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The Micropropagation of Bananas

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ABSTRACT

A study to obtain a method of rapid clonal propagation of bananas was done. The investigated cultivars were Pisang Mas (AA), Pisang Ambon Kuning (AAA), Pisang Barangan (AAA), and Pisang Rajabulu (AAB). The basal medium was MS, the treatment were IAA (0 – 4.5 mg/l) and BAP (0 – 10.5 mg/l), and the explants were suckers from the field. The experiment was designed with Randomized complete design, and repeated in 10 bottles for each treatment. The result of experiment showed that IAA alone significantly induced shoot multiplication. The role of IAA could be replaced by BAP. Cultivars, IAA and BAP interacted each other to induce shoot multiplication. The best treatment to induce shoot multiplication was the combination of IAA and BAP, and the concentration depend on the cultivar. After 8 weeks, the highest number of axilar shoots (12.6 shoots/bottles) was obtained by Pisang Ambon (AAA), followed by Pisang Mas (AA) 8.2 shoot/bottle, Pisang Barangan (AAA) 7.8 shoot/bottle, and Pisang Rajabulu (AAB) 7.6 shoot/bottle.

Key words : Banana, Micropropagation

INTRODUCTION

Bananas is an important fruit in Indonesia, because of nutritive values, as source of vitamins, minerals and carbohydrates. The bananas also easily grow in many areas in Indonesia. The domestic demand of bananas increase 5-10 % per annum, while the exsport markets always open, such as in Japan and Korea. To increase the production of bananas, good quality and quality of seedlings are necessary. Tissue culture technique can be used to achieve that purpose. In this paper, some factors affecting the micropropagation of bananas are reported.

MATERIAL AND METHOD

Investigated cultivars in the experiment were Pisang Mas (*Musa acuminata*, AA group), Pisang Ambon (*Musa acuminata*, AAA group), Pisang Barangan (*Musa acuminata*, AAA group) and Pisang Rajabulu (*Musa Paradisiaca*, AAB group).

Initiation of Culture

The explants were suckers obtained from the field. The suckers were sterilized by soaking them in the mixture of detergent, Agrimycin and Dithane M-45, for overnight. The suckers were then sterilized with NaHCl0 (10%) and sterilized water in the laminar airflow cabinet. The sterilized suckers (diameter 1 cm) were then placed on the solid MS medium containing BAP and IAA, and incubated at 25°C under light illumination for 16 hours per day.

Selection to Get Shoot Multiplication lines

The suckers that could be multiplied into more than 3 shoots were maintained and subcultured for ten times in the MS medium containing BAP and IAA. Number of shoot formed in the cultured were counted every four weeks, and subcultured was done every eight weeks. The axilar shoots that could not multiply were discarded. The produced axilar shoots were used for the following study, to improve medium, for a better propagation rate.

The Effects of BAP and IAA on the Micropropagations of Axilar Shoots.

Single axilar shoot excised from selected lines was used as an explant. The explants were transferred to the MS basal solid medium to eliminate carry over effect of the previous media. The cultures were incubated at the standard condition (25°C, 16 hours/day illumination) for 10 weeks. In order to know the multiplication rate potential of cultivars, the explants were then cultivated on the MS-0 again, and incubated for 8 weeks; subsequently were used for the following experiment. The treatments were IAA (0-4.5 mg/l) and BAP (0-10.5 mg/l), which were added on the MS basal medium. The experiment was designed with Randomized complete design and repeated in 10 bottles for each treatment. Cultures were incubated on the standard condition.

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RESULT AND DISCUSSION

Initiation of Cultures

The suckers were collected from field during dry season (Juni – Agustus, 1992) and wet season (October – December, 1992). Most of suckers (90%) collected from the field during dry season were died in the culture because of browning. Some treatments such as subculturing in a medium containing antioxsidant (Vitamin C, 0.5 mg/l), or transfering to liquid medium were not able to reduce the percentage of death culture. In the contrary, the cultures initiated from suckers collected during rainy season, grew well and number of death cultures were relatifely low (30%).

Tabel 1 showed that Pisang Ambon Kuning produced highest number of shoots than other cultivar. But the multiplication rate depen on the growth regulator. There were some explants which remain to be single without any dome or adventif shoot. Among those four cultivars, Pisang Rajabulu was the most difficult to break its dormancy and time needed to induce dome or shoots was the longest. The percentage of browning and death explants was also the highest. It might relate to cultivar.

The Effects of BAP and IAA on the Micropropagations of Axilar Shoots

Tabel 2 showed the number of axilar shoot produced in plant growth regulator free media.

The results are for the mean of 10 replicates (bottles) containing 1 explant/bottle.

