



UNIVERSITI PUTRA MALAYSIA

**GROWTH AND YIELD RESPONSES OF TOMATO
(LYCOPERSICON ESCULENTUM Mill) TO TRICHODERMA
INOCULANTS IN SOILLESS MEDIUM**

FRANKLIN RAGAI KUNDAT

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By

FRANKLIN RAGAI KUNDAT

**Thesis submitted to the School of Graduate Studies, Universiti Putra Malaysia
in Fulfilment of the Requirements for the Degree of Master of Agricultural Science**

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Toward the end of the thesis write-up, I usually sit down with a glass of cold water and write a thank you to all of the people who helped make this happen. Some are in the line of plant pathology and others have been pulled into it because I live this thesis 25 hours a day. It's difficult to express in short bursts how grateful you are; for the endless hours spent by my brother, sisters and friends towards the completion of this thesis, to my Mum and Dad who let me run off to chase the dreams of a boy only to come home man again and to Sha for a lifetime love and inspiration, you truly are an angel. There are so many people all around to thank, the lecturers, the staffs, the students .I could go on. You see, it's hard to say so much in so little to so many But, if a man could be judged by the friends he keeps, I must be the luckiest man in town.



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February 2002

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Growth and yield responses of tomato to *Trichoderma* inoculants in Soilless medium (mixture of coconut dust and peat) (Cdp) under Protected Environment Agriculture (PEA) system was attempted. Two *Trichoderma* species; *T. harzianum* (UPM 29) and *T. virens* (UPM 23) singly and as mixtures were used in the study. The physical, chemical and microbiological properties of Cdp was suitable for growth of tomato and proliferation and survival of *Trichoderma*. Cdp has good pore size with bulk density within the range of mineral soil (1.0 – 1.8 g/L). Cdp has high water availability of 22.23%. However they has low microbial populations comprising of mainly the genera *Aspergillus* and *Penicillium*.

The effect of soilless media on proliferation and survival of *Trichoderma* was carried out by either adding the inoculant as an additive to the soilless medium or as bio-seed treatment. *T. harzianum* population in Cdp was reduced by 32% after 15 days of application. However, population for *T. virens* when applied singly or as mixtures of *T.*



harzianum + *T. virens* dropped at day 6 but remained stable thereafter until the end of the experiment. *Trichoderma* population was higher on the roots throughout the 21 days of experimental period, suggesting that *Trichoderma* can colonize the germinating roots and live on the root exudates. Application of *Trichoderma* as additives to the soilless medium was a better delivery method as it gave better distribution and easy contact with the growing roots. Therefore, application as additives to germination mixes was selected to evaluate the effect of air-dried preparation of *Trichoderma* inoculants on growth and yield responses of tomato.

Trichoderma inoculants at the rate of 30×10^6 cfu/g dry weight of Cdp have a significant effect ($P < 0.05$) on seed germination and seedling emergence of tomato. Seedling emergence was more than 98% for all *Trichoderma* treatments. Initial establishment and growth of seedlings were better in *Trichoderma* amended Cdp, as shown by high value of stomatal conductance, net photosynthesis and chlorophyll content. However there were no significant difference in vegetative growth and peroxidase activity between treatments, probably due to the influence of plant spacing on light interception and dilution effect of *Trichoderma* inoculant in Cdp with time. Tomato plants grown in *Trichoderma* amended Cdp showed significantly higher fruit production and total fresh weight of fruits as compared to control. The plants were supplied with full strength Cooper solution at rate of 600 ml/day via drip irrigation system



Abstrak thesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk Ijazah Master Sains Pertanian

RESPONS TUMBESARAN DAN HASIL TOMATO (*LYCOPERSICON ESCULENTUM* Mill.) TERHADAP INOKULUM *TRICHODERMA* DI DALAM MEDIA TANPA TANAH

Oleh

FRANKLIN RAGAI KUNDAT

Febuari 2002

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Respons tumbesaran dan hasil tomato terhadap inokulum *Trichoderma* di dalam media tanpa tanah (campuran sabut kelapa dan peat) (Cdp) di bawah system Pertanian Persekitaran Terkawal telah dijalankan. Dua jenis spesies *Trichoderma*; *T. harzianum* (UPM 29) dan *T. virens* (UPM 23) secara beasingan dan gabungan digunakan di dalam kajian ini. Sifat-sifat fizikal, kimia dan mikrobiologi Cdp menunjukkan bahawa ia sesuai untuk pertumbuhan tomato serta pembiakan dan keupayaan hidup *Trichoderma*. Ia mempunyai saiz ruang rongga yang baik dengan ketumpatan pukal berada di dalam lingkungan tanah mineral (1.0 – 1.8 g/L). Cdp mempunyai peratus kandungan air tersedia yang tinggi iaitu 22.23%. Walaubagaimanpun ia mempunyai populasi mikrob yang rendah terutamanya terdiri daripada genera *Aspergillus* dan *Penicillium*.



