EFFECTS OF PLANT GROWTH REGULATORS (6-BENZYLAMINOPURINE AND KINETIN) ON SHOOT PRODUCTION OF CAVENDISH BANANA

NUR ATIQAH BINTI SAAD

FH 2018 28

EFFECTS OF PLANT GROWTH REGULATORS (6-BENZYLAMINOPURINE AND KINETIN) ON SHOOT PRODUCTION OF CAVENDISH BANANA

By

NUR ATIQAH BINTI SAAD

A Project Report Submitted in Partial Fulfillment of the Requirements for the Degree of Bachelor of Forestry Science in the Faculty of Forestry Universiti Putra Malaysia

2018

DEDICATION

I would like to dedicate this thesis especially to my beloved parents,

"SAAD MD SAMAN"

and

"KHALIJAH AHMAD"

And also to my siblings.

Thank you for all of the things that you've done for me, never get tired and always be there to support and motivate me.

Thank you

ABSTRACT

Cavendish Banana is known as one of the most important food crops for the society throughout the world as it provides various functions and needs especially for the food security. Micro propagation is the best alternative way to produce multiple number of planting materials. The main objective of this study was to investigate the effects of using different cytokinin (6-benzylaminopurine and Kinetin) as plant growth regulators with different concentrations to induce shoot productions on sucker of Cavendish Banana. The explants were cultured on Murashige and Skoog (MS) medium supplemented with five levels of cytokinins concentration (1.0, 2.0, 3.0, 4.0 and 5.0mg/L) and 0.0mg/L as a control; each of it was cultured with five replicates. After 30 days of observations, the highest number of shoots and the length of the longest shoots were observed in MS media with 1.0mg/L concentration of BAP which was higher compared to Kinetin.

ABSTRAK

Pisang Cavendish dikenali sebagai salah satu tanaman buah yang menjadi kepentingan bagi masyarakat di dunia kerana ia membekalkan pelbagai fungsi dan keperluan terutamanya di dalam aspek keselamatan makanan . Mikro organism adalah cara alternatif yang terbaik untuk menghasilkan pelbagai jenis bahan penanaman. Tujuan utama kajian ini adalah untuk mengetahui keberkesanan Sitokinin (6-benzylaminopurine dan Kinetin) dengan kepekatan yang berbeza untuk mendorong produksi pucuk pada sulur Pisang Cavendish. Sampel pisang yang diambil dibiakkan dalam media Murashige dan Skoog (MS) dengan tahap kepekatan sitokinin yang berbeza (1.0, 2.0, 3.0, 4.0 dan 5.0mg / L) dan 0.0mg / L sebagai kawalan; masing-masing dibiakkan dengan lima replika. Selepas 30 hari pemerhatian, bilangan pucuk yang paling tinggi dan panjangnya telah direkodkan. Selepas 30 hari pemerhatian, sample pisang yang dibiakkan di dalam media MS dengan kepekatan 1.0mg / L BAP adalah dicatatkan yang lebih tinggi dalam bilangan pucuk dan panjang pucuk berbanding dengan Kinetin.

ACKNOWLEDGEMENTS

I would like to give my full gratitude to my supervisor Dr. Razak Terhem, for his guidance, supports, never stop to remind and motivates me to be a strong person that enables me to finish this thesis successfully.

A special thanks to Biodiversity Unit, Institute of Bioscience; Mrs. Julia Abd Aziz and her assistant Miss Aina Shafiqah, who allowed and guided me to do the laboratory work for tissue culture efficiently. I've learned a lot during the time I'm being there and I will never forget the kindness they showed.

My dearest friends Suraya Anis, Aina Adlina, Natasha Deanna, Shipa and Wan Rodhiah, thank you for help me in completing my thesis, and for being there during the hardships we've been through. We've done our best doing the thesis and hope the good memories will remain forever.

Last but not least, for my beloved parents and siblings that I'm always grateful for all of their support and understandings. Also, for those who are involved directly and indirectly during the completion of my thesis.

APPROVAL SHEET

I certify that this research project report entitled “EFFECTS OF PLANT GROWTH REGULATORS (6-BENZYLAMINOPURIN AND KINETIN) ON SHOOT PRODUCTION OF CAVENDISH BANANA” by Nur Atiqah binti Saad has been examined and approved as a partial fulfillment of the requirements for the Degree of Bachelor of Forestry Science in the Faculty of Forestry, Univeristi Putra Malaysia.