Tabel 1. Average number of shoots produced per explant (suckers), 10 weeks after initiation. The media containing several plant growth regulators

Cultivars	ppm (mg/l)			
	BAP 3	BAP 5	BAP 9	BAP 9 IAA 3
Mas (AA)	3.4 ± 1.9 a	4.8 ± 2.2 a	4.6 ± 2.1 a	5.2 ± 2.4 a
Ambon K. (AAA)	5.3 ± 2.4 a	6.2 ± 2.7 a	4.3 ± 2.2 a	7.4 ± 2.3 a
Barangan (AAA)	4.2 ± 2.6 a	5.4 ± 2.1 a	4.6 ± 2.2 a	6.2 ± 2.1 a
Rajabulu (AAB)	3.2 ± 1.8 a	4.3 ± 2.3 a	4.8 ± 2.7 a	5.2 ± 2.8 a

Tabel 2. The average number of axilar shoots of four cultivats grown in plant growth regulator free medium

Cultivars	Weeks after initiation				
	4		6	7	8
Mas (AA)	1.35 b	1.35 b	1.35 b	1.35 b	1.46 b
Ambon K. (AAA)	2.63 ab	3.07 a	3.22 a	3.38 a	3.5 b
Barangan (AAA)	3.99 a	3.99 a	4.38 a	4.47 a	4.47 a
Rajabulu (AAB)	2.92 ab	2.92 a	2.92 a	3.03 a	3.03 a

Note

Tabel 3. The effect of IAA on the production of axilar shoots of four banana cultivars.

IAA (mg/l)	Weeks after initiation 4	5	6	7	8
			Mas (AA)		
1.5	1.46 f	1.96 g	2.16 g	2.22 f	2.39 g
3.0	3.15 d	4.25 cde	4.56 de	5.12 cd	5.21 de
4.5	1.93 ef	2.29 fg	2.42 fg	2.67 ef	2.81 fg
			Ambon (AAA)		
1.5	4.47 be	5.70 bc	6.68 be	7.50 b	7.97 bc
3.0	6.10 a	8.20 a	9.48 a	10.26 a	10.99 a
4.5	5.16 ab	6.90 ab	8.44 ab	9.68 a	9.99 ab
			Barangan (AAA)		
1.5	3.19 d	4.65 cd	4.98 de	5.45 c	5.70 d
3.0	2.81 de	3.87 de	4.38 de	4.65 cd	4.74 de
4.5	3.63 cd	4.30 cde	4.56 de	5.02 cd	5.21 de
			Rajabulu (AAB)		
1.5	2.81 de	3.11 ef	3.50 ef	3.70 de	3.86 ef
3.0	4.38 bc	5.31 bcd	6.00 cd	6.31 bc	6.47 cd
4.5	3.58 cd	4.21 cde	4.54 de	4.84 cd	4.93 de

Note : Number followed by the same letter in the same column was not significantly different by the Duncan test (5%). The results are for the mean of 10 replicates.

Tabel 4. The effect of IAA and BAP on the average number of shoots per explant of four cultivar banana.

Treatment	Weeks after initiation 5	6	7	8
		Mas (AA)		
I_1B_1	2.96 h-m	3.58 f-g	3.91 h-m	4.04 g-m
I_1B_2	0.99 n	0.99 n	0.99 n	1.19 n
I_1B_3	2.16 l-n	2.16 l-n	2.16 l-n	2.26 l-n
I_2B_1	2.49 j-n	2.70 h-k	2.81 j-n	2.81 j-n
I_2B_2	6.68 a-f	7.23 a-e	7.91 a-g	8.81 j-n
I_2B_3	3.99 f-m	4.25 e-j	5.21 e-j	5.21 e-j
I_3B_1	1.87 mn	1.96 mn	1.96 mn	2.12 ln
I_3B_2	2.32 k-n	2.42 i-k	2.85 j-n	2.99 j-n
I_3B_3	2.70 i-n	2.96 h-k	3.25 i-m	3.38 j-n
		Ambon (AAA)		
I_1B_1	4.79 b-k	5.5 c-h	6.16 d-i	6.42 c-i
I_1B_2	5.90 a-g	6.39 c-g	7.68 a-g	8.14 a-e
I_1B_3	6.52 a-f	8.44 a-d	8.80 a-d	9.42 a-e
I_2B_1	7.91 a-c	8.86 a-c	9.68 a-g	10.32 a-c
I_2B_2	7.85 a-d	9.05 a-c	9.8 a-d	10.32 a-c
I_2B_3	8.92 a	10.59 ab	11.54 ab	12.24 ab
I_3B_1	5.63 b-i	6.26 c-g	7.06 c-h	10.32 a-c
I_3B_2	8.14 ab	11.06 a	11.89 a	12.6 a
I_3B_3	7.28 a-c	8.44 a-d	10.46 a-c	10.72 ab

