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Growth of Sugarcane Plantlets on Media Limiting Concentration of Inorganic Nutrient and Growth Promotor Supplement

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ABSTRACT

Plantlets of sugarcane were cultured on media having different concentration of inorganic nutrient (1, 1/2, 1/4 and 1/8 strength) and different growth promotor supplement (without growth promotor, with 1 mg/l kinetin and 1 mg/l NAA, 10% coconut water or 20 g/l sucrose). Half-strength (1/2) media without growth promotor and with 1 mg/l kinetin and 1 mg/l NAA were found to provide high vigor and short stem plantlets and hence are suitable media for culture of sugarcane plantlets for the purpose of germplasm preservation.

Keywords: sugarcane plantlet, limiting media, growth promotor

INTRODUCTION

Sugarcane is an important vegetative-propagated crop in which *in vitro* technique is suggested to be an interesting alternative method for germplasm preservation. Various methods; i.e; were used to reduce *in vitro* growth for preservation sugarcane germplasm; In this, (preservation of sugarcane callus using daminozide (Visessuwan and Lersrutaiyotin, 1992) and low temperature (Lersrutaiyotin *et al.*, 1992) preservation of sugarcane plantlets using growth suppression chemicals (Sonthichai *et al.*, 1992) low temperature (Lersrutaiyotin *et al.*, in press)) In this study growth reduction of sugarcane plantlets by limiting concentration of inorganic nutrient and growth promotor supplement was investigated.

MATERIALS AND METHODS

1. Production of plantlets

Shoot tips of sugarcane cultivar CO_2 were dissected from shoots, surface sterilized in 10% Clorox solution for 15 minutes and rinsed 3 times with sterized distilled water. Shoot tips were cut into about 5 mm in length and cultured on Murashige and Skoog (MS) media supplemented with 1 mg/l kinetin, 0.5 mg/l gibberellic acid (GA3), 1 mg/l indole-3-butylic acid (IBA), 10% by volume of coconut water and 20 g/l sucrose at pH 5.8. Then, plantlets were subcultured onto modified Murashige and Skoog media supplemented with 1 mg/l 2,4-dichlorophenoxy acetic acid (2,4-D), 0.1 mg/l myo-inositol, 0.02 mg/l glycine, 20 g/l sucrose and 10% by volume of coconut water (CD media).

2. Culture plantlets on media limiting concentration of inorganic nutrient and growth promotor supplement

After subcultured for 2 months, plantlets were cut to approximately 4 cm in length and not less than 1 g in weight. Height of each plantlet was recorded before transfered to the media. The studied media composed by 2 factors. The first factor was concentration of inorganic nutrient (full-strength of inorganic nutrient, half-strength (1/2) of inorganic nutrient, 1/4 strength of inorganic nutrient and 1/8 strength of inorganic nutrient). The second factor was growth promotor supplements (no growth promotor, 1 mg/l kinetin and 1 mg/l NAA (1-naphthyl acetic acid), 10% coconut water and 20 g/l sucrose. Total number

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Table 1 Treatments of media in this study.

Growth promotor	Media concentration			
	Full(1)	Half(1/2)	1/4	1/8
None (N)	1 N	1/2 N	1/4 N	1/8 N
1 mg/l kinetin + 1 mg/l NAA	1 KN	1/2 KN	1/4 KN	1/8 KN
10% coconut water (CW)	1 CW	1/2 CW	1/4 CW	1/8 CW
20 g/l sucrose (S)	1 S	1/2 S	1/4 S	1/8 S

of treatments was 16 as shown in Table 1. There were 6 plantlets for each treatment. All sugarcane plantlets were cultured under 1,000 lux light intensity, 16 hours photoperiod at 27°C. Height of each plantlet was recorded at 75 days after starting of the experiment. The difference of height of each plantlet at the starting time and at 75 days after starting experiment were recorded as the increasing height. Factorial in completely randomized design (CRD) was used to evaluate the effect of each treatment.

Table 2 Increasing height of sugarcane plantlets cultured on media with different inorganic nutrient concentration.

Media concentration	Increase height
1	8.11 bc
1/2	7.09 c
1/4	11.96 ab
1/8	15.07 a

RESULTS AND DISCUSSION

1. Induction of plantlets from sugarcane shoot tips

After culture shoot tips on media for 1 - 2 days, many explants became dark and died owing to the phenolic compounds in medium from shoot tips of sugarcane. Therefore, shoot tips had to be subcultured every 3 days during the first two weeks of culture. Shoot tips cultured for 2 months developed into plantlets with 4 - 10 leaves. These plantlets were used for the study.

