Research

iMedPub Journals
http://www.imedpub.com/

Journal of Plant Sciences and Agricultural Research

2017
Vol.1 No.1:005

Interaction Effects of IBA and Immersion Time on *In Vivo* Rooting Responses of *In Vitro* Raised *Ex-Vitro* Mass Propagated Sugarcane Plants

Gezahegn Terefe[1] and Belay Tolera[2]

[1]Dilla University, School of Natural and Computational Sciences, Department of Biology

[2]Ethiopian Sugar Corporation Research and Development Center, Wonji Research Center, Ethiopia

Corresponding author: Belay Tolera, Ethiopian Sugar Corporation Research and Development Center, Wonji Research Center, Ethiopia, Tel: +251-910-18-16-44; Fax: +251-222-20-01-44; E-mail: belaytolera@yahoo.com

Received date: 24 January, 2017; **Accepted date:** 23 February, 2017; **Published date:** 01 March, 2017

Citation: Terefe G, Tolera B. Interaction Effects of IBA and Immersion Time on *In Vivo* Rooting Responses of *In Vitro* Raised *Ex-Vitro* Mass Propagated Sugarcane Plants. J Plant Sci Agric Res. 2017, 1:1.

Abstract

A study was conducted at Metahara Sugar Estate of Ethiopia Sugar Industry with the objective to determine the effects of different concentrations of IBA and immersion time on ex vitro root induction of detached sugarcane plantlets. Five levels of IBA (0, 0.5, 1, 1.5 and 2 mgL^{-1}) and four levels of immersion time (0, 1, 5 and 10 minutes) with three sugarcane genotypes were combined in factorial treatment combination arrangements with completely randomized design in pot experiment under nursery. The basal end of the detached shoots was dipped in IBA solution for different durations before the shoots were transferred into a polyethylene bag containing a mixed growing medium. The results showed that the interaction effect of IBA, immersion time and the sugarcane genotypes was very highly significant ($p<0.0001$) on number of roots per shoot, average root length and average survival rate of the sugarcane plantlets. Sugarcane plantlets of SP7-1284, C132-81 and C86-56 dipped in 0.5 mgL^{-1} IBA for 10 minutes gave the optimum rooting responses with highest survival rate in all the sugarcane genotypes studied.

Keywords: *In vivo* rooting; Sugarcane genotypes; IBA; Immersion time; SP70-1284; C132-81; C86-56

Introduction

For commercial planting, sugarcane is propagated by vegetative means through stem cuttings. Besides its slow rate of propagation [1-3], production of sufficient quantity disease-free planting material in such large numbers during the planting season is laborious and time consuming. In addition, it requires a substantial quantity of crushable cane that otherwise could be used for sugar and other by-products production. Proper use of tissue culture technology offers an opportunity to mass produce disease-free planting material and is now used to supplement commercial sugarcane propagation in many countries including Ethiopia. Recently, tissue culture technology plays a leading role in rapid multiplication of disease-free and quality planting material and thus, reported to give better cane and sugar yield as compared to the conventional seed sources [4-11].

To utilize this advantage, besides building tissue culture facilities at Metahara, Kuraz, Tendaho and Fincha sugar factories/projects, the Ethiopian Sugar Corporation has been procuring micropropagated sugarcane plantlets from other biotechnology laboratories and will going to require about five million plantlets annually. However, the high tech nature of the technology, limited skills of the supplier organizations and delay of import inputs make the plantlets supply erratic and challenging the sustainable supply. In addition, the cost of procurement is high and increasing from time to time.

The high cost involved in microproagation technology is a major constraint to its popular use in sugarcane [12]. Thus, rapid and low cost *ex-vitro* propagation protocols have been developed to complement microproagation technology for few sugarcane genotypes while reports on *in vivo* rooting are limited. During *in-vitro* propagation, detaching the tillers from the main shoot having common fibrous root system resulted in absence or few roots and resulted in low survival rate of the detached plantlets/tillers.