_____

Dr. Razak Terhem
Faculty of Forestry
Universiti Putra Malaysia
(Supervisor)

_____

Prof. Dr. Mohamed Zakaria Bin Hussin
Dean
Faculty of Forestry
Universiti Putra Malaysia

Date: January 2018

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

g	gramme
L	litre
cm	centimeter
m	meter
mg/L	milligramme/litre
pH	negative logarithm of the hydrogen concentration
min	minute
%	percentage
PGRs	Plant growth regulators
BAP	6-benzylaminopurine
Kin	Kinetin
MS	Murashige and Skoog
HCL	Hydrochloric acid
NaClO	Sodium hypochlorite
NaOH	Sodium hydroxide
Foc	*Fusarium oxysporum* f. sp. *Cubense*
FAO	Food and Agriculture Organization

CHAPTER 1

INTRODUCTION

1.1 General Background

Banana can be labeled as one of the earliest plant cultivation in the aspect of agriculture. The origin of this *Musaceae* family stretches from India to Papua New Guinea which also included Southeast Asia countries (Arvanitoyannis and Mavromatis 2009; De Lange *et al*. 2009). Other than that, it also stated in the Third National Agricultural Policy in the year of 1998 to 2010 that banana has listed as one of the 15 fruit important for commercial cultivation. Combined global production of banana in 2011 was about 145 million tones with a gross production value of US$44.10 billion (FAOSTAT, 2013). Thus, it makes banana as one of the most important food crops in the world.

Banana involved a hybridization and polyploidy between two diploid species, *Musa acuminata* ('AA" genome) and *Musa balbisiana* ("BB" genome) (Heslop-Harrisons and Swarzacher, 2007). There are varieties of banana that exist in the world today and among all of that, the triploid genome groups such as AAA, AAB, and ABB was categorized as the most important.

The popular banana cultivars which also recognized as a dessert banana are called Cavendish banana. It had been cultivated decades ago and still humans need it as a source of fiber, foods, and ornaments plant (Subbaraya 2006; Kennedy 2009). Generally, the cultivated bananas are different from the wild bananas in terms of fruits. Cultivated banana fruit are fully developed which have no seed or some of the species which have a few seeds. High technologies nowadays has made it possible for the banana to transform into varieties with less seed, sterility, oversized pulp, and good development of fruit without the need of fertilization (Ploetz *et al*. 2007; Arvanitoyannis and Mavromatis 2009).

Similar to other plant, the productivity of the banana is also constraint by several biotic and abiotic factors. There are about 80% of world banana productions that has been threatens with Fusarium wilt and Singatoka diseases especially among the banana cultivars (Ploetz, 2005). TR4 or known as Panama disease is caused by the fungus called *Fusarium oxysporum* f. sp. *Cubense* (Foc). This disease caused the banana to wilt and died when the fungus start to spread by going through the xylem vessels from the roots of the plant. The problems occur have a potential to cause a high economic damage and reduce the quality of agricultural crops, especially banana plantation.

People throughout the world are continuously demanding banana as it contains various nutritional benefits such as potassium and vitamins. Banana production is declining as most of the farmers are facing with the fatal diseases of banana that cause the decreased in fruit production. Therefore, one of the way to cope with the problem is to increase the production of saplings using the micro propagation method. It is more favorable rather than the traditional method which consumes more time to grow and have a high risk towards diseases. A single plant or a small plant tissue can be used to increase a production of a free-disease plants and it can be continued throughout the year along with the seasonal changes (Martin *et al*., 2006; Rahman *et al*., 2004).

Plant growth regulators are vital for micro propagation of crop plants; especially cytokinin which helps in plant's shoots induction. Cronauer and Krikorian (1984) cited that the cytokinin contains are depends on the variety of banana and the conditions of the culture. Several types of cytokinin that often used in the research of micro propagation of banana are 6-benzylaminopurine (BAP) and Kinetin. Therefore, the present study was based on the objective; to evaluate shoot production using different cytokinin (6-benzylaminopurine and Kinetin) as plant growth regulators with different concentrations on sucker of Cavendish Banana.

1.2 Problem Statement / Justification

Cavendish banana has been recognized as a high demand of potential crop especially for the market without any problems on the quality of the fruit itself. Local communities are the biggest consumer of banana productions which shows that a good quality of banana cultivar was highly demanded as a source of food, medicinal benefits and also for industrial purposes such as cloth that act as a daily life uses for community. There are many biotic and abiotic factors that are responsible for reducing in yield and production of banana in Malaysia. Cavendish banana is susceptible to infestation by soil borne fungal disease. The fungus *Fusarium oxysporum* f. sp. *Cubense* (Foc) that is responsible to cause highly damaging disease of banana. There is also a concern that was made in the article of the New Scientist which stated that the banana which is the Cavendish could be disappearing in the next ten years (Pearce, 2003).