Kesan Cdp ke atas pembiakan dan keupayaan hidup *Trichoderma* telah dijalankan sebagai tambahan kepada media tanpa tanah dan rawatan biji benih. Populasi *T. harzianum* di dalam Cdp telah menurun sebanyak 32%, 15 hari selepas aplikasi. Walaubagaimanapun, bagi populasi *T. virens* yang digunakan sendirian atau campuran *T. harzianum*+ *T. virens* menurun pada hari ke 6 dan stabil sehingga ke penghujung eksperimen. Populasi *Trichoderma* adalah tinggi pada akar sepanjang 21 hari tempoh ujikaji, menunjukkan bahawa antagonis tersebut boleh mengkoloni akar yang bercambah dan boleh hidup pada cecair rembesan akar. Penambahan inokulum kepada media tanpa tanah memberi taburan yang lebih baik dan mudah untuk bersentuh dengan akar. Dengan itu penambahan kepada media tanpa tanah dipilih sebagai kaedah untuk menilai kesan inokulum *Trichoderma* terhadap tumbesaran dan hasil tomato.

Trichoderma pada kadar 30×10^6 cfu/g berat kering Cdp mempunyai kesan yang beerti ($P < 0.05$) terhadap percambahan biji benih dan kemunculan anak benih. Peratus percambahan anak benih adalah melebihi daripada 98% bagi semua rawatan *Trichoderma*. Pertumbuhan peringkat awal anak benih adalah lebih baik dalam media yang digabungkan dengan *Trichoderma*, sebagaimana yang ditunjukkan oleh konduksi stomata, kadar fotosintesis bersih dan kandungan klorofil. Walaubagaimanapun tiada perbezaan beerti pada pertumbuhan vegetatif dan aktiviti peroxidase di antara rawatan yang mungkin disebabkan oleh pengaruh jarak tanaman terhadap kemasukan cahaya pada tomato dan kesan pencairan inokulum *Trichoderma* di dalam media mengikut masa. Pokok tomato yang ditanam di dalam medium Cdp dan dirawat dengan *Trichoderma* menunjukkan peningkatan dalam pengeluaran buah dan jumlah berat bersih buah jika dibandingkan dengan rawatan kawalan. Pokok tomato diberi baja Cooper solution yang mencukupi iaitu pada kadar 600 ml/hari melalui sistem pengairan titis.

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CHAPTER 1

INTRODUCTION

Peri-urban vegetable production, which occurs in the 'green belt' zones surrounding large population centres in this country, is the primary source of perishable and off-season fresh vegetables for the urban dwellers. However, the demands for high quality and safe vegetables (*pesticides-free*) by this burgeoning urban population poses major challenges for agricultural research.

The simplistic approach to increase vegetable production is to increase the cultivated area for vegetables. However, various constraints exist. The rapid urbanization and suburban housing and industrialization development has placed a premium on agricultural land, and even existing market gardens are being forced out. Currently, the government's policy on industrialization has seen tremendous changes to switch the interest from agriculture to the manufacturing sector.

Utilization of soilless medium in vegetable production in Malaysia at the moment is rather scarce. At present, soil-based growing medium are widely used by the growers. Difficulty in getting good quality topsoils and variation in its quality may promote utilization of non-soil materials such as soilless culture systems using organic-based media. Organic-based media such as rice husk, cocomix coir, sawdust and peat are widely used as growing medium in the soilless culture systems (FAO,1990). Utilization of these materials are considered environmentally sustainable, and has attracted interest among research scientists and horticulturists worldwide.



The movement of commercial greenhouse operations away from soil-based growing medium to soilless culture has meant that the greenhouse crops are initially free of soil-borne pathogens (FAO, 1990). However, plants that are grown in these soilless medium also have lower populations of microorganisms in the rhizosphere including those beneficial for plant growth or disease suppression. Opportunity exists, then, to introduce specific beneficial microorganisms into the soilless environment.

