

The Use of Bean Sprout Extract as Supplement for the Growth of Plaintain Unti Sayang (*Musa paradisiaca* L.) by Tissue Culture

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Abstract

One of the problems in plaintain development is on the plant breeding that has been conducted conventionally by using seedlings, so that it requires time and wide field, different treatment, and potentially brings pests and diseases. The bean sprout extract is potentially useful in improving the explant growth quality of plaintain by tissue culture. This study aims to know the influence of bean sprout extract concentration as supplement in tissue culture media upon the explant growth of plaintain Unti sayang (*Musa paradisiaca* L). There are 4 dosages of bean sprout extract as control. The data was analyzed by using ANOVA and was continued by using BNJ test. The result of the experiment shows that the bean sprout extract influences significantly on the plantlets height, leaf number, root length, and root number. The dosage of 100 g/l and 200 g/l bean sprout extracts provide the best result on the experimenting component of plantlet height, leaf number, root length, and root number. Meanwhile, the experimenting component of leaf number and the wet weight of the plantlets provide the best result on MS without any addition of bean sprout extract.

Keywords: Plaintain *Musa paradisiaca* L, Tissue culture, Bean sprout culture, Murashige and Skoog Media

1. Introduction

Plaintain Unti sayang (*Musa paradisiaca* L.) has a very important role in Indonesia since it is the most consuming fruits, compared to the others. In addition, plaintain has also multifunction since almost all of its parts can be used. For example, the inflorescence can be used as vegetable, the leaves can be used as wrapper of traditional cakes, the pseudo-stem can be used as chips, the skin can be used to make vinegar through fermentation process and to be yogurt raw material, the stem can be used as the cattle foods, compost material, and also it can be processed to be fiber for clothes and paper (Rosdiana, 2009)

The breeding of banana was conducted conventionally by using seedling, namely by slicing the seedlings that have two to three buds. However, breeding by using this technique generally needs a quite long time, wide field, created differently, and potentially brings pests and diseases. Thus, it then becomes the problem in the banana development. Therefore, tissue culture was conducted to get more effective result in the breeding process in order to produce more quality seeds in a greater number. Wetherell (1982) said that this breeding technique is also called as *in vitro*, in which every cell is able to grow to be a new individual.

The growth of the plant parts in tissue culture are significantly influenced by the type of media and components inside (Thymann, 1935). (Gunawan, 1998). In the previous study, the use of bean sprout extract could stimulate orchid more optimally compared to the growth of orchid without bean sprout extract (Soreyowinoto and Moeso, 1977). According to Amaliah and Astuti (2006) and USDA (2009), bean sprout extract contains vitamin and mineral such as vitamin C, Thiamin, Riboflavin, Niasin, Vitamin B, β -karotein, Vitamin A, and Vitamin E. whereas, mineral contained in bean sprout are Calcium (Ca), Ferro (Fe), magnesium (Mg), phosphor (P), sodium (Na), zink (Zn), tembaga (Cu) dan mangan (Mn). Based on the vitamin and mineral content in the bean sprout extract, it is potentially used in the growth of the plaintain explant by tissue culture.

2. Method of the Study

This study was conducted in Tissue Culture Laboratorium Instalation of Regional Duties Implementation Unit, Horticultural Breeding Centers, Agriculture Department of Corps and Horticulture of Sout Sulawesi Province, in February 2012 to May 2012. The planting material used was plaintain inoculum *M. paradisiaca* L. the product of sub culture of 6-month-old bid. The laboratory study used Completely Randomized Design (CRD) with three times repetitions for each treatment. The data resulted was analyzed by using F 0.05 and 0.01 tests; also BNJ 0.05 test to differentiate the treatments.

The study was conducted in several stages continuously, including sterilizing tools and material, formulating stock liquid, formulating bean sprout extract, making media and planting inoculum. This study used basic formulation of MS (Murashige and Skoog) Media with the treatments: MS without adding bean sprout extract (control), MS + 50 g/l bean sprout extract, MS + 100 g/l bean sprout extract, MS + 150 g/l bean sprout extract, and MS + 200 g/l bean sprout extract. The observation components in the age of 12, MST: plantlet height, leaf number, root length, and root number, and plantlet wet weight.

