently the ranking of genotypes are decided based on the mean performances; while the level of projection from axis represent the degree of stability of the genotypes. This AEC view facilitated the genotype comparisons based on mean performance and stability across environments within the mega-environment (Yan 2002). The genotypes were ranked according to genotype main effect (G, i.e. proportional to the rank two approximations of the genotype means; Yan, 2002). Here, since GGE represents G + GE while AEC abscissa approximates the genotype's contributions to G. So, AEC ordinate must approximate the genotype's contributions to GE, which is a measure of their stability (Yan 2001, Yan 2002). Thus, F 144/10 (10) for cane yield, CoJ 64 (29) for CCS% and F 144/10 (10) & CoJ 88 (30) for CCS t/ha were identified as ideal genotypes (While other genotypes with above mean performances i.e. F 90/10 (5), F 714/10 (27), F 194/10 (13), F 751/10 (28), CoJ 88 (30) for cane yield, CoPant 84211 (31), CoS 8436 (36), F 390/10 (19), F 335/10 (18), F 519/10 (21), F 90/10 (5) for CCS%, F 194/10 (13), F 133/10 (9), F 751/10 (28), F 210/10

(14) for CCS t/ha were more stable and located almost on the AEC abscissa and had a near zero projection onto the AEC ordinate; which is in accordance with Yan et al. 2010.

Fig. 7 and Fig. 6 are the same, except that Fig. 7 is based on environment focused scaling. This is the singular value, and entirely partitioned into the environmental scores. That is why, it's appropriate for studying the relationships among environments. This kind of the AEC is referred as 'Discriminating Power vs. Representativeness'

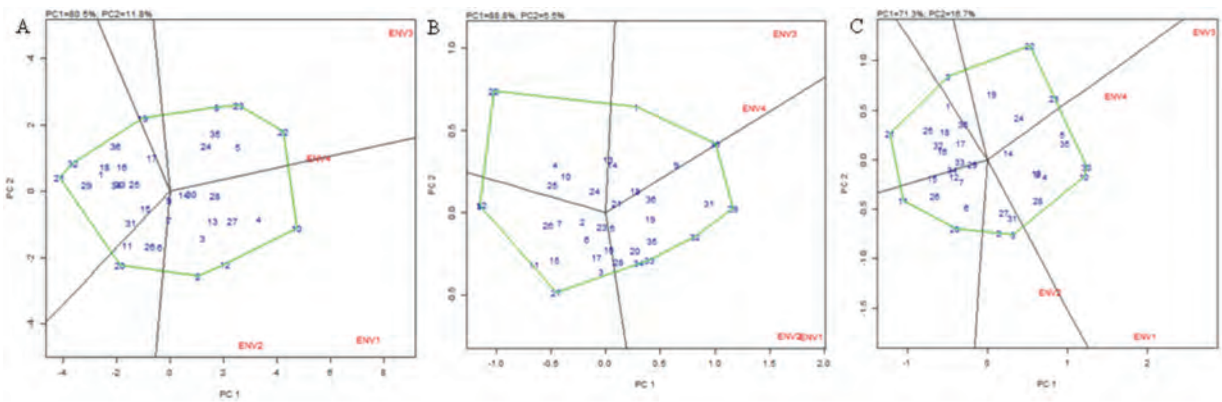

Figure 5: "Which-Won-Where" view of GE Biplot for Cane yielcl t/l1a (A), CCS % (B) mu\ CCS t/l1a (C) in Sugarcane

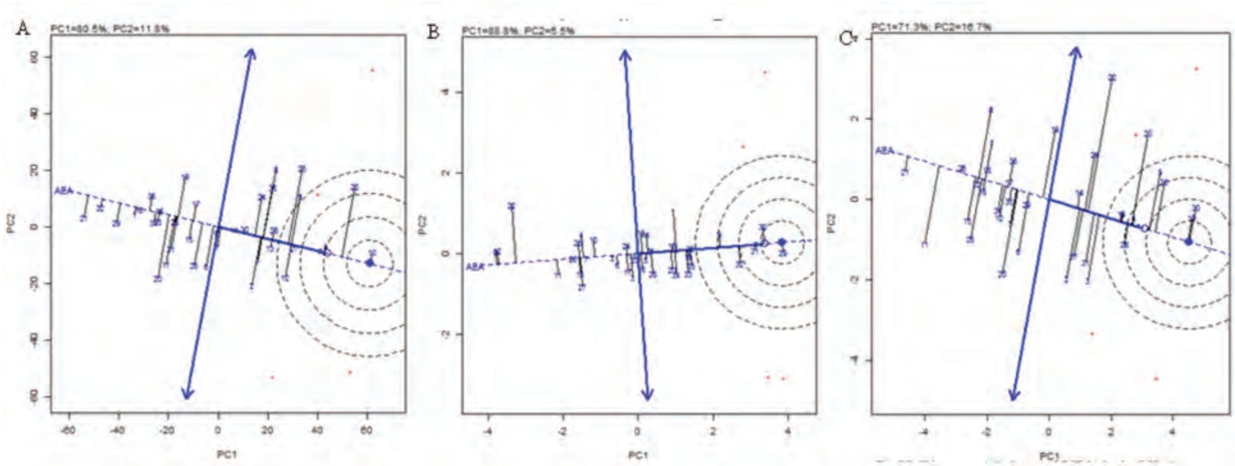

Figure 6: "Mean vs. Stability" view of GE Biplot for Caneyield tha (A), CCS % (B) and CCS tha (C) in Sugarcane