The manipulation of the microbiological communities in the rhizosphere of crop plants for increasing yields and the biological control of diseases has been extensively studied in field crops and greenhouse crops (Menziez and Ehret, 1997). Formulations have been developed and tested for disease suppression and for promoting the growth of plants. An extruded granular formulation with biomass of *Gliocladium virens* and *Trichoderma spp.* significantly reduced the severity of damping-off of eggplant caused by *Rhizoctonia solani* and saprophytic growth of the pathogen in soilless mix (Lewis and Larkin, 1997).

Heemert and Veenstra (1997), found that introduction of *T. harzianum* strains as granules and wettable powder respectively has shown significant results, both as plant growth promoter in several crops, increased in the development of the root system and in prevention against certain root diseases of greenhouse crops. Combination treatment of a fungal antagonist *G. virens* and a bacterial antagonist *Bulkolderia cepia* resulted in improvement in disease severity and fresh weight for pepper and fruit yield for tomato in the field (Fravel and Larkin, 1997). The efficient use of natural resources for vegetable

production was also a move towards developing a production system, which is safe, sustainable, and environment friendly.

This research focused on the microbiological activity of the rhizosphere of greenhouse vegetable plants growing in soilless medium. In the study presented here, tomato (*Lycopersicon esculentum*, L) an economic important food crop is chosen as plant material for trials in the laboratory and greenhouse. *T. virens* (UPM 23, IMI 378843) and *T. harzianum* (UPM 29, IMI 378842) were used as the inoculants. The objectives of this study will include:

- (1) Determination of the physical, chemical and microbial properties of soilless medium.
- (2) Proliferation and survival of *Trichoderma* inoculants in soilless medium.
- (3) Growth responses of tomato plants to *Trichoderma* inoculants.

CHAPTER 2

LITERATURE REVIEW

Tomato Cultivation in Malaysia

The area planted to tomato is slightly more than 500ha (Mardi, 1992). The two most popularly grown lowland tomato varieties are Banting and Local White, which can yield up to 17t/ha. Among more than 50 types of vegetables grown, tomato is among the most expensive to produce. This crop is mainly grown for fresh consumption. Pests and diseases are the major production constraints, apart from marketing, irrigation, infertile soils and unavailability of good planting materials. In the year 2000, Malaysia is importing 5,120 MT of tomato fruits, which is worth RM 4.8 million (Department of Statistics Malaysia, 2001).

In terms of total area of production, tomato ranks seventh among other high value vegetables (Ministry of Agriculture Malaysia, 1999). A total of 500ha were planted to tomato in 1985, producing 9,469 t of fresh tomatoes (Ministry of Agriculture Malaysia, 1996). The high cost of production might be one factor limiting area expansion.

Lowland tomato production in Malaysia is entirely in the hands of small holders. Among the popularly grown varieties are Banting, Local White, MT I, MT II, King Kong and FG 46. An average yield of 15t/ha is obtained by lowland farmers compared to 35t/ha obtained in the highlands (Anon, 1987). Tomato is often intercropped with other vegetables and many farmers practice crop rotation with no fixed pattern.

The majority of farmers save their own seeds from one planting season to another. Only a small amount of seed is imported, mainly from Taiwan and Thailand. A breeding program was initiated by MARDI and have produced MT I and MT II, two varieties that are suitable for lowland cultivation (MARDI, 1992).

Consumption of fresh tomato in this country is relatively high. Most tomatoes produced are marketed as fresh fruit. The marketing pattern is farmers - collectors - truckers - wholesalers - retailers -consumers. By far the most prevalent is selling by consignment in which tomatoes are consigned by the farmers through local collectors for sale at the terminal market with prices determined at the market.

Problems and Constraints

Pests and diseases are the most serious production constraints. Bacterial wilt (*Ralstonia solanacearum*), early blight (*Alternaria solani*), black leafnold (*Cercospora fuligena*), anthracnose (*Collectotrichum capsici*), *Sclerotium* wilt (*Sclerotium rolfsii*), and viruses such as tomato mosaic virus have been reported on tomato in Malaysia (Mardi, 1995). Another serious production constraint besides pests and diseases is the unavailability of suitable soils since most have been utilized for plantation crops. Only marginal land is usually available for vegetable production and high inputs and management skills are needed to produce the crop economically.

