

IN VITRO ADDITION OF BENZYLADENINE (BA) AND THIAMINE ON GROWTH OF ABACA BANANA SHOOTS

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ABSTRACT

Abaca is a type of banana plant used as paper material for securities industries, therefore, it has high economic value. This study therefore aims to determine the effect of Benzyladenine (BA) and thiamine in the induction of Abaca banana shoots in vitro. It was conducted at the Biotechnology laboratory of UPN "Veteran" Yogyakarta with the laboratory experiments arranged in a 2-factor Complete Randomized Design. The first factor in the experiment was the concentration of BA (3 ppm, 4 ppm, 5 ppm) and the second was the concentration of thiamine (10 mg/L, 20 mg/L, 30 mg/L). The results showed an interaction with plantlet height parameters in the treatment of BA 3 ppm and Thiamin 20 mg/L. 5 ppm Benzyl Adenine resulted in the highest yields on shoots length, the number of leaves, and fresh weight plantlets. 10 mg/l Thiamine resulted in highest yields on shoots length, number of leaves, and fresh weight plantlets.

Keywords: BAP, thiamin, abaca banana, in vitro

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INTRODUCTION

Abaca is a type of banana with high economic value, with its stem fiber used in textile and security paper industries. As a superior commodity, its number is relatively limited, with the need of a large planting area to meet the high market demand (1). According to a research (2), abaca fiber is used in making marine ropes due to its strength, weight, and resistance to saltwater. (3) reported that abaca waste is used as material for composting, door ceilings, etc.

In developed countries, seedlings are used to determine crop production for agribusiness. However, with the technological advances, it is produced through tissue culture which provides a new opportunity for agribusiness in Indonesia. Furthermore, the propagation of abaca tissue culture produces higher multiplication compared to conventional methods.

The application of cytokinin in banana plants has been widely carried out. Research (4) shows that addition of 5 mg/l IIBA and 5 mg/l BAP + 5 mg/l 2-ip in Ambon bananas yields 4,4 shoots in 8 weeks. Furthermore (5), the addition of BAP 3 mg/l to Abaca banana produces 9 shoots, and the addition of 1mg/l NAA, increases the amount to 6,67 roots per explants.

The addition of thiamine in culture media optimizes respiration activity in plant tissue, which are needed in the process of cell division, elongation and enlargement in the apical and intercalary meristem which promotes plant growth (6). Therefore, this research provides a complete propagation technique of Abaca bananas in vitro through the addition of cytokinins and thiamine. Efforts to increase the growth of banana shoots in this study were carried out by adding BA and thiamine in the Murashige and Skoog media.

RESEARCH METHODOLOGY

This research was conducted at the Biotechnology Laboratory of UPN "Veteran" Yogyakarta, with the experiment carried

out according to a 2-factor Complete Randomized Design. The first factor consists of BA concentrations which consists of 3 ppm, 4 ppm, 5 ppm and the second Thiamine (10 mg/L; 20 mg/L; 30 mg/L) using all media addition with 250 mg/L Casein Hydrolyzate. Sterilization of the medium by autoclav was carried out at 20 psi, and 121°C for 30 minutes. Each culture bottle was planted with one explant originating from a small group of abaca plantlets which were separated according to the treatment. Furthermore, the plantlets were placed in the incubation room for 12 weeks at 22°C.

RESULT AND DISCUSSION

Shoots induction is performed to prepare the roots for acclimatization using the least number of multiplied plantlets, which was carried out for eight weeks after multiplication. Furthermore, the explants are planted on media containing Benzyl adenine and thiamine to produce shoots and leaves, which are expected to produce endogenous auxins as the basal and induce root formation.

The observations showed that the combination of B2T1, B3T1, B3T2 treatments had higher interactions (synergy) with plantlets than B1T2 and B3T3 (Table 1). The addition of 10 mg/l thiamine and 3 ppm of Benzyl Adenine increases plantlet height parameters. While inserting thiamin 10 mg/l into culture media optimizes respiration activity in plant tissue, which is indicated by an increase in plantlet height. Energy (ATP) which is the result of the respiration process is used to synthesize proteins, carbohydrates, fats, and other essential compounds (6). These are needed in cell division, elongation, and enlargement in the apical and intercalary meristems to make plants grow taller (7). The addition of 4 ppm Benzyl Adenine plays an important role in cell division and enlargement which accelerates the height of apical meristem plantlets in early development (8).

