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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 increasing year round. Tissue-cultured banana seedlings are getting widely used as planting materials among the growers for 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. The research was conducted to develop a nursery technique that will improve these drawbacks in order to improve greater acceptance and responsive level for the growers towards tissue-cultured seedlings. Based on the results, the developed technique has increased 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 grow faster than that of older seedlings with common quality (15-25cm in height). 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 of 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 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 the production of 101992743 million tons in 2012 (FAO, 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 (FAO, 2012). Banana can be consumed in various ways or produced into different products. It can be eaten as fresh fruits, or processed into the cooked product, such as fried banana or banana cakes, or even processed into the industrial product, for example, banana flours or protein powder. Thus banana achieves exceptional high demands in different industries. However, the current supply could not meet the high demand due to inconsistency and non-systematic planting of banana.

The main problems causing inconsistent planting of banana is the difficulty in getting high-quality free disease planting materials (seedlings) through natural propagation methods. But this problem is overcome by the introduction of micropropagation (tissue culture) techniques to mass produced banana seedlings with assured quantity. With tissue culture techniques, mass production of free disease plantlets can be produced in the shorter period. However, these plantlets are highly susceptible to external environment condition, which requires a critical acclimatization process (hardening) before transplanting to the field. The cost of this seedlings becomes relatively high due to the requirement of laboratory process as well as long acclimatization period, 8 to 12 weeks in common (Lule *et al*, 2013; Robinson and Sauco, 2009[a]). On top of that, the hardened seedlings, with recommended physical requirements (Robinson and Sauco, 2009[ab]) are still susceptible to the field environment after transplanting, leading to high mortality rate.

Macro-propagation techniques are developed for the production of banana planting materials worldwide and being practiced, especially by small-scale farmers (Anonymous, 1955; Pillay and Tripathi, 2007; Ahmad Yusuf and Othman, 2014; Njukwe *et al.*, NA). However, these macro-propagation techniques require a relatively higher quantity of disease-free plant source for mass production as compared to tissue culture micropropagation techniques. The quality of seedlings produced is greatly relying on the quality of planting material source (the vegetative parts of the banana plant). Going down from 1st 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. 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 for 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 the cost of nursery setting, and easily adopted 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 met. With the success in providing enough high quality and quantity of seedlings, banana productions can be done year round and this benefits the rural population in Malaysia and other countries.

Non-systematic planting is due to the difficulty in obtaining an adequate supply of uniform free disease seedlings with high yield potential. The adoption of tissue-cultured seedlings is able to overcome this problem but the survival rate in the early post-planting stage is considerably low compared to conventional seedlings, particularly when handled by small scale and inexperienced growers. One of the potential factors 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 these seedlings is being done in considerably “over-protective” environment condition. The nursery settings have created environmental conditions that are still far different from the actual farm environment conditions. This has caused the low adaptable level of seedlings to survive in farm environment later after transplanting, which leads to stunted growth or mortality. Apart from this, in fact, the survival rate of seedlings in nursery stage also remains an uncertainty, which lies between 50 to 90% depends on techniques used (Au *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) environmental 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 and, in turn, reduces the cost of purchasing ready-to-plant seedlings.

This study highlight on the development of alternative nursery technique for banana tissue-cultured seedlings that were more farmer-friendly, or easily practiced by growers. The success of the study would overcome the shortage of planting materials in the market and potentially increases the survival rate at post-planting. This technique revert farmers negative perception about using tissue-cultured seedlings eventually will enhance the productivity of banana cultivation among the rural population. The objectives of this study were to improve the survival rate of the banana tissue-cultured seedlings in nursery stage and to develop a farmer -friendly nursery technique for tissue-cultured banana seedlings.

RESEARCH METHOD

Planting materials used in this study were 6 to 8 weeks banana *Musa accuminata* cv. Berangan (AAA) tissue-cultured explants. They were grown under conventional technique (control) and Tisc-Tech on-farm nursery technique. For, the conventional technique, the media used was soil:sand:chicken dung at ratio 3:2:1. The media was well mixed and filled into 4"x7" poly-bag till 4/5 full. The filled poly-bags were kept for 1 week and ready to be used. Explants are directly planted into poly-bag. Commercial granular fertilizer (NPK) was applied to the plants every 2 weeks at 3g per plant.

For the TisC-Tech technique, the nursery was set at two different stages, vis.: pre-nursery stage and poly-bag nursery stage. There were several experiments tested on both stages to find out the best treatments for acclimatization process. At pre-nursery stage (the primary hardening), the media used was cocopeat:compost at ratio 2:1. Media was mixed well before making into pre-nursery bed at 66cm length x 66cm width x 15cm height (100 explants basis). Planting distance between seedlings was 6 cm. Commercial water soluble fertilizer was applied at 3 days interval. Seedlings were kept in this stage till 14 days (extended to 22 days for research data taking purpose) before transplanting into the poly-bag stage. At poly-bag stage (the secondary hardening). Media used was cocopeat: compost at ratio 2:1 and mixed well before filling into 4"x9" black poly-bag at 3/5 full. 14 days seedlings planted in the pre-nursery stage were transferred to poly-bag. Commercial water soluble fertilizer was applied at 3 days interval.