The traditional way of banana propagation consume more time and will not be able to cope up with the high demands from people. According to Stover and Simmonds (1987), because of the high sterility of the banana varieties, the practice of traditional breeding by the sucker itself is difficult to be done efficiently. In order to overcome the problems mentioned above, a tissue culture technique; micro propagation can be an alternative ways to propagate banana and helps in reducing the attacks by Panama disease and increases the number of its production. A continuous improvement in technological development for producing the alternatives resources is necessary in order to provide the needs

of the world's population (Mohammadi, 2006). Also, there are limited studies about the use of 6-benzylaminopurine and Kinetin with different concentrations on shoot production of Cavendish banana.

1.3 Objective

The objective was to study the effects of using different cytokinin (6-benzylaminopurine and Kinetin) as plant growth regulators with their different concentrations to induce shoot productions on sucker of Cavendish Banana.

REFERENCES

Ali, M. R., Mehraj, H., Jamal Uddin, & A. F. M. (2014). Kinetin (KIN) and Indole 3-Acetic Acid (IAA) on In vitro shoot and root initiation of tuberose. *International Journal of Sustainable Agriculture Technology, 10*(8), 1-4.

Arias D, Dankers C, Liu P, & Pilkauskas P (2003). The World Banana Economy 1985-2002. *Food and Agriculture Organisation of the United Nations, Rome, Italy.*

Arvanitoyannis, I. S., & Mavromatis, A. (2009). Banana cultivars, cultivation practices, and physicochemical properties. *Critical Reviews in Food Science and Nutrition*, *49*(2), 113–135.

Bairu, M. W., Fennell, C. W., & van Staden, J. (2006). The effect of plant growth regulators on somaclonal variation in Cavendish banana (Musa AAA cv. "Zelig"). *Scientia Horticulturae*, *108*(4), 347–351.

Beckman C & Roberts E, (1995). On the nature and genetic basis for resistance and tolerance to fungal wilt diseases of plants. *Advances in Botanical Research*, 21, 35-77.

Bentley, S., N. Moore, J. Pattemore, J. Anderson & K. Pegg. (2001). A DNA – based diagnostic test for 'tropical' race 4 of Fusarium wilt of banana. *Infomusa,* 10,9-10.

Bentley, S., Pegg K.G., Moore N.Y., Davis R.D. & Buddenhagen I.W. (1998) Genetic variation among vegetative compatibility groups of *Fusarium oxysporum f. sp. cubense* analyzed by DNA fingerprinting. *Phytopathology,* 88,1283-93.

Cassells A.C. (1991). Setting up a commercial micropropagation laboratory. In: Bajaj YPS (ed) Biotechnology in agriculture and forestry. Springer, Berlin, (17), 17–3.

Cote, F., Alvard D., Domergue R., Mastache L.N., & Teisson C. (1990). Micropropagation in vitro du bananier. *Fruits,* 45, 112-118.

Cronauer SS. & Krikorian AD (1984). Rapid multiplication of bananas and plantains by *in vitro* shoot tip culture. *Horticulture Science*, 19,234-235.

Dagla & H.R. (2012). Plant Tissue Culture. *Resonance – Journal of Science Education, 17*(8), 759–767.

De Langhe E., Vrydaghs L., Maret PE., Perrier X., & Denham T. (2009). Why bananas matter: an introduction to the history of banana domestication. *Ethnobotany Research and Application*, 7,165–178.

De Langhe EAL (1985). Tissue culture technique for rapid Clonal propagation and storage conditions of Musa (Banana and Plantain). Plant Cell Replication, 4,355-359.

Dita, M. A., Waalwijk, C., Mutua, P., Daly, A., Chang, P. F. L., Corcolon, B. M., … J. Kema, & G. H. (2013). Detecting fusarium oxysporum f. sp. cubense tropical race 4 in soil and symptomless banana tissues. *Acta Horticulturae*, *986*(Table 1), 127–130.

Dita, M., Waalwijk, C. Buddenhagen, I.W. Souza Jr., M.T. & Kema, G.H.J. (2010). A molecular diagnostic for tropical race 4 of the banana Fusarium wilt pathogen. *Plant Pathology,* 59,348-357.

