

BIOFORTIFICATION OF SOILLESS CULTURE USING BENEFICIAL MICROBES AND COMPOST FOR CULTIVATION OF CHILLI (CAPSICUM ANNUUM) UNDER DEFICIT FERTIGATION

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By

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Thesis submitted to the School of Graduate Studies, Universiti Putra Malaysia, in Fulfillment of the Requirement for the Degree of Master of Science

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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfillment of the requirement for the degree of Master of Science

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Chairman: Mohd Razi bin Ismail, PhD Institute: Institute of Tropical Agriculture

Water and fertilizer are crucial in determining plant growth and production. The use of beneficial microbes is one of several approaches that have potential to be an alternative as plant growth promoters even at under water limited conditions. In the present study, attempts were made to enhance the growth of chilli using *Azospirillum brasilense* Sp7, *Bacilluss sphaericus* UPMB10, *Rhizobium* sp. UPMR31 and mycorrhizal fungi inoculated in the root rhizosphere. Preliminary study were carried out using *Azospirillum brasilense* Sp7, *Bacilluss sphaericus* UPMB10, *Rhizobium* sp. UPMR31 as plant enhancer to evaluate the efficacy of the microbes under serial degrees of deficit fertigation. The results showed increase in yield and plant biomass in inoculated plants compared to non-inoculated plants when subjected to 80 %. 60 %, 40 % and 20 % deficit fertigation. The inoculation effect also increased with amendment of empty fruit bunch into growing media of coconut dust fiber.

Generally with decreasing level of water, greater losses in yield were observed. However, in the second experiment, inoculation of Sp7, UPMR31 and mycorrhiza reduced the extent of growth suppression and the bacterial treated plants accumulated more fruit yield and plant dry weight than the untreated plants especially when compost was added. Besides that, microbial inoculation was capable to maintain the physiological status in plants such as photosynthesis and relative water content. In the 3rd experiment, *Azospirillum brasiliense* Sp7 had increased the growth rate of plants and it could be seen in different stages of growth. Inoculation reduced the antioxidant enzymes activities in inoculated plants but increased the proline content. At the end of the study, it was concluded PGPR is able to increase plant's tolerance to deficit fertigation stress especially with the presence of empty fruit bunch compost as the strategy to increase the efficiency of fertigation management. Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Master Sains

PENGUKUHAN KAEDAH MEDIA TANPA TANAH MENGGUNAKAN MIKROB BERFAEDAH DAN KOMPOS KE ATAS TANAMAN CILI (CAPSICUM ANNUUM) DI BAWAH TEGASAN FERTIGASI

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Air dan baja sangat penting sebagai penentu kepada pertumbuhan dan pengeluaran hasil tanaman. Penggunaan mikrob berfaedah adalah salah satu daripada beberapa pendekatan yang berpotensi menjadi satu alternatif sebagai penggalak kepada pertumbuhan pokok walaupun di bawah keadaan air yang terbatas. Dalam kajian ini, percubaan dibuat untuk meningkatkan pertumbuhan tanaman cili dengan menggunakan mikrob *Azospirillum brasilense* Sp7, *Bacilluss sphaericus* UPMB10, *Rhizobium* sp. UPMR31 dan kulat mikoriza yang diinokulasi di sekitar rizosfera akar. Kajian permulaan dijalankan menggunakan *Azospirillum brasilense* Sp7, *Bacilluss sphaericus* Sp7, *Bacilluss sphaericus* UPMB10, *Rhizobium* sp. UPMB10, *Rhizobium* sp. UPMR31 sebagai penggalak pertumbuhan untuk menilai keberkesanan mikrob tersebut di bawah beberapa paras kekurangan air. Keputusan menunjukkan peningkatan hasil dan biojisim pokok yang diinokulasi dengan mikrob berbanding dengan pokok yang tidak diinokulasi di bawah paras 80 %. 60 %, 40 % dan 20% kekurangan fertigasi.

Kesan penginokulatan juga meningkat dengan tambahan kompos buah tandan kelapa sawit kosong ke dalam media penanaman habuk sabut kelapa.

