

Evaluation of Tissue Culture Raised Sugarcane Planting Materials against their Donor Conventional Seed Sources as Initial Source of Seed Cane at Tendaho Sugar Development Project, North-Eastern Ethiopia

Mohammad Ibrahim^{1,2}, Belay Tolera^{1*}, Jamal Aman² and Tadesse Negi³

¹Ethiopian Sugar Corporation, Research and Training Division, Tendaho Research Station, Ethiopia

²Ethiopian Sugar Corporation, Research and Training Division, Variety Development Research Directorate, Biotechnology Research Team, Ethiopia

³Ethiopian Sugar Corporation, Research and Training Division, Agronomy and crop protection Research Team, Wonji Research Center, Wonji, Ethiopia

*Corresponding author: Belay Tolera, Ethiopian Sugar Corporation, Research and Training Division, Tendaho Research Station, Ethiopia, Tel: +251-910181644; Fax: +251-222200144; E-mail: belaytolera@yahoo.com

Received date: Nov 30, 2015; Accepted date: Dec 31, 2015; Published date: Jan 04, 2016

Copyright: © 2016 Ibrahim M, et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Abstract

Healthy seed cane supply is the lifeline in the productivity and profitability of sugarcane and hence the sustainability of sugar industry, the Ethiopian sugar estates/projects have been using tissue cultured sugarcane planting materials since 2011/12. However, there is no study that shows the practical advantage of tissue culture over the conventional seed sources at Ethiopian situations. Thus, the current work was carried out to evaluate the two seed sources of two sugarcane genotypes. Accordingly, analysis of variance proved that the interaction effects of genotype by seed source is highly significant ($p < 0.001$ at $\alpha = 5\%$) in all the response variables tested. Similarly, the two seed sources showed statistically significant differences throughout all the responses. The two sugarcane genotypes also showed marked variation in all the responses except the number of live buds per stalk, stalk height (cm) and number of two bud setts produced per stalk. In sugarcane genotype B52-298, the rate of propagation for tissue culture seed source is 1:44.68 against 1:13.72 of its donor from conventional seed source. In NCo-334, tissue culture seed source produced a propagation rate of 1:40.11 against 1:13.98 of its donor conventional seed source. Regardless of the other benefits, planting a hectare B52-298 and NCo-334 provided a direct net benefit of US\$ 5448 and US\$ 3999, respectively. Thus, the current result revealed that tissue culture seed source is a realistic and better alternative over the conventional seed source in sugarcane as initial seed cane at Tendaho Sugar Development Project. Evaluation of the two seed sources for all genotypes at each sugar estates/projects in successive three tier system of seed cane production and commercial stages could be the future line of work.

Introduction

The Ethiopia sugar industry plays a leading role in the socio-economy of the country. Besides its agricultural and industrial investments, foreign exchange earnings, high employment, linkages with major suppliers, support industries and customers; development of new sugar development projects provide access road, clean water, education and health facilities for the local communities. In addition, the country has great sugar production potentials and opportunities which include specifically identified irrigable suitable fertile land, favorable weather conditions, cheap and productive labour force, high demand for sugar and other by-products and huge market outlets to the nearby countries. To utilize these opportunities and satisfy the current national sugar shortage and export the surplus; the Ethiopian Sugar Corporation is undertaking large scale expansion and new sugar development projects in different regions of the country. Within the last four years (2011-2015) the sugarcane plantation area increased from 30,000 hectares to about 100,000 hectares and following the completion of all the expansion and first phase new development plan, the total cane area shall grow to 500,000 ha. As a result, the existing annual sugar production of 0.30 million tons shall be increased to 2.25 million tons with increase in other by products. Micropropagation based seed cane production is conducive to decrease spread of systemic diseases like RSD, leaf scald, chlorotic streak, mosaic virus, leaf gall and other diseases that spread through seed cane with subsequent

reduction in cost of disease control and increase in yield. In addition, tissue culture derived seed cane is superior in sprouting, growth, cane and sugar yield than their donor plants propagated by conventional method [1-7].

