

Research Article

Effect of BA and NAA for Shoot Induction *In vitro* Culture of Sugarcane

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Abstract

The experiment was conducted at the Laboratory of Breeding Division of Bangladesh Sugarcane Research Institute (BSRI), Ishurdi, Pabna during the period of 2011 for the effect of BA and NAA for shoot induction *in vitro* culture of sugarcane. The regeneration of sugarcane varieties with supplementation of 1 mg/l of BA and different concentration of NAA (0.5, 1.0 and 2.0 mg/l) were used in this experiment for shoot induction. In varietal response to application of BA and NAA, among five varieties, the early days to shoot initiation were found in Isd 2-54 and Isd 40. The highest number of shoots per culture and the highest shoot length were observed in Isd 40 and Isd 37 respectively. The effect of concentrations of BA and NAA, 1.0 mg/l BA and 0.5 mg/l of NAA were showed the highest number of shoot per culture and the highest shoot length. Regarding interaction effect of varieties and the doses of BA and NAA, 1 mg/l of BA and 0.50 mg/l of NAA produced the highest number of shoots per culture and shoot length in Isd 40 and Isd 37 respectively.

Keywords

- Sugarcane
- *In vitro* Culture
- BA
- NAA
- Shoot induction

ABBREVIATIONS

Isd: Ishurdi; LJ-C: Latari Jaba-C

INTRODUCTION

The technique of plant tissue culture is being routinely used for producing large number of clonal plants by *in vitro* culture of explants from wide range of species throughout the world. Tissue culture techniques play an important role in the genetic improvement of vegetatively propagated crops like sugarcane [1-3]. The *in vitro* plant regeneration from callus capable of producing somaclonal variants for different traits like high yield, more sugar recovery, disease resistance, drought tolerance and early maturity etc.

Rapid callus formation has been obtained mostly from young leaf sheath [4,5]. To promote regeneration, callus was transferred to medium with different growth hormones [6]. Somaclonal variation has emerged as an important parasexual tool for crop improvement. This technique has been developed as a breeding tool for improving the quality and production of vegetatively propagated crops such as sugarcane [3].

The present study is the regeneration potential of sugarcane varieties with supplementation of auxin (NAA) and cytokinin (BA) in sugarcane on defined MS media for shoot induction *in vitro* culture of sugarcane.

MATERIALS AND METHODS

Five hundred callus of five sugarcane varieties *viz.*: Isd 2/54, LJ-C, Isd 17, Isd 37 and Isd 40 were used as experimental materials in the study. The developed callus was aseptically transferred into fresh medium containing the different hormonal treatments (*viz.*, BA 1.0 and NAA 0.25, 0.50 and 0.75 mg/l) for proliferation and development of shoots. To maintain and ensure aseptic condition precautions were taken in every step of works. All inoculation and aseptic manipulation were carried out by using a laminar air flow cabinet. Hands were properly washed with soap before starting work in laminar airflow cabinet. During the operation hands were rubbed with 70% ethyl alcohol frequency with cotton and wiped cabinet base for maintaining clean condition. Successful shoot formation became evident when small green fresh leaves began to emerge. It is the first sign of regeneration. These tiny leaves when developed in their actual shape were subcultured in fresh medium containing the best combination of NAA at different concentrations showing better performance. Subculture was carried out regularly at an interval of 2-3 weeks. Data were recorded on days to shoot initiation, number of shoots per culture and shoot length (cm). The collected data were analyzed statistically following the analysis of variance (ANOVA) technique and the mean differences were adjudged by Duncan's Multiple Range Test (DMRT) using the statistical computer package program, MSTAT-C (Gomez and Gomez, 1984) [7].

RESULTS AND DISCUSSION

Varietal response to application of BA and NAA

Varietal response to media supplemented with BA and NAA for days to shoot initiation, number of shoots per culture and shoot length are shown in Table (1).

Among the five varieties, the early days to shoots initiation (9.33) took by the variety Isd 2-54 and Isd 40. The highest number of shoots per culture (7.04) and the highest shoot length (3.31cm) were observed in Isd 40 and Isd 37 respectively. The lowest number of shoots per culture (4.06) and the lowest shoot length (1.09cm) were found in Isd 40 and Isd 2-54 respectively (Table 1). The findings were supported by Alam et al. [8], for sugarcane variety Isd 18.

Hormonal effect of BA and NAA

The effect of BA and NAA on days to shoot initiation, number of shoots per culture and shoots length are shown in Table (2).

