ISSN 0258-7122 (Print), 2408-8293 (Online)
Bangladesh J. Agril. Res. 45(2): 209-213, June 2020 **Short Communication**

EFFECT OF CYTOKININ (BA and Kn) ON SHOOT INDUCTION FROM SUGARCANE CALLUS

M. M. RAHMAN[1], R. ALAM[2], H. M. TARIQUE[3]
M. A. M. HUSSEN[4] AND M. G. AZAM[5]

Sugarcane (*Saccharum officinarum* L.), a perennial tropical crop is grown for the sugar stored in its stem and propagated through stem cuttings. It is one of the economically important food-cum cash crops widely cultivated in the tropics to sub tropics and annually provides around 60 to 70 % of the world sugar (Shah *et al.*, 2009). Disease-ridden planting materials can pass diseases like grassy shoot, red rot, ratoon stunting, leaf scald, etc. to succeeding crops. This leads to financial loss due to a reduction in cane yield and sucrose recovery. Plant tissue culture offers a way to overcome these issues and to produce disease-free planting material. This technique has been developed as a breeding tool for improving the quality and production of vegetatively propagated crops such as sugarcane (Siddiqui *et al.*, 1994). *In vitro* regeneration of sugarcane has also been reported by Heinz *et al.*, (1977). MS Basal medium supplemented with benzylamineopurine (BA) and kinetin (Kn) effectively used for rapid shoot multiplication (Ali and Shahid, 2001; Singh *et al*., 2006 and Mekonnen *et al*., 2014). Technique of plant tissue culture is the most successful and tranquil way to produce ample amount of pathogen free and vigorous planting material. In this study various concentration of cytokinin (BA and Kn) were used for shoot regeneration from sugarcane callus.

A total of five hundred callus of five non- flowering sugarcane varieties (which are commercially cultivated) such as, Isd 2-54, LJ-C, Isd 17, Isd 37 and Isd 40 were transferred to the MS medium supplemented with different concentration (0.5, 1.0 and 2.0 mg/l) of BA and Kn for proliferation and development of shoots. The 30 treatments were arranged in a RCB design in each of 5 replications. These were placed on breeding laboratory at BSRI, Ishurdi, Pabna where a temperature of 10-15 ^{0}C and a light intensity of 35000 lux were maintained and ensure aseptic condition precautions were taken in every step of works.

All inoculations were carried out by using a laminar air flow cabinet maintaining proper sterilization and aseptic condition. Successful shoot

[1,2,3] Breeding Division, Bangladesh Sugarcrop Research Institute (BSRI), Ishurdi-6620, Pabna, [4]Bangladesh Agricultural Research Council (BARC), Dhaka-1215. [5]Pulses Research Centre, Bangladesh Agricultural Research Institute (BARI), Ishurdi-6620, Pabna, Bangladesh.

formation became evident when small green fresh leaves began to emerge. It is the first sign of regeneration. These tiny leaves when developed their actual shape, were sub cultured in fresh medium containing the best combination of BA and Kn at different concentrations showed 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 using the statistical computer package program, MSTAT-C and the mean differences were adjudged by Duncan's Multiple Range Test (DMRT) following Gomez and Gomez, 1984.

The days to shoot initiation and the number of shoot production per culture differed among the varieties, shooting media and sources of callus media.

Varietal Response to application BA for shoot initiation

The results on the effects of different varieties and BA treatments on days to shoot initiation and the number of shoot formation are shown in Table 1.

Table 1. Varietal response to application of BA for shoot initiation in *in vitro* callus culture of sugarcane

Varieties	Days to shoot initiation	No. of shoots /culture
Isd 2-54	17.00	1.66
LJ-C	20.00	1.57
Isd 17	19.66	1.61
Isd 37	19.66	1.55
Isd 40	17.33	2.35
LSD (5%)	0.929	0.152

The shortest time to shoot initiation (17 days) and the lowest number of shoots per culture (1.55) was found in variety Isd 2-54 and Isd 37, respectively. The highest number of shoots per culture (2.35) was observed in variety Isd 40 indicating differential response of varieties. The results of Niaz and Quraishi, (2002) supported the present finding.

Shoot Induction by BA

Results on the effect of different concentrations of cytokinin on shoot induction *in vitro* culture of sugarcane are shown in Table 2.

Table 2. Effect of different concentrations of BA on shoot initiation in *in vitro* callus culture of sugarcane

Doses of BA (mg/l)	Days to shoot initiation	No. of shoots/culture
0.50	21.40	1.51
1.0	16.20	2.48
2.0	18.60	1.26
LSD (5%)	0.249	0.118

Among the three concentrations, 1.0 mg/l of BA took the shortest time to shoot initiation (16.20 days) and produced highest number of shoot per culture (2.48). The concentration of 1mg/l of BA was found to have better response for shoot initiation and number of shoots per culture as also reported by Niaz and Quraishi, (2002). Biradar *et al.,* (2009) reported that MS medium containing 1.0 mg/l of BA and Kn produced maximum number of shoots. These findings were similar to the study. In this study the effect of Kn was very poor and hence was not discussed in detail.