Healthy seed cane supply is the lifeline in the productivity and profitability of sugarcane and hence the sustainability of sugar industry. In line with this, except Omo kuraz sugar development project, all the eight Ethiopian sugar estates/projects are using micropropagated sugarcane plantlets within the three tier system of sugarcane seed cane production since 2011/12. Among the eight sugar estates/projects, Tendaho and Tana-Beles sugar development project are the leading ones that used tissue culture raised clean sugarcane planting materials over a large plantation area. Owing to its distant location and harsh environmental condition, transport of bulky conventional sugarcane planting materials and rapid desiccation and deterioration is among the major challenges in sugarcane plantation establishment at Tendaho. In addition, salt affected soils, poor soil and irrigation drainage, labor shortage with limited output, invasive prosopis weeds, sugarcane shoot and stalk borer are among the major challenges in cane and planting material. Thus, use of clean and healthy planting material to cover large area of land within the shortest possible time to supply adequate malleable cane for the 26,000 TCD factory of Tendaho bypassing all the challenges was a key target of the time.

To satisfy the short term planting material requirement, the Ethiopian Sugar Corporation made an agreement with Mekelle Technology Institute and Narus Biotechnology and Agro-Industry PLC to had total of 170 million primary acclimatized sugarcane planting materials of 14 different sugarcane genotypes among which about 33.5 million plantlets were delivered to different sugar estates of Ethiopia [8]. However, there is no research work that shows the yield magnitude or advantage of tissue culture derived seed source over the conventional propagated seed source in the Ethiopian sugar estates. Thus, the main aim of the current work is to compare tissue culture derived seed source with the conventional propagated seed source as a source of initial seed cane.

Materials and Methods

Description of the study site

Tendaho Sugar Development Project is one of the nine Ethiopian sugar estates/projects located at Afar Regional State with an elevation of the area laying between 400 m.a.s (around Tendaho) and 340 m.a.s (near Assayta). The average mean monthly maximum temperature varies between 32.3°C and 43.2°C with the mean annual rainfall of about 222 mm. The soils of the area were derived from different parent materials mainly from recent alluvium (near Assayta), lacustrine sediments or old alluvium (near Dubti), and young riverine alluvium (left and right bank of Awash River) and the major ones were fluvisols, vertisols, solonchak-solontez and regosols while the dominant soil type is fluvisols followed by vertisols [9].

Field survey procedures and response variables collected

Tissue culture raised sugarcane plantlets derived from apical meristem cultures of two selected genotypes of sugarcane namely: NCo-334 & B52-298 were delivered from Mekelle Technology Institute Tissue Culture Laboratory. Plantlets having intact coco-peat from primary green house acclimatization were planted directly to field without any secondary acclimatization using 25 cm spacing between plants and 145 cm spacing between furrows. Pre-planting furrow irrigation was give a day before planting to moisten the soil and cool down the harsh temperature (45°C air temperature and 68°C soil temperature) of the field.

Similarly, planting of the two genotypes (NCo-334 and B52-298) of sugarcane using conventional seed source was made using the estate recommended planting technique (5 cm overlapping) on the same soil type (fluvisols) on the same date. Field management for both seed sources was employed as per the recommendation that had been made for both sugarcane seed production of the estate. Data were collected from five sample plots with each 19.63 m² (2.5 radius) area. Sample plots were stratified diagonally at 24 meters interval leaving 10 meters to avoid border effect. The data were collected from randomly selected sample plants on number of tillers per hectare at three, four and five months, stalk population per hectare, number of live buds per stalk, cane height (cm), number of setts per stalk and number of setts produced per hectare. , number of plants established per hectare at two months, and rate propagation at ten months for both seed source genotypes were collected from five representative sample plots with 19.63 m² (2.5 radius) area. Data for survival percent (for tissue culture plantlets) and percent sprout (for conventional seed source) was collected at 45 days after planting from every 3rd and 5th row one after the other. Then, collected data were subjected to Analysis of variance using statistical analysis software SAS version 9.2. Mean separation was

made using the procedure of REGWQ Multiple Range Test at 5% probability level.

Result and Discussion

Analysis of variance revealed that the interaction effects of genotype by seed source has a very significant effect ($p < 0.001$) on planting material requirement per hectare, survival rate (for plantlets), percent sprout (for sett), number of plants established per hectare, number of tillers per hectare, stalk population per hectare, number of live buds per stalk, stalk height, number of two bud setts per stalk, number of two bud setts produced per hectare, and propagation rate (Table 1). The two sugarcane genotypes also showed marked variation for all the responses tested i.e. planting material requirement per hectare, survival rate (for plantlets), percent sprout (for sett), number of plants established per hectare, number of tillers per hectare, stalk population per hectare, number of two bud setts produced per hectare and propagation rate except number of live buds per stalk, stalk height and number of two bud setts per stalk (Table 2). Similarly, the two seed sources also showed statistically significant variation for all the tested responses (Table 3).