Most reports of *ex vitro* rooting of plant species have involved treatment with exogenous auxin such as indole-3-acetic acid (IAA), indole-3-butyric acid (IBA), and 1-naphthalene-acetic acid (NAA) with dipping the basal end for different durations for better root induction. Therefore, this study was aimed to determine the effect of different concentrations of IBA and immersion time on *ex vitro* rooting of three sugarcane genotypes (SP70-1284, C132-81 and C86-56).

Result and Discussion

Analysis of variance proved that the Effects of IBA, Immersion Time and sugarcane genotypes have a highly significant (IBA*time* genotype=$p<0.001$) effect on the number of Roots

per shoot and average root length (cm) of all the sugarcane genotypes tested: C132-81, C86-56 and Sp70-1284 **(Table 1).**

Table 1 ANOVA Summary for the interaction effects of IBA and immersion time on *in vivo* Rooting of sugarcane plantlets.

Source of Variation	DF	Mean Squares	
		Number of Roots per shoot	Average root length(cm)
IBA	3	113.61^{ns}	16.80^{*}
Time	2	960.41^{**}	42.43^{***}
IBA*Time	6	699.46^{***}	25.77^{***}
Genotype	2	337.13^{*}	33.72^{***}
IBA*Genotype	6	39.05^{ns}	8.31^{ns}
Time*Genotype	4	164.92^{ns}	22.94^{***}
IBA*Time*Genotype	12	291.62^{**}	25.86^{***}

Remark: DF=Degree of freedom; P ≤ 0.0001=***; P>0.0001 and P<0.001=** P>0.001 and P<0.05=*; P>0.05=ns; IBA =Indole-3-butyric acid.

Table 2 Comparison of the sugarcane genotypes by their rooting responses.

Sugarcane genotype	Pooled value of the response variables	
	Number of roots per shoot	Average root length(cm)
C86 - 56	41.38^{a}	13.90^{c}
C132 - 81	39.14^{b}	17.01^{a}
SP70 -1284	34.97^{c}	15.46^{b}

Remark: a,b,c means with the same letter are not significantly different.

All the sugarcane genotypes showed statistically significant variation both in the number of roots per shoot and average root length (cm) **(Table 2).** In sugarcane genotype SP70-1284, the lowest number of roots per shoot(15.0) and the lowest average root length (9.67 cm) were recorded at 0 mg/l IBA and 0 minute immersion time while the lowest average survival count (20 plants per plot) was recorded at 0.5 mg/l IBA+5 minutes immersion time. Similarly, in C86-56, the lowest number of roots per shoot(19.0) and lowest average survival count(24 plants per plot) were obtained on the control treatment(0 mg/l IBA+0 minute immersion time) while the lowest average root length (11.83 cm) was obtained at 0.5 mg/l IBA+5 minutes immersion time. In sugarcane genotype C132-81, the lowest number of roots per shoot (16.67) and lowest average survival count (21 plants per plot) were found on the control treatment (0 mg/l IBA +0 minute immersion time) while the lowest average root length(11.8 cm) was found at 2 mg/l IBA with 1 minute immersion time **(Table 2).** In sugarcane genotype SP70-1284, the maximum number of roots per shoot (51.67) was recorded at 1.5 mg/l IBA+1 minute immersion time while the highest average root lengths were recorded at 0.5 mg/l IBA+10 minutes immersion time; and 2 mg/l IBA+5 minutes immersion time. However, in this genotype (SP70-1284), the optimum number of roots per shoot (48.33), average root length (19.66 cm) and average survival count (30 plants per plot) were obtained at 0.5 mg/l IBA with 10 minutes immersion time.