Doreswamy, R., Srinivasa Rao N. K. & Chacko E. K. (1983). Tissue culture propagation of banana. Science Horticulture, 18, 247-252.

Drew, R.A. & Smith M.K. (1990). Field evaluation of tissue cultured bananas in southeastern Queensland. *Australian Journal of Experimental Agriculture,* 30, 569-574.

Escalant, J.V., Tession C., & Cote F. (1994). Amplified somatic embryogenesis from male flowers of triploid banana and plantain cultivars (Musa spp.) *In vitro. Cellular and Developmental Biology*, 30, 181-186.

FAOSTAT, (2013). Food and Agriculture Organization Statistical database. http://www.fao.org/faostat/en/#home

Farahani F, Aminpoor H, Sheidai M, Noormohammadi Z, & Mazinani MH (2008). An improved system for in vitro propagation of banana (Musa acuminate L.) cultivars. *Asian Journal Plant Science*, *7*(1), 116-118.

Fourie, G., Steenkamp E.T., R.C. Ploetz, Gordon T.R., & Viljoen A. (2011). Current status of the taxonomic position of Fusarium oxysporum formae specialis cubense within the Fusarium oxysporum complex. *Infection Genetic and Evolution*, 11,533-42.

Johri B.M. (1982). Experimental embryology of vascular plants, Springer Verlag, Berlin, Heidelberg, New York.

Gamborg O.L, Murashige T., Thorpe T.A, & Vasil I.K, (1976). Plant tissue culture media. *In vitro*, 12, 473-478.

Gaspar T, Kevers C, Penel C, Greppin H., Reid D.M., & Thorpe T.A. (1996). Plant hormones and plant growth regulators in plant tissue culture. *In Vitro Cell Developmental Biology Plant, 32*(4),272–289.

Gordon, T.R. & Martyn R.D., (1997). The evolutionary biology of *Fusarium oxysporum*. *Annual Review of Phytopathology*, 35,111-128.

Hare, P., Cress, W., (1997). Metabolic implications of stress-induced proline accumulation in plants. *Plant Growth Regulation*, 21, 79-102.

Hartmann H.T., Kester D.E., Davies F.T., & Geneve R.L. (1997). Plant Propagation: Principles and practices. 6th Edition. New Jersey: *Prentice Hall, Inc. Englewood Cliffs*, 770p.

Heslop-Harrisons JS & Swarzacher T. (2007). Domestication, genomics and the future for banana. *Ann Botany*, 100,1073–1084

Huang L. & T. Murashige, (1977). Plant tissue culture media: major constituents: their preparation and some applications. *Tissue Culture Associations Manual*, 3, 539-548.

Kennedy J. (2009). Bananas and people in the homeland of genus Musa: not just pretty fruit. *Ethnobotany Research and Application*, 7,179–197.

Khalid, N. (2011). Effect of Benzylaminopurine (BAP) Pulsing on in Vitro Shoot Multiplication of Musa acuminata (Banana) cv. Berangan. *African Journal of Biotechnology*, 10, 2446-2450.

Madhulatha, P., Anbalagan, M., Jayachandaran, S. & Sakthivel, N. (2004). Influence of liquid pulse treatment with growth regulators on In vitro propagation of banana (Musa sp. AAA). *Plant Cell Tissue Organ Cultivation*, 76, 189-192.

Maina, SM., Emongor, Q., Sharma, KK., Gichuki, ST., Gathaara, M. & de Villiers, S.M. (2010). Surface sterilant effect on the regeneration efficiency from cotyledon explants of groundnut (Arachis hypogea L.) varieties adapted to eastern and southern Africa. *African Journal of Biotechnology, 9* (20), 2866 – 2871.

Mantell, S.H., Mathews J.A., & McKee R.A. (1985). Principles of Biotechnology. Blackwell Scientific PubIlcation. Oxford, UK p. 269.

Martin KP, Pachathundikandi SK, Zhang CL, Slater A, & Joseph M (2006). RAPD Analysis of a variant of banana (Musa sp.) cv. Grande Naine and its propagation via shoot tip culture. *In vitro Cellular and Developmental Biology Plant*, 42,188-192.

Meghwal, PR., Sharma, HC. & Singh, SK. (2000). Effect of surface sterilizing agents on in vitro culture establishment of guava (Psidium guajava L.). *Journal of Applied Horticulture, 2* (2), 94-95.

Mohammadi IM. (2006). Agricultural waste management extension education (AWMEE) *The ultimate need for intellectual productivity. American Journal of Environmental Science, 2*(1),10–14.