In sugarcane genotype B52-298, the two seed sources (conventional and tissue culture) showed a significant difference in all the tested response variables except number of live buds per stalk, stalk height and number of two bud setts per stalk (Table 4). Using B52-298 for planting a hectare of land requires 26,945 two bud setts (53890 buds) which cost about US\$ 300 while planting the same hectare of land through tissue cultured plantlets requires 27,600 plantlets that costs about US\$ 4140. In genotype NCo-334, planting a hectare of land requires 4250 buds (2125 two bud setts) which is significant less than the requirement for B52-298 owing to the longer internodes NCo-334 than that of B52-298. At Tendaho sugar development project, sugarcane genotype B52-298 produced 383067 two bud setts per hectare with propagation rate of 1:13.72 from conventional seed source while the same genotype produced 1060682 two bud setts with 1:44.68 propagation rates under similar environmental and agronomic management practice. Thus, planting a hectare of micropagated plantlets of B52-298 can produce planting materials for more 30.96 hectares as compared to the conventional seed source.

Generally, planting a hectare of land with micropagated sugarcane genotype of B52-298 at Tendaho sugar development project gave a direct net profit of US\$ 5448 without any opportunity cost like land saving, agronomic management practice (weeding, fertilization, irrigation, pesticides, labor, etc.,) with subsequent propagation rates and yield. In sugarcane genotype NCo-334, the conventional seed source produced 350649 two bud setts per hectare with 1:13.98 propagation rates against 852074.5 two bud setts with 1:40.11 rate of propagation of the micropagated seed source. Planting a hectare of NCo-334 derived from tissue culture seed source can produce planting material for 26.13 ha of land than the conventional source resulting in direct net profit of US\$ 3999. Even if the initial plant establishment for conventional seed source is better than that of the tissue culture seed sources in both genotypes, the larger number of tillers followed by more number of stalk population in tissue culture seed source resulted in production of more number two bud setts and hence the propagation rate (Table 4). From this result, it can be deduce that the use of tissue culture seed source at Tendaho sugar development project is by far more profitable than the conventional seed source. The current result is in agreement with the findings of [2,3,5-7] except the number of live buds per stalk and stalk height. Evaluation of the two

seed sources at successive three tier system of seed production and at production commercial level will be the future line of work.

Source of variations	DF	Mean Squares									
		Seed requirement (no of buds or plantlets/ha)	Survival / Germination rate (%)	No of Plants established/ha	Number of tillers/ha	Stalk population/ha	No of live buds per stalk	Height(cm)	Setts per stalk	No of setts/ha	Propagation rate
Genotype	1	129891***	207.36***	110390***	51840***	704955***	0.000225***	370.27***	0.000756***	0.667***	18.576***
Seed source	1	16960***	670.81***	6874621***	20070***	3768995***	0.004225***	347.54***	0.002756***	0.667***	3259.268***
Genotype * Seed source	1	12989***	47.61***	305532***	46240***	2626668***	0.013225***	370.27***	0.000156***	0.667***	23.328***
CV (%)	11		9.4	9.5	10.7	8.3	9.8	9.2	7.5	9.7	12

Table 1: ANOVA for comparison of tissue culture raised and conventional propagated seed sources of sugarcane genotypes. The symbol “***” indicates very highly significant difference.

Genotypes	REGWQ Grouping of Genotypes									
	Seed requirement (No. of buds or plantlets/ha)	Survival / Germination rate (%)	No. of Plants established/ha	Number of tillers/ha	Stalk population/ha	No. of live buds/stalk	Height(cm)	No. of Two bud Setts/ stalk	No. of two bud setts/ha	Propagation rate (ha)
B52-298	40745a	93a	33882a	141000a	111759a	12.5a	175a	7a	721875a	29.20a
NCo-334	35047b	85.8b	32221b	105000b	98483b	12.4a	174a	7a	601362b	27.05b

Table 2: Comparison of sugarcane genotypes based on REGWQ Grouping.

Seed source	REGWQ Grouping of Genotypes									
	Seed requirement (No. of buds or plantlets/ha)	Survival / Germination rate (%)	No. of Plants established/ha	Number of tillers/ha	Stalk population/ha	No. of live buds/stalk	Height(cm)	No. of Two bud Setts/ stalk	No. of two bud setts/ha	Propagation rate (ha)
Tissue culture Raised	48192a	95.9a	39607a	235000a	153656a	13.5a	199a	6.75a	956378a	42.40a
Conventional propagation	27600b	82.9b	26497b	11000b	56586b	12.2b	176b	6.25b	366858b	13.85b

Table 3: Comparison of Tissue culture raised and conventional propagated seed sources based on REGWQ Grouping.

