ON-FARM GROWER-FRIENDLY NURSERY TECHNIQUE FOR ACCLIMATIZATION OF TISSUE-CULTURED BANANA SEEDLINGS

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Abstract

Banana is one of the common fruit preferences in the world. The market demand for this crop is increasing year round. Tissue-cultured banana seedlings is getting widely used as planting materials for the growers with its quality assured properties, particularly for commercial plantation. However, most of the small scale growers are still using conventional planting sources (water suckers and sword suckers). They do not prefer tissue-cultured seedlings due to its expensive price, delicate to handle and often result in high post-transplanting mortality rate. The considerably high cost is due to the process of producing the tissue-cultured seedlings at the nursery stage (acclimatization). Even at the nursery stage the conventional acclimatization process of the plantlets in the nursery results in non-uniformity of establishment and high mortality rate. Research was conducted to develop a nursery technique that will improve these drawbacks in order to implement greater acceptance and responsive level for the growers towards tissue-cultured seedlings. Based on the results, the developed technique has increase the survival rate (reducing mortality rate) of seedlings as compared to the conventional technique. In addition, this developed technique can be adopted on-farm instead of using protected nursery as conventionally practiced. Using this technique, the seedlings would be more easily adapted to the farm after transplanting. Ordinary farmers prefer younger seedlings (10-15cm in height) for planting as they growing faster than that of older seedlings with common quality (15-25cm in height). Therefore, girth size of seedlings would be the higher consideration in meeting the requirement for transplanting purpose. The developed technique has shown better growth performance in both height and girth parameters in the first 36 days as compared to conventional technique. Meaning that, this helps to shorten the nursery period and create an alternative standard for seedlings to be transplanted.

Keywords: Banana, tissue culture, nursery technique, acclimatization
Introduction

Banana is one of the members in the family Musaceae. It is a unique group of plants within Zingiberales, and contributes to the nourishment and culture of millions of people worldwide, especially in tropical regions. Currently, it is one of the most important fresh fruits that grown in mass for trade and export purposes, particularly in tropical and subtropical countries (Kamaludin et al., 2012).

Banana cultivation is one of the top ranking commodities production in the world, which ranked at 12th place with production of 101992743 million tons in 2012 (Anon., 2012). Malaysia is one of the countries that develop banana production as the major crop in the country. It is the fourth economical crop production after oil palm, rubber and paddy in Malaysia (Anon., 2012). Banana can be consumed in various ways or produced into different products. It can be eaten as fresh fruits, or processed into cooked product, such as fried banana or banana cakes, or even processed into industrial product, for example banana flours or protein powder. The availability of the crop causes it achieves exceptional high demands in different industries. However, the current supply could not meet the high demand of this crop due to inconsistence planting of banana.

Previously, the main problems that cause the inconsistence planting of banana is the difficulty in getting high quality free disease planting materials (seedlings) through natural propagation methods. But this problem has soon overcome by the introduction of micro-propagation (tissue culture) techniques in producing banana seedlings that are quality and quantity assured. With tissue culture techniques, mass production of free disease plantlets can be produced in shorter period. However, these plantlets are highly susceptible to external environment condition, which required a critical process, called acclimatization process (hardening) before transplanting to the field. The cost of this seedlings becomes relatively high due to the involvement of laboratory process as well as long acclimatization period, 8 to 12 weeks in common (Lule M et al, 2013; Robinson, J. C. and Galan Sauco, V., 20091,2). On top of that, the hardened seedlings, with recommended physical requirements (Robinson, J. C. and Galan Sauco, V., 20091,2) are still susceptible to the field environment after transplanting, leading to high mortality rate.

Macro-propagation techniques are developed for production of banana planting materials worldwide and being practiced, especially for small-scale farmers (La multiplication, 1955; Pillay and Tripathi, 2007; Ahmad Yusuf and Suhaimi Othman, 2014; Njuwe et al., NA). However, these macro-propagation techniques require relatively higher quantity of disease-free plant source for mass production as compared to tissue culture micro-propagation techniques. The quality of seedlings produced are greatly relying on the quality of planting material source (the vegetative parts of the banana plant), as going down from generation, the quality of the banana would be lower from the previous one, therefore the quality of produced seedlings from these sources may become lower. Therefore, large-scale farmers or plantations still prefer tissue-cultured seedlings. Though, alternative acclimatization techniques should be introduced in order to make tissue-cultured seedlings be more accessible and affordable for all ranges of farmers and plantations.
The development of techniques in improving the hardening process of tissue-cultured seedlings would make these planting materials available, accessible and affordable. The hardening technique should be practiced in a simpler and cheaper in cost of nursery setting, and easily adapted by growers. And, the produced seedlings should have higher survival capability in both nursery and field planting condition. Through this, the cost of tissue-cultured seedlings can be reduced and demands on seedlings can be fed. With the success in providing enough high quality and quantity of seedlings, banana productions can be done year round and this benefits to the other banana related production industries, in Malaysia and other countries.