Table 1. Average plant height (cm)

Treatment	B1 (3 ppm)	B2 (4 ppm)	B3 (5 ppm)	Mean
T1 (10 mg/L)	13.43 abc	16.17 a	16.50 a	15.37
T2 (20 mg/L)	11.17 c	14.67 abc	17.17 a	14.33
T3 (30 mg/L)	15.33 ab	16.00 ab	10.33 c	13.89
Mean	13.31	15.61	14.67	(+)

Note: Numbers in one column followed by the same letters indicate no significant difference in the 5% DMRT test.
(+) = there is an interaction

Table 2 shows the number and length of shoots, as well as leaves, and weight of each explant tends to decrease with increase in BA concentrations (4 ppm to 5 ppm). According to (9), auxin generally inhibits bud growth, whereas the combination of high cytokinin concentrations with low auxin is important in the formation of shoots and leaves.

The use of concentrations of growth regulators need to be inappropriate with the ability to inhibit the growth and development of plantlets. This is caused by interactions between growth regulators in a medium that affects cell differentiation (10). It defines Benzyl Adenine as a growth regulator which regulates division and increases the enlargement of stem cells. The increase in the number of shoots was due to the cell size (11).

While cytokinins are growth regulators that play a role in cell division, enlargement of young leaves, regulate leaf growth and shoots. (12) stated that the addition of high dose cytokinin results in the formation of primordial stems or shoots, while the formation of shoots in vitro largely determines the success of seed production.

The addition of 10 mg/l thiamine in culture media optimizes respiration activity in plant tissue. It is indicated by the increase of shoot length, the number of leaves, and fresh plantlet weight. The energy in the form of ATP synthesizes proteins, carbohydrates, fats, and other essential compounds (6).

Table 2. The average number of shoots, shoots length, number of leaves, fresh weight, and dry weight.

Treatment	Number of shoots	Shoots length (cm)	Number of leaves	Fresh weight (g)	Dry weight (g)
Thiamine					
T1(10 mg/L)	4.89 a	4.00 a	6.78 a	5.97 a	0.38 a
T2(20 mg/L)	1.67 b	3.00 b	5.22 b	2.72 b	0.15 a
T3(30 mg/L)	1.67 b	2.56 b	5.22 b	2.59 b	0.13 a
Benzyl Adenine					
B1(3 ppm)	1.67 p	3.17 q	4.44 q	2.53 q	0.12 p
B2(4 ppm)	1.67 p	3.06 q	5.33 q	3.49 q	0.15 p
B3(5 ppm)	1.89 p	5.33 p	7.44 p	6.26 p	0.19 p
Interaction	(-)	(-)	(-)	(-)	(-)

Note: Numbers in one column followed by the same letters indicate no significant difference in the 5% DMRT test.
(-) = there is no interaction

Fresh plant weight is the accumulation of water weight from respiration and cell metabolism (protein). It is also the accumulation of photosynthesis results which is obtained through diffusion and contact between the media and the root surface. The addition of thiamine spurs the division and enlargement of cells which affects plantlet fresh weight. Besides, thiamine also stimulates cultured tissue organogenesis which accelerate plantlet growth.

The concentration of Benzyl Adenine and thiamine treatments showed no significant difference in the dry weight parameters. The fresh weight is necessarily dry due to the comparison of the content which assimilates and the presence of water in the plant. (13) stated that dry weight is an indicator of plant growth that represents the accumulation of assimilates from total plant growth and development during its lifetime. According to (14), the dry weight is an accumulation of organic matter in the form of biomass which reflects the energy produced from the photosynthesis process.

CONCLUSION

In conclusion, the following analysis were obtained:

1. There was an interaction in the combination of 4 ppm BA and 10 mg/l thiamine in the plantlet height parameter.
2. 5 ppm Benzyl Adenine resulted in the highest yields on shoots length, the number of leaves, and fresh weight plantlets.
3. 10 mg/l Thiamine resulted in highest yields on shoots length, number of leaves, and fresh weight plantlets.

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