Genotype	Seed source	Mean ± SE									
		Seed requirement (No. of buds or plantlets/ha)	Survival / Germination rate (%)	No. of Plants established/ha	Number of tillers/ha	Stalk population/ha	No. of live buds/stalk	Stalk Height (cm)	No. of Two bud Setts/ stalk	No. of two bud setts/ha	Propagation rate (ha)
B52-298	PC	53890 ± 1.73a	77.6 ± 0.18c	41819 ± 0.32a	10000 ± 1.25d	59172 ± 0.20c	12.95 ± 0.5a	1.91 ± 1.8a	12.95 ± 0.55a	383067 ± 1.92c	13.72 ± 2.4c
	TC	27600 ± 1.73c	94 ± 0.18b	25946 ± 0.32d	200000 ± 1.25b	164345 ± 0.20a	12.93 ± 0.5a	1.90 ± 1.8a	12.93 ± 0.55a	1060682 ± 1.92a	44.68 ± 2.4a

NCo-334	PC	4249 1.73b	3±	88.25 ± 0.18c	37394 ± 0.32b	12000 1.25c	±	54000 ± 0.20d	12.9 ± 0.5a	1.95 1.8a	±	12.94 ± 0.55a	350649 ± 1.92d	±	13.98± 2.4c
	TC	27600 ± 1.73c		97.75± 0.18a	27048 ± 0.32c	270000 1.25a	±	142966 ± 0.20b	12.99 0.5a	±	1.9 1.8a	7±	12.99 ± 0.55a	852074.5 ± 1.92b	40.11± 2.4b

Table 4: Comparisons of sugarcane genotypes for different seed sources. “PC” stands for conventional propagation seed source while “TC” indicates tissue culture raised seed source.

Conclusion

Unlike the costly procurement cost of initial planting material, sugarcane planting materials derived from tissue culture technology produced significant number of tillers and stalk population per hectare resulting in a very high propagation rate (1:42.4). Regardless of the other opportunity benefits, the current result clearly showed that the use of tissue cultured seed source is by far more profitable than using the conventional seed source in terms of the rate of propagation. Thus, in the multitude challenges of sugarcane plantation establishment of Tendaho sugar development project, use of tissue cultured sugarcane planting material is a realistic and best alternative than the conventional seed source.

Acknowledgments

We would like to thank the committed staffs of Tendaho Sugar Development Project for their allocation of resources and our gentle car driver Ato Yared Midheksa for his safe driving at the harsh environmental conditions of the project site.

References

1. Anonymous (2002) Micropropagation: Tissue culture techniques in sugarcane. Indian Institute of sugarcane Research. Directorate of Sugarcane Development.
2. Comstock JC, Miller JD (2004) Yield comparison: Disease free tissue cultures versus bud propagated planted sugarcane plants and healthy Versus yellow leaf virus infected plants. J Amer Soc Sugar Cane Techno 24.
3. Geetha S, Padmanabhan D (2001) Effect of hormones on direct somatic embryogenesis in sugarcane. Sugar Tech 3: 120-121.
4. Lakshmanan (2012) Sugarcane tissue culture. Sugarcane for the future.
5. Nand L, Ram K (1997) Yield comparison in sugarcane crop raised from conventional and mericlone derived seed cane. Ind Sugar 47: 617-621.
6. Ramanand Lal M, Singh SB (2005) Comparative performance of micropropagated and conventionally raised crops of sugarcane. Sugar Tech 7: 93-95.
7. Sood N, Gupta PK, Srivastava RK, Gosal SS (2006) Comparative studies on field performance of micropropagated and conventionally propagated sugarcane plants. Plant Tissue Culture and Biotechnology 16: 25-29.
8. Tolera B, Diro M, Belew D (2014) Response of Sugarcane (*Saccharum officinarum* L.) Varieties to BAP and IAA on In vitro Shoot Multiplication. Adv Crop Sci Tech 2: 126.
9. Food and Agriculture Organization of the United States of America (FAO) (2003) Values and Use of industrial crop. Southern Nations, Natural History Society. FAO. Ethiopia, pp: 15-27.