Growing Medium

Peat

Peat is an important natural resource comprising highly of modified plant remains that accumulate in certain wet habitats. It is used primarily as a fuel and as a horticultural medium (Molitor and Bruckner, 1997). Peat has many properties that make it valuable to horticulturists. Works at research stations, and general experience in many countries have shown that chemical and physical properties of peat endow it with great advantages for vegetable production (Molitor and Bruckner, 1997). However the physical and chemical properties of peat depend primarily on the nature and origin of the plant remains of which it is composed and their degree of decomposition. These two criteria together form the basis for a broad practical evaluation (Puustjarvi, 1974, Robertson, 1993).

Properties of peat are mainly associated with its degree of decomposition and textural and structural characteristics. Peat varies in quality depending on source, season, harvesting method, factory processing and even container load and storage (Bunt, 1988, Handreck *et al.*, 1991). Peat has a fairly good base exchange and buffering capacity and a very high water holding capacity besides being a structure that allows good aeration and resistance to decomposition (Handreck *et al.*, 1991). Nevertheless, most peats, are naturally very low in essential plant nutrients due to its low pH value. Therefore, liming and fertilization are necessary (Lucas *et al.*, 1971). The fertilization formulation used in peat will depend greatly upon the nature of the crop.

Peats in greenhouse plant production can be used alone as a growing medium. Peats are often mixed, however, with either mineral soil or soil substitutes such as

vermiculite, perlite and coconut dust (Yahya and Razi, 1997). Mixes containing substitute materials have become popular in some countries because of the uniformity, good physical properties, low density and few weed and soil-borne pathogen problems. Peat is also suitable for improving the difficult soils such as light sand and heavy clays (Lucas *et al.*, 1971). Most foliage plants are grown using peat based soilless media (Olli *et al.*, 1993). Therefore, peat is a valuable asset in many facets of protected cropping systems.

Coconut Coir Dust

There is a great deal of interest in the use of coconut coir dust or commercially known as cocopeat as a growing medium (Wever *et al.*, 1994). Utilization of cocopeat is considered environmentally sustainable and has attracted interest among research scientists and horticulturists worldwide (Martinez *et al.*, 1996). Sri Lanka, which produces more than 2.5 billion coconut fruit each year, has become the leading processor of coconut coir dust into a form sustainable for horticulturist use (Handreck, 1993).

Cocomix is the name given to the fibrous material that constitutes the thick mesocarp tissue, or husk, of the coconut fruit. The husk contains 60%-70% pith tissues, with the remainder being fiber of varying lengths. After grinding the husk, the long fibers of the coir are removed and used for various industrial purposes. Traditionally, the short fibers (2mm or less) and pith left behind have accumulated as a waste product or commonly referred to as waste grade coir. The waste grade coir may be screened to remove part or the entire fiber, and the remaining product is referred to as coirdust as for which no industrial use has been discovered (Verdonck *et al.*, 1996). Coir dust is light to

dark brown in colour and consists primarily of particles in the size range 0.2-2.0mm (75%-90%).

There is a great interest in the use of coir as a growing medium (Wever *et al.*, 1994). Published results using coir dust as a growing medium or as soil amendment have reported good results (Merrow, 1994). Prasad (1996) concluded that coirdust in many cases has suitable physical, chemical and biological properties to be used as growing medium. He added that physical properties could be adjusted to meet the aeration requirements of crops by adding coconut fibre to the coirdust. Cresswell (1992) confirmed that it has superior structural stability, water absorption ability and drainage, and cation exchange capacity compared to either sphagnum peat or sedge peat. Apart from that it is hydrophilic to sphagnum peat and rehydrates readily. Coirdust seems to be an acceptable substitute for sphagnum peat or sedge peat in soilless container media, although nutritional regimes may need to be adjusted on a crop-by-crop basis (Merrow, 1994). Utilization of peat-coir mixes as growing medium has been analyzed in some reports (Merrow, 1994; Argo *et al.*, 1996). The responses of crops to a specific mixture of coirdust-based medium vary depending on plant species (Yahya and Razi, 1997).

Trichoderma spp.

Trichoderma belongs to the mitosporic (*Hyphomycetes*) (Samuels, 1999). The colony grew well on potato dextrose agar (PDA) with smooth, white and clear surface at first, later turning into greenish colour. It is normally classified as imperfect fungi that produced one-celled conidia or chlamyospore in single or ellipsoid, with phialospore at the end of each phialide. Asexual spores are produced on hyphae freely exposed to the