2. Growth of sugarcane plantlets cultured on media limiting concentration and supplements

Increasing height of sugarcane plantlets after cultured for 75 days was at the highest level when cultured on medium supplemented with 1/8 strength which was not significantly different from the increasing height of plantlets cultured on 1/4 strength medium (Table 2). On the other hand, the increasing height of the plantlets cultured on 1/2 strength medium was at the lowest level which was not significantly different from the increasing height of plantlets cultured on full-strength media. The increasing height of sugarcane plantlets cultured on media without supplement and supplemented with kinetin+NAA, coconut water or sucrose at each concentration of inorganic nutrient shown in Figure 1 indicated that the effect of media concentration on increasing height of sugarcane plantlet differred for each growth promotor supplement. In the high-strength media (full- and half- strength of inorganic nutrient), coconut water and sucrose tended to affect the increasing height of plantlet. But sugarcane plantlets cultured on the low-strength (1/8 strength) media supplemented with sucrose had low level of increasing height compared to the other growth promotor supplement treatments.

With various concentration of inorganic nutrient, growth promotor supplement did not clearly affect the increasing height of sugarcane plantlets (Table 3). The low level of increasing height was observed in plantlets cultured on media supplemented with 1 mg/l kinetin and 1 mg/l NAA. However, growth promotor supplement of media affected the increasing height of plantlets at different level of each

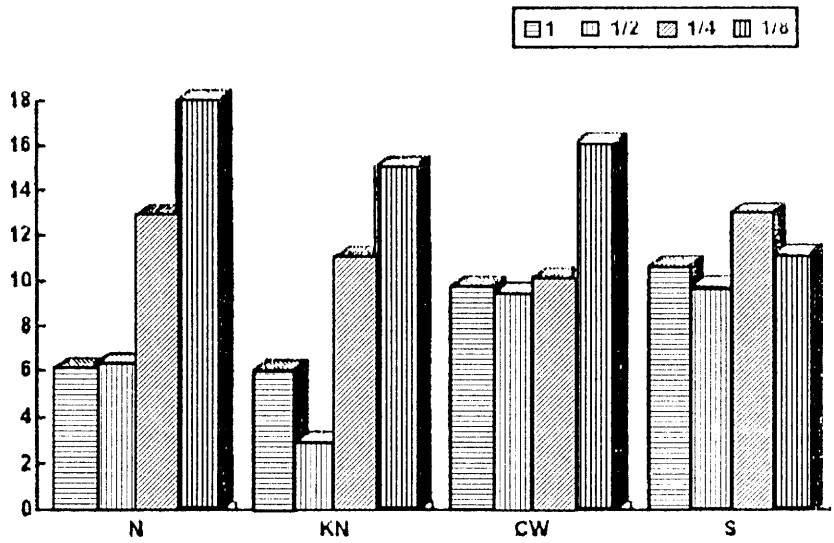


Figure 1 Increasing height of sugarcane plantlets cultured on media supplemented with different growth promotor supplement with different concentration of inorganic nutrient.

concentration of inorganic nutrient. Without growth promotor supplement, height at sugarcane plantlets tended to increase at higher level compared to the plantlets cultured on media with growth promotor supplement in which the increasing height of sugarcane plantlets cultured on 1/8 strength medium was about 2 times of those cultured on 1/4 strength medium and was about 3 times of those cultured on full-strength or 1/2 strength medium (Figure 2). With growth promotor supplement, height of sugarcane plantlets cultured on media supplemented with 1 mg/l kinetin and 1 mg/l NAA tended to increase at lower level compared to those cultured on media supplemented with 10% coconut water or with 20 g/l sucrose. Moreover, height of sugarcane plantlets at 75 days cultured on media supplemented with 20 g/l sucrose tended to be more stable. The stability of the level of increasing height in sugarcane plantlets cultured on media supplemented with 20 g/l sucrose and 10% coconut water may relate to the ability of plantlets in increasing height at the high strength of inorganic nutrient.

Table 3 Increasing height of sugarcane plantlets cultured on media with different growth promotor supplement.