3. Result

3.1 Plantlet Height

The result of observation of plantlet height average of 12-week plaintain after planting is presented in Table 1. The analysis of variance shows that the treatment of bean sprout extract in every concentration influences significantly on F 0.01 test taraff on the plantlet height of the plaintain.

Table 1. The Plantlet Height Average (cm) of the Plaintain of *M. paradisica* L. in week 12th after planting in Various Bean Sprout Extract.

Treatment	High Planlet (cm)	$\sqrt{X+0,5}$	NP BNJ 0.05
D0 = MS without the addition of bean sprouts extract	5.39	2.43 ^b	1.111
D1 = MS + extract 50 g/l	6.30	2.54 ^b	
D2 = MS + extract 100 g/l	15.30	3.94 ^a	
D3 = MS + extract 150 g/l	11.48	3.45 ^{ab}	
D4 = MS + extract 200 g/l	13.73	3.77 ^a	

Information: The Average value that is followed by same letter is insignificantly different in BNJ 0.05 test taraff.

The result of BNJ in Table 1 shows that the highest average of plantlet height is obtained in the treatment of MS + 100 g/l bean sprout extract (D2), significantly different from the tratment of MS + 50 g/l bean sprout extract (D1), and MS without addition of bean sprout extract (D0). However, it is insignificantly different with the treatment of MS + 150 g/l bean sprout extract (D3) and MS + 200 g/l bean sprout extract (D4).

3.2 Leaf Lenght

The result of observation data analysis of leaf length average of plaintain in week 12th after planting shows that the addition of bean sprout extract into clture media significantly influences on F 0.05 test on the leaf length of the plaintain.

Table 2. The Leaf Length Average (cm) of the Plaintain of *M. paradisica* L.in week 12th after planting in Various Bean Sprout Extract.

Treatment	Long Leaf (cm)	$\sqrt{X+0,5}$	NP BNJ 0.05
D0 = MS without the addition of bean sprouts extract	2.65	1.77 b	0.835
D1 = MS + extract 50 g/l	2.89	1.80 b	
D2 = MS + extract 100 g/l	6.84	2.69 a	
D3 = MS + extract 150 g/l	5.14	2.37 ab	
D4 = MS + extract 200 g/l	7.23	2.77 a	

Information: The Average value that is followed by same letter is insignificantly different in BNJ 0.05 test taraff.

The result of BNJ 0.05 test on the leaf length of the plaintain in week 12th after planting (Table 2) shows that the treatment of MS + 200 g/l bean sprout extract (D4) provides the best and significant different from MS without addition of bean sprout extract (D0) and MS + 50 g/l bean sprout extract (D1). However it is insignificantly different in the tratment of MS + 100 g/l bean sprout extract (D2) and MS + 150 g/l bean sprout extract (D3).

3.3 Leaf Number

The result of the observation of leaf number average of the plaintain in week 12th after planting is provided in Table 3.

Table 3. The Leaf Number Average of the Plaintain of *M. paradisica* L. in the age of week 12th after planting in Various Bean sprout Extract.

Treatment	Number of leaves	$\sqrt{X+0,5}$	NP BNJ 0.05
D0 = MS without the addition of bean sprouts extract	8.33	2.97 ^a	0.502
D1 = MS + extract 50 g/l	2.50	1.72 ^c	
D2 = MS + extract 100 g/l	4.75	2.28 ^b	
D3 = MS + extract 150 g/l	3.50	2.00 ^{bc}	
D4 = MS + extract 200 g/l	3.67	2.03 ^{bc}	

Information: The Average value that is followed by same letter is insignificantly different in BNJ 0.05 test taraff.

The result of BNJ 0.05 test on the leaf number formed in week 12th after planting (Table 3) shows that MS without addition of bean sprout exstract (D0) indicates the most leaf number and insignificantly different from the tratment of MS + 50 g/l bean sprout extract (D1), MS + 100 g/l bean sprout extract (D2), MS + 150 g/l bean sprout extract (D3) and MS + 200 g/l bean sprout extract (D4).

3.4 Root Lenght

The result of the observation of root lenght average of the plaintain *M. paradisica* L. in week 12th after planting is provided in Table 4. The analysis of variance shows that the treatment of bean sprout extract in every consentration influences significantly on F 0.01 test taraff on the root lenght.