Secara umumnya dengan mengurangkan kadar pemberian air, penurunan kadar pertumbuhan telah direkodkan. Dalam eksperimen kedua, apabila pokok dirawat dengan Sp7, UPMR31 and mikoriza, tahap ketahanan pertumbuhan pokok dikurangkan dan pokok yang mendapat rawatan bakteria telah meningkatkan hasil buah dan berat kering yang tinggi berbanding pokok yang tidak diberi rawatan dan ia lebih jelas dengan kehadiran kompos. Selain itu, inokulasi mikrob juga mampu mengekalkan status fisiologi seperti fotosintesis dan kandungan air relative dalam tumbuhan. Dalam eksperimen ketiga, *Azospirilium brasiliense* Sp7 dapat meningkatkan kadar pertumbuhan pokok dan ia boleh dilihat di pelbagai peringkat pertumbuhan. Di samping itu, kadar aktiviti antioksida yang rendah didapati pada pokok yang diberi rawatan mikrob tetapi peningkatan kandungan proline. Di akhir kajian, ia dapat disimpulkan bahawa pengunaan PGPR mampu meningkatkan toleransi tumbuhan untuk menghadapi keadaan tegasan fertigasi terutamanya dengan kehadiran kompos tandan kosong kelapa sawit sebagai strategi untuk meningkatkan kecekapan pengurusan fertigasi.

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I certify that a Thesis Examination Committee has met on 08/06/2012 to conduct the final examination of Mohd Fauzihan bin Karim on his thesis entitled "Biofortification of Soilless Culture Using Beneficial Microbes and Compost for Cultivation of Chilli (*Capsicum annuum*) Under Deficit Fertigation" in accordance with the universities and University Colleges Act 1971 and the Constitution of the Universiti Putra Malaysia [P.U. (A) 106] 15 March 1998. The Committee recommends that the student be awarded the Master of Science.

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DECLARATION

I declare that the thesis is my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously, and is not concurrently, submitted for any other degree at Universiti Putra Malaysia or at any other institution.

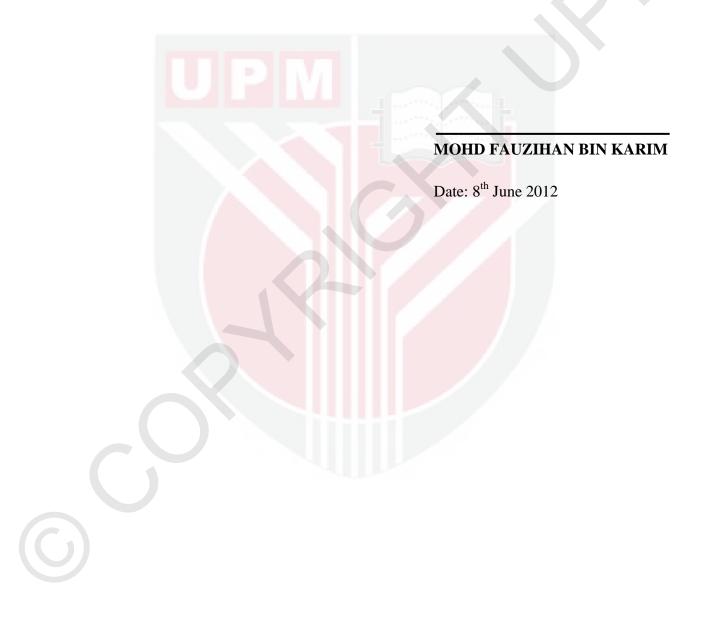


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LIST OF ABBREVIATIONS

ACC	1-aminocyclopropane-1-carboxylate
AM	arbuscular mycorrhizal-fungi
ANOVA	Analysis of variance
APX	Ascorbate peroxidase
ATP	Adenosine triphosphate
cm	Centimetre
CD	Coconut coir dust
CAT	Catalase
DI	Deficit irrigation
DF	Deficit fertigation
DOA	Department of Agriculture
EFB	Empty fruit bunch
EDTA	ethylenediaminetetraacetic acid
FAO	Food and Agriculture Organization
g	gram
GPX	Guaiacol peroxidase
H_2O_2	Hydrogen peroxide
IAA	Indole-acetic acid
IUE	Irrigation use efficiency
μΙ	Microlitre
L	litre
min	minutes
ml	mililitre
mM	Milimoles
μM	Micromoles
PGPR	Plant growth promoting rhizobacteria
RNS	Reactive nitrogen species
ROS	Reactive oxygen species
S	Second
v/v	Volume to volume
WUE	Water use efficiency