**Problem Statement**

Banana become one of the important fresh fruits for trade and export since being the 12th place in world top ranking commodities production (Anon., 2012). Non-systematic planting leads by difficulty in obtaining adequate supply of uniform free disease banana seedlings with high yield potential causes inconsistent production. Tissue-cultured seedlings have solved this problem but somehow the survival rate in early field planting stage are considerably low compared to conventional seedlings, particularly handling by small scale and inexpeirence growers. One of the potential factor that lead to this issue is due to the acclimatization (hardening) process of tissue-cultured seedlings. Most of the conventional or common hardening process of this seedlings are being done in considerably “over-protective” environment condition. The nursery settings have create environment conditions that are still far different from the actual farm environment conditions. This has causes low adaptable level of seedlings to survive in farm environment later after transplanting, which lead to stunted growth or mortality. Apart from this, in fact the survival rate of seedlings in nursery stage are also remain uncertainty, which lies between 50 to 90% depends on techniques used (Au, V.H. et al., 2012; Kavoo-Mwangi et al., 2013). Therefore, there is a need to develop techniques that could overcome these drawbacks. This research focused on creating an alternative hardening technique that able to be practiced in less protective (closer to farm environment) environment condition, in which potentially contribute to higher adaptation level of seedlings in field planting. On top of that, with simple nursery setting, small scale growers able to harden the tissue-cultured explants on their own, in which reduces the cost of purchasing ready-to-plant seedlings. Eventually, seedlings shortage problem can be solved.

**Significant of Study**

This study highlight on the development of alternative nursery technique for banana tissue-cultured seedlings that are more farmer-friendly, or easily practiced by growers. The success of the study would enable in solving the shortage of planting materials in the market, and potentially increases the survival of banana seedlings grow in farm area, eventually overcome banana shortage problem.

**Objectives**

1. To improve the survival rate of the banana tissue-cultured seedlings in nursery stage.
2. To develop a farmer-friendly nursery technique for tissue-cultured banana seedlings.
Materials and methods
Planting material: 6 to 8 weeks banana *Musa acuminate* cv. Berangan (AAA) tissue-cultured explants

Control method (Conventional / common used method)
The media used is soil:sand:chicken dung at ratio 3:2:1. The media is well mixed and filled into 4"x7" poly-bag till 4/5 full. The filled poly-bag are kept for 1 week and ready to be used. Explants are directly planted into poly-bag. Commercial granular fertilizer (NPK) is applied to the plants every 2 weeks at 3g per plant.

Research method (TisC-Tech)
The nursery are set at two different stages, which are pre-nursery stage and poly-bag nursery stage. There are several experiments tested on both stages to find out the best treatments for acclimatization process.

Pre-nursery stage (Primary Hardening)
The media used are cocopeat:compost at ratio 2:1. Media are mixed well before make into pre-nursery bed at 66cm length x 66cm width x 15cm height (100 explants basis). Planting distance between seedlings is 6cm. Commercial water soluble fertilizer is applied at 3 days interval. Seedlings are kept in this stage till 14 days (extended to 22 days for research data taking purpose) before transplanting into poly-bag stage.

Poly-bag stage (Secondary Hardening)
4"x9" black poly-bag is selected to be used. Media used are cocopeat:compost at ratio 2:1. Media are mixed well before fill into poly-bag at 3/5 full. 14 days seedlings planted in pre-nursery stage are transferred to poly-bag. Commercial water soluble fertilizer is applied at 3 days interval.

Environment setting treatment
Both control and research method are being tested in 2 different nursery environment setting in order to examine the effectiveness and suitability of the methods in different environment condition.

Control environment (conventional / common nursery setting)
Conventional greenhouse (netted house) at 3m in height from the floor (cemented or covered with gravels), with transparent rain shelter on top of the greenhouse and 2 layers of 50% black netting under the rain shelter. 4 side of greenhouse are walled using white gaze. Racks and misting irrigation system (or sprinkler irrigation system) are set in the netted house. The seedlings are placed right on the racks throughout the hardening process.