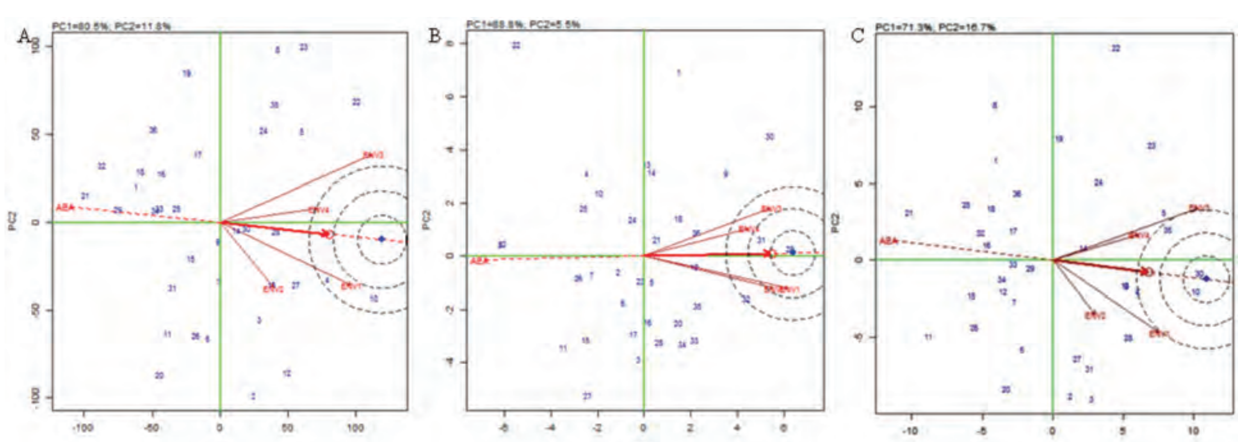

Figure 7 : "Discriminating Power vs Representativeness" view of GE Biplot for Cane Yield t/ha (A), CCS % (B) and CCS t/ha © in Sugarcane

view of 'GGE biplot'. Among four environments, two environment i.e. ENV1 & ENV2 (N0 environment) for cane yield, CCS % and CCS t/ha was observed to be more representative of mega environment with high discriminating power for genotypes. Because these environment(s) are having long vectors (i.e. vector length is proportional to standard deviations of genotype mean in the environment, which is the measure of discriminating power of environment with assuming comparable test environment experiment errors) and small angles (i.e. more representative of mega environment because angle cosine between any environment vector and average environment axis approximates the correlation coefficient between the genotype values in that environment and the genotype means across the environments) with the AEC abscissa which is useful for selecting superior genotypes with stable performances as by Yan et al. 2007.

Conclusion

Variability with respect to cane yield (t/ha) between N0 and N150 environments was linear and, NUE of genotypes could be judged on the basis of relative magnitude of variances. The genotypes had varying level of NUE over the different N fertility levels for cane yield; while for biochemical traits especially to Brix %; it was also observed significant but less as compared to agro-morphological traits. Among cane and sugar yield contributing traits under both N0 as well as N150 environment, NMC for CY, Pol % for CCS % and cane yield t/ha for CCS t/ha were observed to have major role in their expression, respectively. However under sufficient soil N available environment, comparatively major role of SCW was observed along with NMC in determining CY. Cane yield and its contributing traits were negatively correlated to sucrose content and its contributing traits. So based on the correlation & regression coefficient, these traits i.e. NMC, Brix %, Pol % and CCS % could be considered in formulating the counteracting selection index for development of superior genotypes having higher cane as well as sugar yield under different levels of soil Nitrogen availability. Further, NMC and Pol % (under N0 & N150) along with SCW (under N0) together among independent traits could be considered as major cane and sugar yield contributing traits during formulation of selection index. The genotypes F 194/10 (10) for cane yield and CCS t/ha, CoJ 64 (29) for CCS % and CoJ 88 (30) for CCS t/ha were identified as the ideal genotypes to available soil nitrogen environment. However for both cane as well as sugar yield trait, four NUE genotypes suitable to high soil available nitrogen environment i.e. F 90/10 (5), F 751/10 (28), CoJ 88 (30), CoH 119 (35), and only one NUE genotype suitable to low soil available nitrogen environment i.e. F 194/10 (10) were identified. It was observed that Eberhart & Russell's model was better suited for evaluating genotypic adaptation to particular environment as well as to a wide range of environments. However, the environments' ability to discriminate the genotypes could be effectively evaluated using GGE Biplot analysis.

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