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CHAPTER 1

INTRODUCTION

In 2010, the area of chilli cultivation in Malaysia is estimated about 2850 hectares with 32780 tonnes/year or 11.5 tonnes/ha/ year of yield (DOA, 2011). However since 1998, Malaysia has been allocating a large budget to import chilli from other countries in order to meet local demand, and the value is increasing by the years (Appendix 1). Statistical data from FAO (2010) shows that Malaysia becomes one of the major importers of chilli in the world since ten years ago. The simplistic approach to increase crop production is to increase the cultivated area for chilli. However, various constraints such as rapid urbanization, suburban housing and industrialization programs which are heading on agricultural land have disturbed the food production chains from field to the market. In fact, the existing market gardens are also being forced out. The previous government policy which gave priority on industrialization programs has changed the interest of people from agriculture to manufacturing sector.

Currently, most of the cultivated area for vegetables is soil-based system and widely used by growers. Nevertheless due to the difficulty to get good soil quality and risk of soil-borne pathogens has slowly force the farmers to find other alternative such as soilless culture media which using non-soil materials as growing medium. Soilless culture media are commonly cultivated under protected environment and it is still scarce in the country. In Malaysia, the system only covered small portion from 86000 hectares of total vegetables production area in 2010 (DOA). Advancement in the knowledge of rhizosphere manipulation and technologies in soilless culture will ensure the continuality of yield production and may promote the system to be used widely. However, beginning cost for glass house and rain shelter structures are always become controversial. Rising cost for variable inputs especially fresh water and chemical fertilizers also requires a large investment and because of that, more effective methods need to be discovered.

Nowadays, environmental stresses present the most limiting factors to food productivity. Environmental stress impact not only on crops which are presently being cultivated, but also become significant barriers to the introduction of crop into new land. Water deficit is one of the most important environmental factors limiting crop productivity. Water deficit develop when water loss by transpiration exceeds absorption by root. Plants generally experience some degree of water deficit in the open field or under protected environment.

Rhizobacteria symbiotically colonize plant roots and consume root exudates and lysates (Antoun and Prevost, 2006; Pieterse *et al.*, 2002). Certain strains are referred to as plant growth-promoting rhizobacteria (PGPR), which can be used as biofertilizers (Kennedy *et al.*, 2004). The PGPR can directly benefit plant growth by increasing nitrogen uptake through nitrogen fixation process, synthesis of phytohormones, solubilization of minerals, and iron chelation (Bowen and Rovira, 1999). Some PGPR may suppress soil-borne pathogens by producing siderophores, antimicrobial metabolites, or competing for nutrients and/or niches (Nelson, 2004). Indirectly, some PGPR stimulate an increase in resistance to pathogens and pests that feed on leaves by activating the formation of physical and chemical barriers in the host, a phenomenon referred to as induced systemic resistance (Bostock, 2005; Persello-Cartieaux *et al.*, 2003; Ryu *et al.*, 2003).

Many plants respond positively to inoculation with bacteria and mycorrhiza. Legumes are well-known example of plant benefiting from inoculation by its symbiotic partner, rhizobia, and have been exploited in many parts of the world (Shamsuddin *et al.*, 1988). Mycorrhiza too has been reported to benefit many different species of plant crops. Meanwhile, plant growth promoting rhizobacteria (PGPR) have been shown to benefit vegetables and cereal crops (Okon and Labandera-Gonzales, 1994). These PGPR include *Azospirillum*, *Pseudomonas*, *Bacillus* and *Agrobacterium* species. The mechanism to promote plant growth include fixation of atmospheric dinitrogen, production of indole acetic acid (IAA) ad production of siderophores (Kloepper *et al.*, 1980). In addition, since ten to fifteen years ago, PGPR and mycorrhiza have been previously used as a part of plant defence mechanism against various types of environmental stresses especially water deficit and drought.

1.1 Objectives

The objectives of this study were:

- 1. To evaluate the effectiveness of selected microbes on growth and yield of *Capsicum annuum* under different level of fertigation regimes.
- 2. To evaluate the effect of empty fruit bunch compost amendment on plant growth, physiological activities and nutrient status.
- 3. To determine the catalase, ascorbate peroxidase, guiacol peroxidase and proline activities in plant tissue in response to deficit fertigation.

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