Both conventional and TisC-Tech techniques were tested in 2 different nursery environments settings, vis.: conventional greenhouse and simple netted house, in order to examine the effectiveness and suitability of the techniques in different environment condition. The conventional greenhouse was 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. The 4 sides of the greenhouse were walled using white gaze. Racks and misting irrigation system (or sprinkler irrigation system) were set in the greenhouse. The seedlings were placed right on the racks throughout the hardening process. The simple netted house was erected in a farm area and 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 were placed on the floor (soil) throughout the hardening process. Irrigation is applied manually using pipe water.

Data were collected for 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

The growth pattern of seedling height (under two different nursery techniques, with two different nursery environment) for first 22 days right after transferred from in-situ laboratory to ex-situ laboratory stage, is shown in Figure 1. Based on the growth pattern, it showed that the explants grown in developed technique (TisC-Tech) were slightly better than those in the conventional technique. Seedlings grow in conventional technique in the on-farm environment (less protective environment) are quite fluctuate whereas more stable in the 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 was different from the growing pattern of seedling girth.

The growth pattern of seedling's girth at first 22 days right after transferred from in-situ laboratory to ex-situ laboratory stage is shown in Figure 2. Based on the growth pattern, it showed that the seedlings grown in developed technique (TisC-Tech) was better than that in the conventional technique, even in the on-farm environment. Explants start increase girth size since 8^{th} 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 the poly-bag stage could be done after 12 to 14 days in the pre-nursery stage.