Table 4. The Root Lenght Average (cm) of the Plaintain of *M. paradisica* L.in week 12th after planting in Various Bean Sprout Extract.

Treatment	The root length (cm)	$\sqrt{X+0,5}$	NP BNJ 0.05
D0 = MS without the addition of bean sprouts extract	1.77	1.49 ^b	1.126
D1 = MS + extract 50 g/l	3.66	2.02 ^{ab}	
D2 = MS + extract 100 g/l	7.50	2.76 ^{ab}	
D3 = MS + extract 150 g/l	5.88	2.51 ^{ab}	
D4 = MS + extract 200 g/l	8.46	2.97 ^a	

Notes: The number that is followed by same letter is insignificantly different in BNJ 0.05 test.

The result of BNJ test in Table 4 shows that the longest root length average is obtained in the treatment of MS + 200 g/l bean sprout extract (D4) and significantly different from MS without addition of bean sprout extract (D0). However it is insignificantly different in the treatment of MS + 50 g/l bean sprout extract (D1) and MS + 100 g/l bean sprout extract (D2) as well as MS + 150 g/l bean sprout extract (D3).

3.4 Root Number

The result of the observation of root number average of the plaintain in week 12th after planting is provided in Table 5. The analysis of variance shows that the treatment of bean sprout extract in every concentration influences significantly on F 0.01 test taraff on the plantlet root number formed.

Table 5. The Root Number Average (cm) of the Plaintain of *M. paradisica* L. in week 12th after planting in Various Bean Sprout Extract.

Treatment	number of Roots	$\sqrt{X+0,5}$	NP BNJ 0.05
D0 = MS without the addition of bean sprouts extract	1.67	1.42 ^b	0.742
D1 = MS + extract 50 g/l	1.67	1.46 ^b	
D2 = MS + extract 100 g/l	4.42	2.20 ^a	
D3 = MS + extract 150 g/l	4.08	2.14 ^a	
D4 = MS + extract 200 g/l	4.92	2.32 ^a	

Information: The number that is followed by same letter is insignificantly different in BNJ 0.05 test taraff.

The result of BNJ test in Table 5 shows that the root number average is mostly obtained in the treatment of MS + 200 g/l bean sprout extract (D4) and significantly different from MS without addition of bean sprout extract (D0). However it is insignificantly different in the treatment of MS + 50 g/l bean sprout extract (D1) and MS + 100 g/l bean sprout extract (D3) as well as MS + 150 g/l bean sprout extract (D3).

3.5 Plantlet Wet Weight

The result of the observation of plantlet wet weight average of the plaintain *M. paradisica* L. in week 12th after planting is provided in Table 6. The analysis of variance shows that the treatment of bean sprout extract in every concentration influences significantly plantlet wet weight.

Table 6. The Plantlet Wet Weight Average (gram) of the Plaintain of *M. paradisiaca* L. in week 12th after planting in Various Bean sprout Extract.

Treatment	Wet weight (g)	$\sqrt{X+0,5}$	NP BNJ 0.05
D0 = MS without the addition of bean sprouts extract	2.83	1.81 a	
D1 = MS + extract 50 g/l	0.61	1.04 b	
D2 = MS + extract 100 g/l	1.86	1.52 ab	0.57
D3 = MS + extract 150 g/l	1.07	1.24 b	
D4 = MS + extract 200 g/l	1.45	1.39 ab	

Information: The number that is followed by same letter is insignificantly different in BNJ 0.05 test taraff.

The result of BNJ 0.05 test on the plantlet wet weight of plaintain *M. paradisiaca* L. in week 12th after planting (Table 6) shows that MS without addition of bean sprout extract (D0) provides the best influence and significantly different from the tratment of MS + 50 g/l bean sprout extract (D1) and MS + 150 g/l bean sprout extract (D3); while it is insignificantly different from MS + 100 g/l bean sprout extract (D2) and MS + 200 g/l bean sprout extract (D4).

4. Discussion

The growth and development of plantlet in tissue culture technique is influenced by the media formulation that is used. The media formulation consists of several mineral, vitamins, and organic materials. To increase the quality of particular growth of the plantlets, the addition of growth hormon or organic material in the form of plant or fruit extracts can be added.