		Barangan (AAA)		
I_1B_1	5.02 b-j	5.50 c-h	6.05 d-i	6.47 c-i
I_1B_2	4.38 e-i	4.38 e-j	4.70 g-l	4.84 e-l
I_1B_3	4.56 d-e	5.02 d-l	5.75 e-j	5.75 d-j
I_2B_1	5.40 a-l	6.58 c-f	7.00 c-h	7.23 b-g
I_2B_2	1.90 mn	1.90 mn	1.90 mn	1.90 mn
I_2B_3	4.65 c-l	5.40 c-h	5.80 c-j	5.80 d-j
I_3B_1	3.07 g-m	3.42 g-j	3.42 l-m	3.42 l-n
I_3B_2	6.21 a-f	6.54 c-f	7.34 b-h	7.85 b-f
I_3B_3	3.87 f-m	4.04 c-j	4.61 g-l	4.84 e-l
		Rajabulu(AAB)		
I_1B_1	2.96 h-m	3.07 h-j	3.38 l-m	3.74 h-m
I_1B_2	4.74 c-k	5.40 c-h	5.60 f-j	5.90 d-j
I_1B_3	1.90 mn	2.36 i-k	2.36 k-n	2.36 k-n
I_2B_1	4.30 lm	4.43 e-j	4.74 g-l	4.74 g-l
I_2B_2	5.70 a-h	6.68 b-f	7.01 c-h	7.17 b-h
I_2B_3	6.05 a-f	7.12 a-e	7.28 b-h	7.68 b-f
I_3B_1	4.70 c-l	5.07 d-i	5.21 f-j	5.21 ek
I_3B_2	4.12 e-m	4.38 e-j	4.38 e-j	4.38 f-m
I_3B_3	3.70 f-m	4.25 e-j	4.88 g-k	5.21 e-k

Note Number followed by the same letter in the same column was not significant on the Duncan test (5%). The results are for the mean of 10 replicates.

I1 : 1.5 mg/l IAA
I2 : 3.0 mg/l IAA
I3 : 4.5 mg/l IAA
B1 : 3.5 mg/l BAP
B2 : 7.0 mg/l BAP
B3 : 10.5 mg/l BAP

The potential axilar shoots produced per explant on the MS basal medium without plant growth regulator was different among cultivars from cultivar to other cultivars. This data showed that there were differences on the capability of cultivars to produce axilar shoot in the plant growth regulator free medium.

The lowest number of shoots was obtained by Pisang Mas, while the highest one was achieved by Pisang Barangan. There were no significant differences on the number of axilar shoots among Pisang Barangan, Ambon Kuning and Rajabulu. Pisang Mas (*Musa acuminata*, AA) is belong to diploid banana, while other cultivar are belong to triploid cultivar. It seems that the multiplication rate of triploid cultivar is higher than diploid cultivar. According to Vuylsteke and de Langhe (1984) the multiplication rate of the AA diploid cultivar was lower than the AAA triploid cultivar, and the AAA triploid cultivar was lower than multiplication rate of the AAB triploid cultivar.

The addition of BAP and IAA on the medium induced higher shoot multiplication. The interaction effect between IAA and cultivar on the shoot multiplication was showed on Tabel 3.

The Pisang Mas, Ambon and Rajabulu tend to produce the highest number of axilar shoots on the medium containing 3 mg/l IAA, and the 4.5 mg/l IAA tend to decrease number of axilar shoots per explant. In case of Pisang Barangan, different concentration of IAA did not induce significant axilar shoot production. The addition of exogenous auxin changed the balance of endogenous plant homone then were followed by morphogenesis.

The concentration of BAP added to the treatments were high enough but there was no significant result on the shoot multiplication of the four cultivars. It was possible the endogenous cytokinin of bananas was high enough, therefore the addition of exogenous cytokinin did not give significant result. Another possibility, the concentrations of cytokinin were in the same range that could not induce significant result.

Tabel 4 showed the interaction effect of IAA – BAP on the average number of axilar shoots of four cultivars. At eigh weeks after initiation, in case of Pisang Mas, the best treatment to induce multiple shoot was 3.0 mg/l IAA in combination of 7.0 mg/l BAP (8.2 shoots). In case of Pisang Ambon Kuning, the best treatment was 4.5 mg/l IAA in combination with 7.0 mg/l BAP (12.24 shoots). In case of Pisang Barangan, the best treatment was 4.5 mg/l IAA in combination with 7.0 mg/l BAP (7.8 shoots). In case of Pisang Rajabulu, the best treatment was 3 mg/l IAA in combination with 10.5 mg/l BAP (7.6 shoots).

This result showed that the multiplication rate was depend on the cultivar, while the role of growth regulator was as trigger to induce the potential of shoot multiplication of each cultivar.

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