Interaction Effect of Varieties and Doses of BA

Results of interaction effect of sugarcane variety and BA and Kn are shown in Table 3.

Table 3. Interaction effect of varieties and doses of BA and Kn on regeneration of shoot from callus of sugarcane

Varieties and doses of BA/Kn (mg/l)	Days to shoot initiation		No. of shoots /culture	
	BA	Kn	BA	Kn
Isd 2-54 × 0.50	19.00	-	1.60	-
× 1.0	15.00	20	2.20	1
× 2.0	17.00	27	1.20	1
LJ-C × 0.50	26.00	-	1.23	-
× 1.0	16.00	20	2.40	1
× 2.0	18.00	22	1.10	1
Isd 17 × 0.50	21.00	-	1.40	-
× 1.0	18.00	20	2.20	1
× 2.0	20.00	23	1.23	1
Isd 37 × 0.50	22.00	-	1.06	-
× 1.0	17.00	19	2.40	2
× 2.0	20.00	20	1.20	2
Isd 40 × 0.50	19.00	-	2.26	-
× 1.0	15.00	19	3.20	1
× 2.0	18.00	21	1.60	1
LSD (5%)	1.611	1.78	0.263	0.28

The doses of 1mg/l of BA induced shoot initiation in shortest time (15.00 days) in Isd 2-54 and Isd 40; and the highest number of shoots per culture (3.20) was recorded in Isd 40. The lowest number of shoots per culture (1.06) was recorded in Isd 37 on the shooting media of 2 mg/l of BA. One mg/l of BA was found to be comparatively more effective in producing shoot initiation in shortest time and higher number of shoot per culture. The concentration of 1mg/l of BA was found to have better response for number of shoot per culture as also reported by Niaz and Quraishi, 2002.

In general, response of kinetin on shoot initiation was not encouraging. Among three concentrations of kinetin 1.0 and 2.0mg/l induced only one shoot from the callus within 19-27 days (Table 3). But 0.5 mg/L concentrations could not induce any shoot. Gopitha *et al.* (2010) reported similar response for Kn.

The results revealed that BA showed better response than Kn for shoot induction in sugarcane production among the varieties. The concentration of 1.0 mg/l of BA was suitable for shoot initiation and shoot production but the response of Kn on shoot initiation and production was very poor.

Acknowledgement

The authors are thankful to the scientist of breeding division of Bangladesh Sugarcrop Research Institute for their technical support. We also acknowledge the financial assistance from the ministry of Science and Information & Communication Technology of Bangladesh for supporting NSICT fellowship to carry out the study.

References

Ali, K. and Shahid A. 2001. Rapid multiplication of sugarcane through micro-propagation technique. *Pak Sugar J*. **16** (6): 11- 14.

Biradar, S., Biradar, D. P., Patil, V. C., Patil, S. S., Kambar, N. S. 2009. *In vitro* plant regeneration using shoot tip culture in commercial cultivar of sugarcane. Karnataka J. of Agri. Sci. **22** (1): 21- 24.

Gomez, K. A. and Gomez, A. A. 1984. Statistical procedures for agricultural research. John Wiley and Sons, New York. p. 97-411.

Gopitha, K., A. L. Rhavani, and J. Santhilmanickam. 2010. Effects of the different auxins and cytokinen callus induction, shoot, root regeneration in sugarcane. *International J. Pharma Bio Sci.* **1** (3): 1-7.

Heinz, D. J., Krisnamutri, M., Nickell, L. G. and Maretzi, A. 1977. Cell tissue and organ culture in sugarcane improvement. *In*: Applied and Fundamental Aspects of Plant Cell, Tissue and Organ Culture. Springer, Berlin, Heidelburg, New York, pp. 3-17.

Mekonnen, T., Diro, M., Sharma, M. and Negi, T. 2014. Protocol optimization for *in vitro* mass propagation of two sugarcane (*Saccharum officinarum* L.) clones grown in Ethiopia. *Afr. J. Biot.*. **13** (12): 1358-1368.

Niaz, F. and Quraishi. A. 2002. Effect of growth regulators on the regeneration potential of two sugarcane cultures SPE-213 and CPF 237. *Pak. J. Bio. Sci*. **5** (10): 1081-1083.

Shah, A. H., Rashid N. H., Saleem, M. S., Tahir, F. M. and Iqbal, J. 2009. An efficient short and cost-effective regeneration system for transformation studies of sugarcane (*Saccharum officinarum* L.). *Pak J. Bot.* **42** (2): 509-514.

Siddiqui, S. H., Khatri, A., Khan, M. A., Javed. A. L. 1994. *In vitro* cultures: a source of genetic variability and an aid to sugarcane improvement. *Pak. J. Agric. Res.***15**: 127-133.

Singh, N., Kumar, A., and Garg, G. K. 2006. Genotype dependent influence of phytohormone combination and subculturing on micropropagation of sugarcane varieties. *Indian J. Biotechnol*. **5**: 99-106.