Media supplement	Increase height
None	10.86
10% coconut water	8.79
20 g/l sucrose	11.47
1 mg/l kinetin + 1 mg/l NAA	11.11

With the high-strength (full- and half-strength) of inorganic nutrient, increasing height of sugarcane plantlets cultured on media supplemented with 10% coconut water and with 20 g/l sucrose was about 10 cm, while those supplemented with 1 mg/l kinetin and 1 mg/l NAA and without supplement were about 6 cm. For the purpose of germplasm preservation, plantlets should have the slow growth rate, especially in height. The high level of increasing height of the plantlets cultured on media with high concentration of inorganic nutrient was due to the extension of leaf in which most of the leaves were slim and had pale green color. The plantlets having long leaf and stem are easily contaminated by fungi. Moreover, these plantlets also had the low vigor which may result in the slow recovering rate of plantlet when culture on the normal media. Therefore, the full-strength and half-strength media with out growth promotor supple-

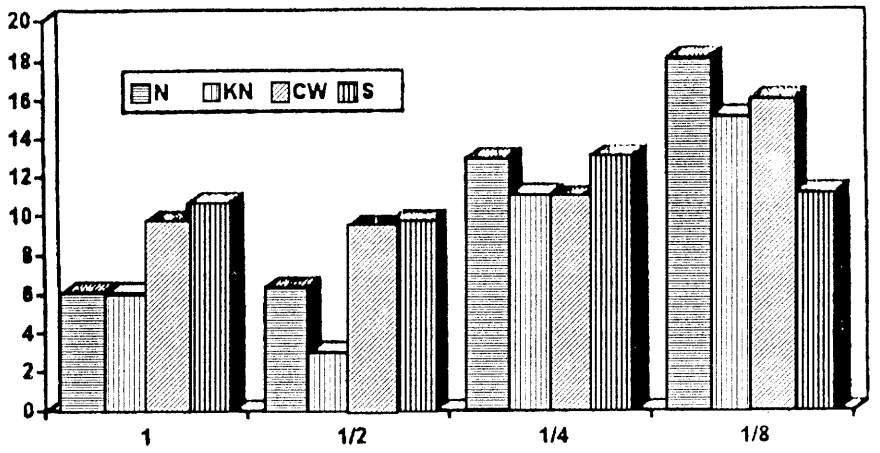


Figure 2 Increasing height of sugarcane plantlets cultured on media with differrent concentration of inorganic nutrient with different growth promotor supplement.

N = no growth promotor
CW = 10% coconut water
S = 20 g/l sucrose
K = 1 mg/l kinetin and 1 mg/l NAA

1 = full-strength of inorganic nutrient
1/2 = half-strength of inorganic nutrient
1/4 = 1/4 strength of inorganic nutrient
1/8 = 1/8 strength of inorganic nutrient

ment or with supplement of kinetin and NAA are suggested, especially the half-strength media supplemented with 1 mg/l kinetin and 1 mg/l NAA.

Sugarcane plantlets cultured on media supplemented with coconut water or sucrose had higher vigor level than supplemented with kinetin and NAA or without growth promotor supplement (Table 4). Coconut water and sucrose are suggested to be necessary for the vigor of sugarcane plantlet especially when culture on low concentration of inorganic nutrient. Nevertheless, plantlets cultured on half-strength media without growth promotor supplement also showed the high level of vigor. As the increasing height of these plantlets was low, the half-strength of inorganic nutrient without growth promotor supplement was suggested as media for *in vitro* preservation of sugarcane plantlets, especially if the period of storage is not so long (about 3 months).

Table 4 Vigor of sugarcane plantlets cultured on media with different inorganic nutrient concentration and growth promotor supplement.

Media concentration	Media supplement			
	N	CW	S	KN
Full strength	++[1/]	++++	++++	+
Half strength	++++	+++	++++	++
1/4 strength	+++	++++	+++	++
1/8 strength	++	++	+++	+

1/ ++++ = high vigor, +++ = moderate high vigor, ++ = moderate vigor, + = low vigor

CONCLUSION

Sugarcane plantlets cultured on media limiting concentration of inorganic nutrient tended to have high rate of extension of leaf and low vigor. Half-strength media supplemented with 1 mg/l kinetin and 1 mg/l NAA or without growth promotor supplement are observed to be suitable media for culture sugarcane plantlets for the purpose of germplasm preservation as sugarcane plantlets cultured on these media had low level of increasing height while maintaining high vigor.

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