Research environment
A simple netted house is being placed in farm area, set at 2m in height from the floor, with 1 layer of 50% black netting covered on top and 2 sides, leaving another 2 sides open. The seedlings are placed right on the floor (soil) throughout the hardening process. Irrigation is applied manually using pipe water.
**Data collection**

Data collection parameters included **survival rate**, leaf numbers (only the fully opened leaves), leaf length, leaf width, **plant height** (from the plantlet base to the vertex of the hem of the most recently open leaf), **girth size** (1.5-2cm above from the base), and leaf area (width of the leaf multiple length of the leaf multiple with 0.8).

**Results and discussion**

**Comparison of plant’s growth**

Since the planting techniques are different, the plant growth would be compared at 2 stages, which is in first 22 days after transferred from laboratory to hardening period between conventional method (direct polybag planting, 0th to 22nd days) and research method (pre-nursery stage, 0th to 22nd days), and another 22 days between conventional method (direct polybag planting, 16th to 38th days) and research method (polybag nursery stage, 0th to 22nd days), in both control environment and research environment setting.

**Plant’s height in first 22 days**

![Plant Height Graph](image)

The graph above showing the growth pattern of plant’s height at first 22 days right after transferred from in-situ laboratory to ex-situ laboratory stage, in two different nursery techniques, with two different nursery environment. Based on the growth pattern, it showed that the explants grow in developed technique (TisC-Tech) are slight better than that in conventional technique. Explants grow in conventional technique in on-farm environment (less protective environment) are quite fluctuate whereas more stable in conventional environment. Explants in on-farm environment condition experienced slight transplanting shock at first 12 days but started positive grow after this. Unlike in conventional nursery environment, explants grow in developed technique has no showing obvious transplanting shock after planting. Though, this condition are different from the growing pattern of plant’s girth.
The graph above showing the growth pattern of plant's girth at first 22 days right after transferred from in-situ laboratory to ex-situ laboratory stage. Based on the growth pattern, it showed that the explants grow in developed technique (TisC-Tech) are better than that in conventional technique, even in on-farm environment. Explants start increase girth size since 8th days after transplanting. This positive growth showed that the developed technique could be practice as better alternative hardening technique for tissue-cultured seedlings. Both growth pattern in plant's height and girth using developed technique have shown that the transplanting of seedlings to poly-bag stage could be done after 12 to 14 days in pre-nursery stage.
Based on the graph above, only seedlings grow in conventional technique at conventional nursery environment condition showed obvious fluctuate pattern. On top of that, both conventional technique in on-farm nursery (less protective) environment and conventional nursery environment showed relatively slow grow compared to developed technique (TisC-Tech). Seedlings hardened with developed technique in on-farm nursery show better growth pattern compared to that in conventional nursery environment. This again showed that the developed technique can be practiced in conventional nursery, as well as on-farm nursery.

Plant's girth between 14 to 36 days (overall nursery period)

The plant's girth growth pattern are quite similar as that of plant's height. Only seedlings grow in conventional technique at conventional nursery environment condition showed fluctuate pattern. This again showed that the developed technique (TisC-Tech) can be practiced in conventional nursery, even better in on-farm nursery.
Bar graph above showed the survival rate of seedlings in both different hardening techniques, in 2 different nursery environments. The first 14 days results have proven that the importance of pre-nursery (primary hardening) existence in hardening process. Developed technique (TisC-Tech) has increased the seedling’s survival rate during acclimatization. 82% survival rate at on-farm nursery (less-protective) has proven that hardening of tissue-cultured can be done in much simple and low cost condition.

Conclusion
This research has shown the importance of pre-nursery stage (primary hardening) involving in the overall tissue-cultured seedlings hardening process. The success of seedlings grow in less protective nursery environment enable to reduce the cost of producing seedlings. Besides, this developed technique (TisC-Tech) is considerably easier than conventional nursery techniques. This farmer-friendly technique enable growers to practice and produce their own seedlings by just only purchase explants instead of ready-to-plant seedlings, which is much cheaper in cost. In addition, seedlings grow in on-farm nursery are potentially more adaptable to the farm after transplanting since seedlings has already experienced almost similar environment condition as that in farm, hence potentially solve mortality problem after planting. However, this have to be undergo further study for proven. Enhancement study, fertilization application for example, can be take into consideration to even fasten the overall nursery period of improve greater seedlings growth. In a nut shell, seedlings shortage problem can be solved with this developed technique, in which beneficial for both seedlings growers and banana planters.

References