In sugarcane genotype C86-56, at 0.5 mg/l IBA, increasing the duration of immersion time from 0 to 1 minute, from 1 to 5 minute, from 5 to 10 minute increased the average number of roots per shoot from 19.0 to 33.67 cm, from 33.67 to 48.33 cm and from 48.33 to 51.33 cm, respectively. In addition, the maximum number of roots per shoot (65.33), highest average root length (20 cm) and maximum survival count (30 plants per plot) were recorded 1 mg/l IBA+5 minutes immersion duration, 1 mg/l IBA+10 minutes immersion time and 0.5 mg/l IBA+10 minutes immersion time respectively. However, the optimum treatment combination selected was 0.5 mg/l IBA with 10 minutes immersion time that produced 51.33 roots per shoot with 19.66 cm root length and an average survival count of 30 plants per plot **(Table 3).**

In sugarcane genotype C132-81, even though the maximum number of roots per shoot (52.0) and highest average root length (22.0 cm) were found at 2 mg/l IBA+1 minute immersion time and 1.5 mg/l IBA+10 minute immersion time, respectively, the optimum treatment combination selected was 0.5mg/l IBA +5 minutes immersion time that gave 31.33 roots per shoot with 14.16 average root length and an average survival count of 29 plants per plot. The absence of IBA in the treatment (control) resulted in reduced number of roots per shoot and hence the survivals count of the plantlets per plot. However, high concentration of IBA (2 mg/l) gave the highest number of roots per shoot (52.0) with the shortest average root length (11.8 cm) with subsequent reduced survival count (26 plants per plot).

Even though the response of different genotypes differs for the same treatment, use of high concentration of IBA may result in excessive rooting with short root length followed by low survival count. Thus, uses of optimum concentration of IBA with appropriate immersion duration is essential in inducing *in vivo* rooting in sugarcane and help improve survival rate of *in vivo* detached sugarcane plantlets in ex-vitro propagation system (EPS). Comparison of the current results with the previous findings was impossible due to the scarcity of research reports on similar activities.

Table 3 Interaction effects of IBA and Immersion time on *in vivo* rooting of sugarcane.

IBA(mg/l)	Immersion Time (minute)	Response of Sugarcane Genotypes								
		SP70-1284			C86-56			C132-81		
		Number of roots per shoot	Average root length (cm)	Average survival count per plot	Number of roots per shoot	Average root length (cm)	Average survival count per plot	Number of roots per shoot	Average root length (cm)	Average survival count per plot
0	0	15.00^{y}	9.67^{q}	21^{g}	19.00^{w}	12.27^{n}	24^{e}	16.67^{x}	16.50^{h}	21^{g}
0.5	1	38.67^{n}	13.66^{m}	25^{f}	33.67^{p}	17.33^{g}	28^{c}	48.67^{g}	21.06^{b}	28^{c}
0.5	5	36.67^{o}	18.33^{f}	20^{g}	48.33^{g}	11.83^{o}	28^{c}	31.33^{q}	14.16^{k}	29^{b}
0.5	10	48.33^{g}	19.66^{d}	30^{a}	51.33^{d}	19.66^{d}	30^{a}	41.67^{l}	21.00^{b}	29^{b}
1	1	43.67^{j}	13.33^{m}	29^{b}	50.00^{e}	13.83^{m}	28^{c}	50.00^{e}	17.16^{g}	29^{b}
1	5	37.33^{n}	15.33^{i}	29^{b}	65.33^{a}	17.33^{g}	29^{b}	51.33^{d}	16.66^{h}	29^{b}
1	10	35.33^{o}	17.66^{g}	28^{c}	21.67^{v}	20.00^{c}	28^{c}	26.00^{r}	15.16^{i}	29^{b}
1.5	1	51.67^{d}	15.83^{i}	28^{c}	59.00^{b}	16.66^{h}	27^{d}	37.00^{n}	15.16^{i}	29^{b}
1.5	5	25.00^{t}	15.33^{i}	28^{c}	33.00^{p}	15.43^{i}	28^{c}	38.33^{n}	15.00^{i}	27^{d}
1.5	10	25.67^{s}	14.00^{l}	27^{d}	34.67^{p}	17.33^{g}	27^{d}	44.67^{i}	22.00^{a}	27^{d}
2	1	49.33^{f}	16.50^{h}	28^{c}	42.50^{k}	13.97^{m}	28^{c}	52.00^{c}	11.80^{p}	26^{e}
2	5	23.00^{u}	19.33^{d}	28^{c}	34.00^{p}	18.75^{f}	27^{d}	35.00^{o}	14.00^{i}	26^{e}
2	10	25.00^{t}	12.33^{n}	27^{d}	46.00^{h}	12.95^{n}	27^{d}	39.00^{m}	19.50^{d}	27^{d}
CV		10.95			8.64					
SE		0.63			1.41					