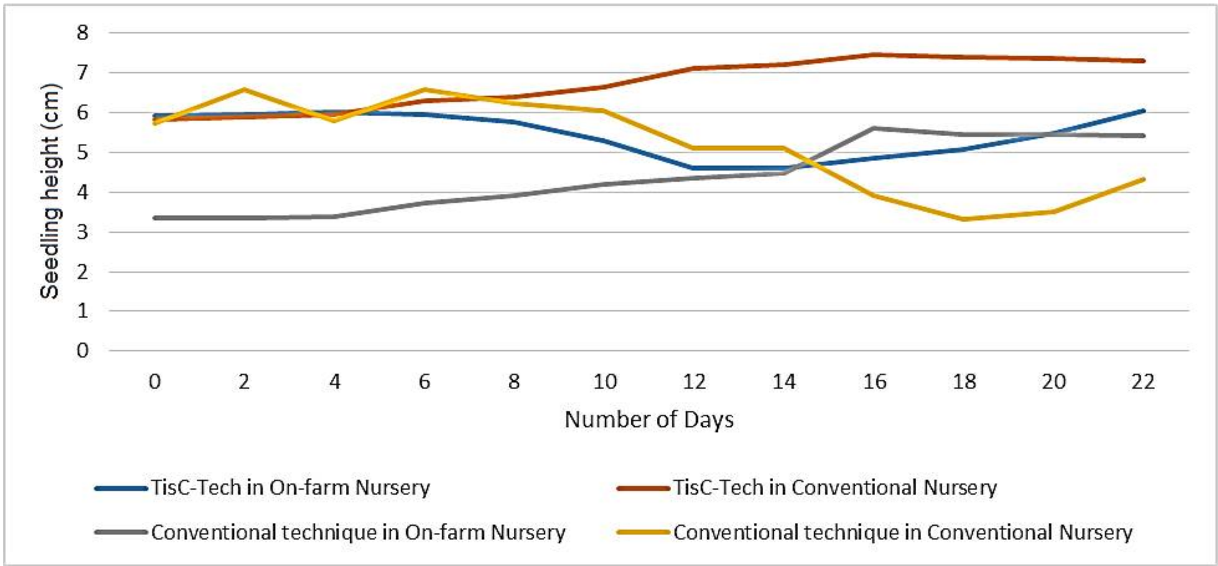

Figure 1. Seedling height comparison in first 22 days

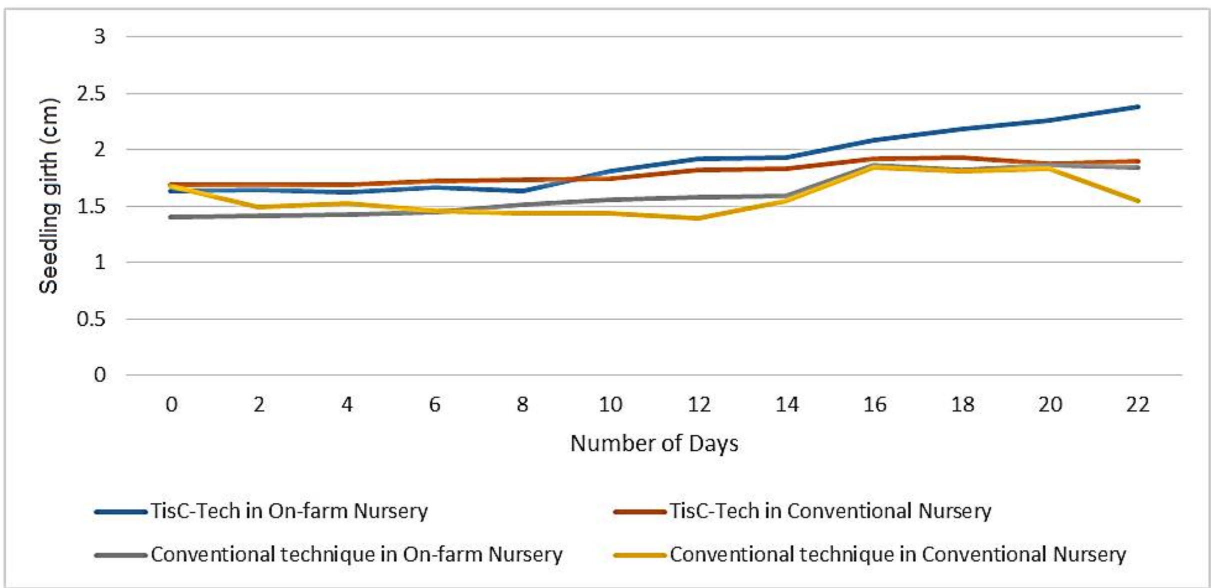

Figure 2. Seedling girth comparison in first 22 days

Figure 3 indicated that only seedlings grown in conventional technique at conventional nursery environment condition showed obvious fluctuation pattern. On top of that, both conventional technique in on-farm nursery (less protective) environment and conventional nursery environment showed relatively slow growing compared to developed technique (TisC-Tech). Seedlings hardened with the developed technique in on-farm nursery show better growth pattern compared to that in the conventional nursery environment. This again showed that the developed technique can be practiced in the conventional nursery, as well as the on-farm nursery.

The seedling girth growth pattern was quite similar as that of seedling's height (Figure 4). Only seedlings grow in conventional technique at conventional nursery environment condition showed fluctuation pattern. This again showed that the developed technique (TisC-Tech) can be practiced in the conventional nursery, even better in the on-farm nursery.