The result of the study provides a description, showing that the addition of bean sprout extract supplement can be used to improve the quality of plaintain explant growth in the tissue culture media. The same result has been showed in the study of Amaliah and Astuti (2006), in which the addition of bean sprout extract in several concentrations gives different influence on the growth of Moth Orchid (*Phalaenopsis amabilis* L).

Bean sprout is sprout that emerges from green bean, containing carbohydrate, protein, Ca, P, Fe, and vitamin A, B1, C, in the form of soluble compounds, so that it is easier to be absorbed by the plant (Amiliah and Astuti, 2006). By the provision of the compounds, the plaintain can grow better.

Protein can be decomposed as a nitrogen source (N) that can fertilize the plant. The role of N element for soil is to stimulate the vegetative growth of the plant since it can be used as the basic material in amino acid and protein syntheses. Amino acid compound (Tryptophan) is precursor of the formation of auxin hormone which can boost the cell division so that the plant can be longer and taller (Mo'o, 1992).

Tryptophan is the most important organic substance in IAA biosynthesis process (auxin).

According to Sriyanti and Wijayanti (1994), auxin effect may increase the osmotic pressure, enhance the protein synthesis, boost the cell permeability toward water, and soften the cell wall, so that the water can get into the cell and increase the cell volume. In low concentration (based on the plant need), auxin can stimulate the growth of root, while with high concentration it can resist the coleoptile elongation rate (root tip) and stem (Sriyanti, 2000).

According to Sriyanti and Wijayanti (1994), food stock supports the energy transfer in chemistry reaction in plant body. Phosphor is significantly required in seedlings phase. In addition, Sriyanti (2000) said that phosphor (P) element is mostly required by the plant for the vegetative growth and to strengthen the plant part, so that the plant can be stronger. Similarly, Mo'o (1992) said that P element will stimulate the growth of the root. Thus, it enables the bean sprout extract to be able to increase the growth of root, root number, and the plant leaf length.

Another element contained in bean sprout is vitamin. According to Sriyanti and Wijayanti (1994), thiamin (vitamin B1) is an essential vitamin for almost all of plant tissue culture. The function of thiamin is to boost the cell division in root meristem and act as coenzyme in reaction that produce energy from carbohydrate.

The treatment of MS + 200 g/l bean sprout extract (D4) provides leaf length, root length, and root number better than other treatments. It is due to the addition of 200 g/l bean sprout extract (D4) into MS media that can indicate the existence of optimum compounds and nutrient in a good media. Bean sprout also contains nutrients that are useful for the banana growth; one of which is calcium. According to Sutiyoso (2013), calcium acts as cell wall, managing the permeability of the cell wall, boosting the plant growth due to the active cell division and cell extension, protein synthesis, carbohydrate transportation to the target parts, as well as in roots growing point.

Meanwhile, in MS media without bean sprout (D0) and MS + 50 g/l bean sprout (D1), it shows that plantlet wet weight is higher. However, it seems that the plantlet performance seems to be weaker and vigorous. This condition will cause resistance on the plantlet in acclimation phase due to the addition of protoplasm in plantlet, causing the cell wall soft and more permeable. As the result, water will easily enter the cell and make the cell bigger. Hence, it causes the plantlet wet weight higher compared to the other treatments. Unfortunately, this condition is poorly good for the plantlet development for the plantlet will be easily attacked by disease, fragile, and weaker compared to the plantlet with bean sprout extract treatment.

Furthermore, in MS without bean sprout extract (D0), it shows that the plant height and root growth components are not significantly better. It relates to the auxin and vitamin contents that are less in number, so that the plantlet roots growth becomes slower (Anonim, 2012). According to Hoflich (1995), this is due to natural auxin hormone in bean sprout that can stimulate the root cells.

5. Conclusion

The addition of bean sprout extract as supplement in basic media of MS (Murashige and Skoog) can increase the growth and development of plantain *M. paradisiaca* L. that is bred by tissue

culture.

The addition of bean sprout between 100-200 g/l of media provides a better effect on the plantlet height, leaf length, root length, and root number of plantain *M. paradisiaca* L.

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