The early days to shoots initiation (8.20) was found in concentration of 1mg/l of BA and 0.25 mg/l of NAA. The concentration of 1mg/l of BA and 0.50mg/l of NAA produced the highest number of shoots per culture (9.21) and the highest shoot length (2.84cm). The lowest number of shoots per culture (3.33) was found in concentration of 1mg/l of BA and 0.75 mg/l of NAA. The result of this study was consistent with the hypothesis of Skoog and Miller [9]. Similar response was also obtained by Alam et al. [8], in sugarcane variety Isd 18 at the combination of 1.0 mg/l BA + 0.5 mg/l NAA.

Table 1: Varietal response to application of cytokinin and auxin (BA and NAA) for shoot initiation from *in vitro* callus culture of sugarcane.

Varietiesa	Days to shoot initiation	No. of shoot/culture	Shoot length (cm)
Isd 2-54	9.33	6.33	1.09
LJ-C	10.33	5.66	1.72
Isd 17	11.33	5.44	1.41
Isd 37	12.33	4.06	3.31
Isd 40	9.33	7.04	1.99
LSD (5%)	0.786	0.132	0.627

Table 2: Effect of different concentrations of cytokinin and auxin (BA and NAA) for shoot initiation from *in vitro* callus culture of sugarcane.

Cytokina and auxin (mg/l)	Days to shoot initiation	No. of shoot /culture	Shoot length (cm)
BA1.0 + NAA 0.25	8.20	4.58	1.78
BA1.0 + NAA 0.50	10.20	9.21	2.84
BA1.0 + NAA 0.75	13.20	3.33	1.57
LSD (5%)	0.609	0.102	0.485

Table 3: Interaction effect of variety and hormones (cytokinin and auxin) on regeneration of shoot from callus of sugarcane.

Variety and doses of hormone (cytokinin and auxin)	Days to shoot initiation	No. of shoots / culture	Shoot length (cm)
Isd2-54 × BA1.0+ NAA0.25	7.0	5.06	1.73
× BA1.0+ NAA0.50	9.0	10.46	2.61
× BA1.0+ NAA0.75	12.0	3.46	1.36
LJ-C × BA1.0+ NAA0.25	8.0	4.73	1.53
× BA1.0+ NAA0.50	10.0	9.33	2.24
× BA1.0+ NAA0.75	13.0	2.93	1.39
Isd 17 × BA1.0+ NAA0.25	9.0	4.26	1.19
× BA1.0+ NAA0.50	11.0	8.66	1.81
× BA1.0+ NAA0.75	14.0	3.40	1.23
Isd 37 × BA1.0+ NAA0.25	10.0	3.26	2.80
× BA1.0+ NAA0.50	12.0	6.33	5.08
× BA1.0+ NAA0.75	15.0	2.60	2.06
Isd 40 × BA1.0+ NAA0.25	7.0	5.60	1.70
× BA1.0+ NAA0.50	9.0	11.26	2.45
× BA1.0+ NAA0.75	12.0	4.26	1.83
LSD (5%)	1.362	0.229	1.086

Interaction effect of varieties and doses of BA and NAA

Days to shoot initiation, number of shoot per culture and shoot length were found to depend on varieties and shooting media. Results of interaction effect of variety and doses of hormones (cytokinin and auxin) on regeneration of shoot from callus of sugarcane are shown in Table (3).

According to the (Table 3) the days to shoot initiation ranged from 7.0 to 15.0 which showed significant variation among the concentration of BA and NAA. The highest day to shoot initiation (15) was recorded in Isd 37 on shooting media supplement with 1mg/l of BA and 0.75 mg/l of NAA. The early days to shoot initiation [7] were found in variety Isd 2-54 and Isd 40 on same shooting media of 1mg/l of BA and 0.25 mg/l of NAA. The early days to shoot initiation were preferable for shoot production. The combination effect of 1 mg/l of BA and 0.50 mg/l of NAA produced the highest number of shoots per culture and shoot length in Isd 40 and Isd 37 respectively. The results of this study were consistent with the hypothesis of Skoog and Miller [9]. Similar response was also obtained by Alam et al. [8], for sugarcane variety Isd 18 at the combination of 1.0 mg/l BA + 0.5 mg/l NAA.

CONCLUSIONS

The combination of 1mg/l of BA and 0.5mg/l of NAA were found to produce the highest number of shoot per culture and the highest number of shoot length in sugarcane varieties in Isd 40 and Isd 37.

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