Murashige T & Skoog F (1962). A revised medium for rapid growth and bioassays with tobacco tissue cultures. *Physiology Plant*, 15,473-497.

Narayanaswamy S. (1994). Plant Cell and Tissue Culture. *New York: Tata McGrawhill Publishing*, 652p.

Omamor, IB., Asemota, AO., Eke, CR. & Eziashi, EI. (2007). Fungal contaminants of the oil palm tissue culture in Nigerian Institute For Oil Palm Research (NIFOR). *African Journal of Agricultural Research, 2* (10), 534-537.

Pearce (2003, January 18). *Going bananas*. Retrieved from https://www.newscientist.com/article/mg17723784-800-going-bananas/

Ploetz R. (2004). Diseases and Pests: A review of their importance and management. *Infomusa*, 13, 11-16.

Ploetz R.C. (2005). Management of Fusarium wilt of banana: A review with special reference to tropical race 4. *Crop Protection,* 73, 7-15.

Ploetz R.C., Kepler AK, Daniells J & Nelson SC (2007). Banana and plantain—an overview with emphasis on Pacific island cultivars, version 1. In: Elevitch CR, editor. Species profiles for Pacific Island agroforestry. Hōlualoa: Permanent Agriculture Resources (PAR).

Ploetz, R.C. (2006). Fusarium wilt of banana is caused by several pathogens referred to as *Fusarium oxysporum f. sp. cubense. Phytopathology,* 96,653-656.

Pradeep, K. P., Zachariah G., Estelittanad S., & Suma A. (1992). Field performance of banana tissue culture plants of variety Nendran (Musa AAB). *South Indian Hon., 40*(1), 1- 4.

Qi Y. X. , Zhang X., Pu J.J., Xie Y. X., Zhang H.Q. & Huang S. L. (2008). Race 4 identification of *Fusarium oxysporum* f. sp. *Cubense* from Cavendish cultivars in Hainan province, China. *Australasian Plant Disease Notes* 3, 46-47.

Rabbani, M.G, Ali, M.H. & Mondal, M.F. (1996). Effect of BAP and IAA on microporpagation of some banana cultivars. *Bangladesh Horticulture, 25*(1&2), 47-52.

Rahman MZ, Nasiruddin KM, Amin MA & Islam MN (2004). *In vitro* Response and Shoot Multiplication of Banana with BAP and NAA. *Asian Journal Plant Science,* 3,406-409.

Sagi L, Gregory DM, Remy S, & Swennen R (1998). Recent developments in biotechnological research on bananas (Musa spp.). *Biotechnology Genetic Engineering Reviews,* 15, 313–317.

Shivas, R. G., Wood, P. M., Darcey, M. W., & Pegg, K. G. (1995). First Record of Fusarium-Oxysporum F-Sp Cubense on Cavendish Bananas in Western-Australia. *Australasian Plant Pathology*, *24*(1), 38.

Simmonds, N. W. (1966). Banana. 2nd Ed. Logmans, London, UK.

Simmonds, N. W., & Shepherd, K. (1955). Taxonomy and origins of cultivated bananas. J. Linn. Soc. Bot. (London) 55.302-312.

Simmonds, N.W. (1960). Bananas (1st Ed.) Longman, London, United Kingdom (U.K.), 466 p.

Smith M., Wiley A., Searle C., Langdon P., Schaffer B., & Pegg K., (1998). Micropropagated bananas are more susceptible to *Fusarium* wilt than plants grown from conventional material. *Australian Journal of Agricultural Research*, 49, 1133-1139.

Snyder W. & Henson H. (1940). The species concept in *Fusarium*. *American Journal of Botany*, 27, 64-67.

Stover RH, & Simmonds NW (1987). Bananas. Longman, London.

Stover, R.H. & Malo, S.E., (1972). The occurrence of Fusarial wilt in normally resistant Dwarf Cavendish banana. *Plant Disease Reporter,* 56, 1000-1003.

Subbaraya U (2006) Potential and constraints of using wild Musa. In: Farmers' knowledge of wild Musa in India. *Food and Agriculture Organization of the United Nations*, pp 33–36.

Vuylsteke DR & Ortiz R (1996). Field Performances of Conventional vs. *In vitro* Propagules of Plaintain (Musa sp., AAB group). *Horticulture Science,* 31,862-865.

Vuylsteke, & D. (1998). Shoot tip culture for the propagation, conservation, and distribution of Musa germplasm. *International Institute of tropical Agriculture, Ibadan, Nigeria*. 82 pp.