

SHORT COMMUNICATION**EFFECT OF PROPAGULE TRIMMING ON SHOOT MULTIPLICATION RATE
IN SUGARCANE MICROPROPAGATION****Swapanil Yadav^{1*}, Aquil Ahmad¹, Jyoti Rastogi² and Madan Lal³****Abstract**

With a view to study the effect of trimming of propagules on the rate of shoot multiplication during micropropagation of sugarcane, the shoot cultures were subcultured on fresh multiplication medium after trimming the leaves of shoot clumps to different lengths i.e. 3.0, 4.0, 5.0 and >6.0 cms. The results showed that trimming of propagules to a height of 3.0 cm during subculture enhanced the rate of shoot multiplication as well as shoot growth in variety CoSe 01235 suggesting that this practice could be helpful in producing more number of plantlets in a seed multiplication programme through *in-vitro* micropropagation.

Key words: Micropropagation, sugarcane, shoot trimming

Sugarcane, an important cash crop of the tropics and subtropics, is cultivated in about 70 countries of the world. Generally, sugarcane is propagated through conventional method by planting small cane pieces containing 2-3 buds. A number of high yielding and high sugared varieties have been developed which have the yield and recovery potentials greater than the national average. But due to slower multiplication ratio, it takes several years to build up sufficient stock of seed cane through conventional methods of multiplication, by the time the varieties start deteriorating in yield and quality. *In vitro* micropropagation technique is emerging as a complimentary tool to conventional methods of seed multiplication. And now there has been a considerable interest in applying this technique in multiplication of newly released varieties of sugarcane for rapid

spread over a large area in a comparatively shorter period of time. Various protocols for *in-vitro* micropropagation of sugarcane have been described by several investigators during past decades (Sauvaire and Glazy 1978; Hendre et al. 1983; Lee 1987; Sreenivasan and Sreenivasan, 1992; Lal and Singh 1994; Ramanand and Lal 1994; Shukla et al. 1994).

The regeneration responses are highly influenced under varied growth room conditions and also with different subculturing methods/practices which enormously affect the rate of shoot multiplication. Much work has been done on some of the factors mentioned above but information regarding the effect of subculturing methods on the rate of shoot multiplication is scanty. Present work was undertaken to investigate the effect of trimming of

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shoots on the rate of multiplication during in vitro micropropagation of sugarcane variety CoSe 01235.

Fresh tops were collected from 8-10 months old field grown healthy plants of an early maturing sugarcane variety CoSe 01235 (popularly known as 'Rapti') growing at the farms of Sugarcane Research Institute, Shahjahanpur. After removing all the open green leaves from the tops, about 6 cm long spindle segments were dissected out and washed under running tap water for about half an hour. The segments were rinsed with 1% aqueous detergent solution for 5 min followed by thorough washing with tap water. The washed segments were dipped in 70% ethanol for 10 sec and again rinsed with sterile distilled water several times. Finally, these segments were surface sterilized with 0.1% aqueous mercuric chloride (HgCl_2) solution for 10 min followed by several washings with sterile distilled water.

About 1.0 cm long shoot tip explants containing apical dome along with 1-2 leaf primordia were carefully excised from the sterilized segments and immediately inoculated on agar solidified (0.8%) MS medium (Murashige and Skoog 1962) supplemented with BAP (0.5 mg/l), Kinetin (0.5 mg/l) and sucrose (30 g/l) for shoot initiation. The pH of the medium was adjusted to 5.8 before autoclaving. Cultures were incubated at $25 \pm 2^\circ\text{C}$ under 16 h illumination of 4000 lux provided by cool white fluorescent tubes. The established shoot cultures were multiplied and used for experimentation. The leaves of shoot clumps were trimmed with the help of a sterile scissor so as to maintain their final length at various levels (i.e. 3.0, 4.0 and 5.0 cm). A set of shoot clumps having length of >6.0 cm was used without trimming the leaves which served as control (Fig. 1). The inocula of different lengths were



Fig. 1. Shoot cultures after trimming the leaves upto various length i.e. 3.0, 4.0, 5.0 and >6.0 cm (control)

subcultured up to four cycles at 15 day interval. Trimming of propagules was done at the time of each subculture onto fresh media, to maintain the length of propagules corresponding to the previous cycle. Cumulative number of shoots produced at each cycle was recorded. The average multiplication ratio was calculated at the end of 4th cycle (i.e. after 60 days of multiplication).

The total number of shoots produced at the end of fourth cycle decreased with increasing length of propagules (Table 1). A maximum of 2372 shoots could be produced at the end of fourth cycle when 3.0 cm long propagules were used which was about two fold higher than the number of shoots (1190) produced in case of untrimmed propagules (Fig. 2). It was also noticed that irrespective of length of propagules, the number of shoots produced in subsequent cycles (i.e. from 1st cycle to 4th cycle) increased considerably; however, the magnitude of increase was higher in case of smaller propagules than the longer ones. This indicated that the rate of multiplication in subsequent cycles was dependent on length of propagules used.

Table 1. Effect of trimming of propagules on rate of shoot multiplication and shoot growth in sugarcane variety CoSe 01235

Responses	Number of cycles (15 day each)	Length of shoot clumps (cm)			
		3.0	4.0	5.0	>6.0 (Control)
Cumulative number of shoots produced at the end of each cycle (figures within parentheses denote multiplication ratio)	1st	16 (1:8.0)	16 (1:8.0)	14 (1:7.0)	13 (1:6.5)
	2nd	110 (1:6.8)	88 (1:5.5)	72 (1:5.1)	64 (1:4.9)
	3rd	540 (1:4.9)	390 (1:4.4)	364 (1:5.0)	304 (1:4.8)
	4th	2372 (1:4.4)	1810 (1:4.6)	1570 (1:4.3)	1190 (1:3.9)
Average multiplication ratio per cycle	-	1 : 6.0	1 : 5.6	1 : 5.3	1 : 5.0
Average shoot length* at 4 th cycle (cm)	-	4.1±0.5	4.3±0.3	5.6±0.4	5.8±0.5
Shoot vigour at 4 th cycle	-	Good	Good	Moderate	Moderate

*Shoots less than 3.0 cm in length were not considered.



Fig. 2. Cultures showing shoot formation from trimmed (left) and un-trimmed (control) shoot clump (right)

Enumeration of multiplication ratio revealed a decreasing trend with the increasing length of propagules at each cycle. At the end of the first cycle, maximum multiplication ratio (1:8.0) obtained in case of 3.0 cm long propagules, was reduced to

(1:6.5) when untrimmed propagules were used. A similar decreasing trend was observed in case of propagules of other sizes as well (Table 1).

The mean multiplication ratio recorded at the end of fourth cycle was highest (1:6.0) in case of 3.0 cm long propagules followed by 1:5.6, 1:5.3 and 1:5.0 in inoculums of 4.0, 5.0 and >6.0 cm length, respectively. The shoots produced from shorter propagules (3.0, 4.0 cm) were more vigorous showing good growth as compared to the longer propagules (5.0, >6.0cm) which showed moderate growth. The average shoot length recorded at the end of fourth cycle showed a gradual increase with increasing length of propagules which varied from 4.1±0.5 to 5.8±0.5 cm. The results showed that trimming of leaves of shoot clumps during subculture enhanced the rate of shoot multiplication as well as shoot growth in variety CoS 01235 suggesting that this practice can be helpful in producing more number

of plantlets in a seed multiplication programme through *in-vitro* micropropagation.

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