Remark: Alphabets means with the same letter are not significantly different.

Conclusion

The conventional method of sugarcane planting material propagation has diverse limitations while procurement of large quantity microproagation sugarcane planting materials to complement the conventional method is costly and erratic. Similarly, on *ex-vitro* propagation protocols developed for three sugarcane genotypes: SP70-1284,C132-81 and C86-56; to complement microproagation technology, separation of tillers from the main shoot having common fibrous root system resulted in absent or few roots with subsequent plant death and hence reduced survival rate. Thus, ex-vitro root induction protocol for ex vitro propagation three sugarcane genotypes 'SP70-1284, C132 and C86-56' has been developed. The result proved that *ex-vitro* rooting of *ex vitro* propagated sugarcane plants is highly dependent on the interaction effects IBA, Immersion duration and the sugarcane genotypes. Treatment combination containing 0.5 mgL^{-1} IBA for 10 minutes was found to give optimum *ex-vitro* rooting response with the highest survival rate of the plantlets in all the three sugarcane genotypes studied. On this treatment combination, sugarcane genotype SP70-1284 produced 48.33 roots with 19.66 cm root length resulting in 100% survival rate of the plantlets while C86-56 gave 51.33 roots having 19.66 cm average root length with 100% survival rate of the plantlets. Similarly, C132-81 produced 41.67 roots with 21.0 cm root length and 96.67% survival rate. Thus, the current findings will help improve the root induction response of the sugarcane genotypes and hence their survival rate.

References

1. Sluis CJ (2006) Integrating automation technologies with commercial microproagation. Plant Tissue Culture Engineering, Springer, Dordrecht, The Netherlands, pp: 231-248.
2. Ethiopian Sugar Corporation (ESC) (2014) Sweet Newsletter 4:1.
3. Jalaja NC, Neelamathi D, Sreenivasan TV (2008) Micropropagation for Quality Seed Production in Sugarcane in Asia and the Pacific.

Food and Agriculture Organization of the United Nations and Asia–Pacific Consortium on Agricultural Biotechnology, Asia Pacific Association of Agricultural Research Institutions.

4. Ibrahim M, Tolera B, Aman J, Negi T (2016) 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. J Hortic 3:168.
5. Anonymous (2002) Micropropagation: Tissue culture techniques in sugarcane. Indian Institute of sugarcane Research. Directorate of Sugarcane Development.
6. 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 AmerSoc Sugar Cane Techno 24.
7. Geetha S, Padmanabhan D (2001) Effect of hormones on direct somatic embryogenesis in sugarcane. Sugar Tech 3: 120-121.
8. Lakshmanan (2012) Sugarcane tissue culture. Sugarcane for the future.
9. Nand L, Ram K (1997) Yield comparison in sugarcane crop raised from conventional and mericlone derived seed cane. Ind Sugar 47: 617-621.
10. Ramanand, Lal M, Singh SB (2005) Comparative performance of micropropagated and conventionally raised crops of sugarcane. Sugar Tech 7: 93-95.
11. Sood N, Gupta PK, Srivastava RK, Gosal SS (2006) Comparative studies on field performance of micropropagated and conventionally propagated sugarcane plants. Plant Tissue Cult and Biotech 16: 25-29.
12. Pandey RN, Rastogi J, Sharma ML, Singh RK (2011). Technologies for cost reduction in sugarcane microproagation. African Journal of Biotechnology 10: 7814-7819.