Figure 5 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

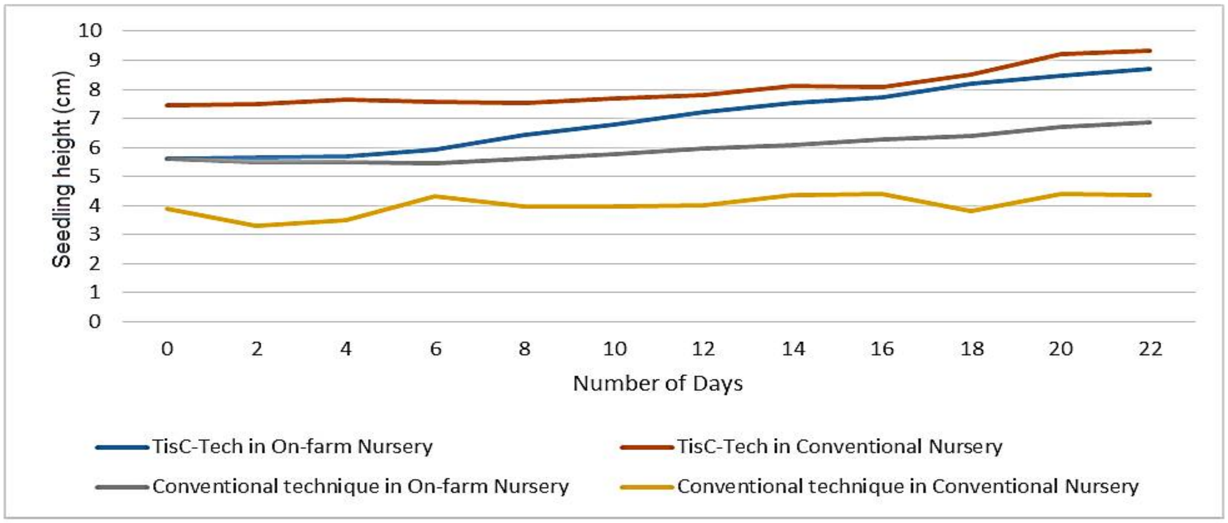

Figure 3. Seedling height comparison between 14 to 36 days

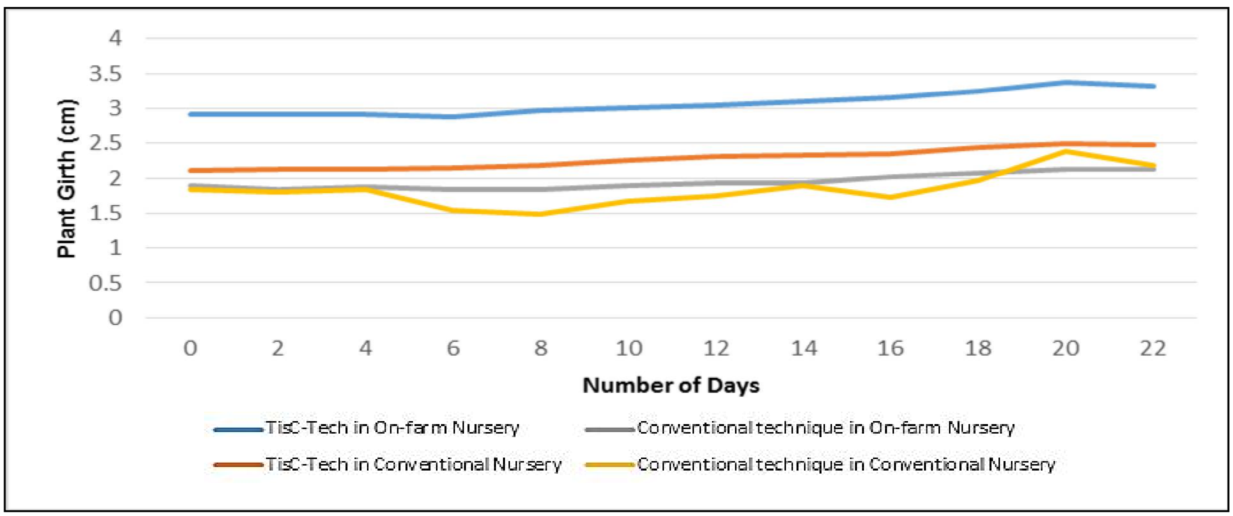

Figure 4. Seedling girth comparison between 14 to 36 days

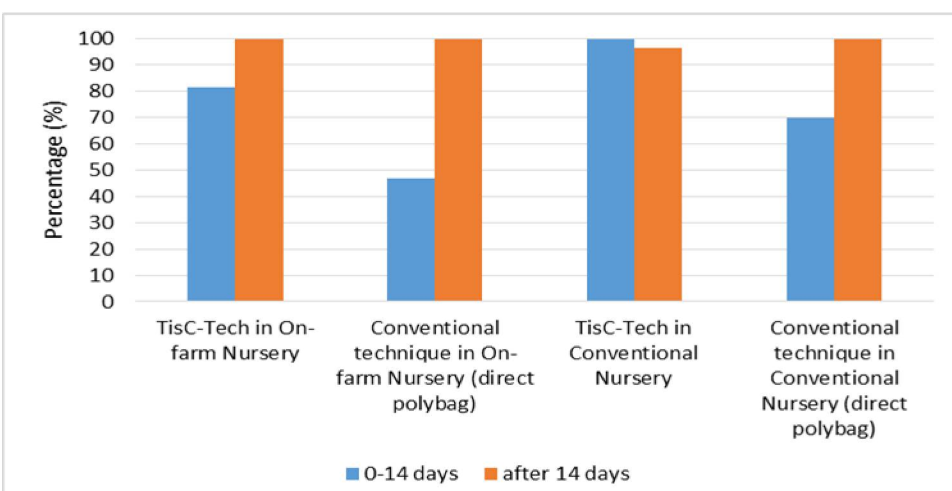

Figure 5. Seedling survival rate comparison in both primary and secondary hardening period

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

These results showed the significant importance of pre-nursery stage (primary hardening) in the overall tissue-cultured seedlings hardening process. The success of seedlings grow in less protective nursery environment would reduce the cost of producing quality seedlings. This developed technique (TisC-Tech) was conveniently practiced than conventional nursery techniques. This farmer-friendly technique enables growers to produce their own seedlings using purchased explants instead of ready-to-plant seedlings, which is much cheaper in cost. In addition, seedlings grown in the on-farm nursery were potentially more adaptable to the farm after transplanting, since the seedlings have already experienced almost similar on-farm environmental condition before planting, hence potentially reduce mortality problem after planting. Enhancement study through fertilization application can further be taken into consideration to even shorten the overall nursery period with improved seedlings growth. In a nut shell, seedlings shortage problem can be solved using this developed technique, hence it is beneficial to both seedlings growers and banana planters.

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