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Bacteria from an Oil Palm Agricultural System and Their Interactions with *Ganoderma boninense* and *Trichoderma harzianum*

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ABSTRAK

Kulat Trichoderma harzianum *(pencilan FA 1132) telah mempamerkan potensi sebagai agen kawalan biologi terhadap patogen kelapa sawit,* Ganoderma boninense. *Kulat ini kini sedang dihasilkan pada skala besar sebagai kompos-Trichoderma untuk ujian di lapangan. Kajian ini telah memencilkan bakteria daripada kompos tersebut dan juga daripada rizosfera kelapa sawit. Kesemua bakteria pencilan disaring untuk tindak balas antagonis terhadap* G. boninense, *selepas itu stren-stren terpilih akan diuji untuk keserasian dengan FA 1132. Empat daripada enam spesies bakteria yang dipencilkan telah menunjukkan aktiviti fungistatik terhadap* G. boninense. Corynebacterium urealyticum *memberi peratus perencatan radius pertumbuhan (PIRG) paling tinggi pada 86.6%, tetapi stren ini menunjukkan ketidakserasian dengan FA1132. Kecuali* C. urealyticum, *kesemua pencilan bakteria menunjukkan keserasian dengan FA 1132. Kajian ini merumuskan* Chromobacterium violaceum *dan* Burkholderia cepacia *masing-masing mungkin mempunyai keupayaan untuk bertindak terus sebagai inokulan tersendiri untuk merencatkan pertumbuhan* G. boninese *di samping juga mempunyai potensi digunakan sebagai ko-inokulan di dalam konsortium bersama* T. harzianum *FA 1132, untuk peningkatan keberkesanan di dalam sesuatu formulasi agen kawalan biologi.*

ABSTRACT

The fungus Trichoderma harzianum *(isolate FA 1132) has shown potential as a biological control agent of the oil palm pathogen,* Ganoderma boninense. *It is currently being produced on a large scale as Trichoderma-infused compost for field trials. This study isolated bacteria from such a compost as well as from oil palm rhizospheres. They were screened for antagonistic reactions towards* G. boninense *after which the selected strains were tested for compatibility with FA 1132. Four out of six species of bacteria showed fungistatic activity towards* G. boninense. *Corynebacterium urealyticum gave the highest percentage inhibition of radial growth (PIRG) at 86.6%, but was incompatible with FA 1132. Except for* C. urealyticum, *all the other isolates showed compatiblity with FA 1132. This study deduced that* Chromobacterium violaceum *and* Burkholderia cepacia *may have the capacity to act as individual inoculants to inhibit the growth of* G. boninense, *as well as have the potential to be used as co-inoculants in consortium with* T. harzianum *FA 1132, for enhanced performance in a biological control formulation.*

INTRODUCTION

Ganoderma boninense (Pat.) is a fungal pathogen which causes basal stem rot (BSR) of oil palms. Chemical treatments carried out on a trial basis on infected palms showed somewhat limited success (MPOB 2003) and the disease still persists in regions where the palms are grown. Ilias and Abdullah (1998) conducted an *in vitro* screening of soilborne fungi and found one strain of *T. harzianum* and *T. virens*, that showed promise as biocontrol agents of *G. boninense.* Repeated greenhouse trials using *T. harzianum* (isolate FA 1132) as a potential biocontrol agent of BSR confirmed the efficacy of this strain (Ilias 2000; Abdullah *et al.* 2003; Nagappan 2005; Sundram 2005).

Strains of bacteria also do behave as biocontrol agents. Among others, *Bacillus*

subtilis have been investigated for the biocontrol of Cercospora leaf spot in sugar beet (Collins and Jacobsen 2003); against the pathogenic *Fusarium oxysporum* (Knox *et al.* 2000) and against crown rot of tomato in the field (Nemec *et al.* 1996). Although some bacteria are used individually as biocontrol agents, many others are used as synergistic co-inoculants in a biocontrol system (Bennet *et al.* 2003; De Jensen *et al.* 2002; Knox *et al.* 2000). Many bacteria-based biocontrol products are already commercialized; among them is Deny® which has *Burkholderia cepacia* in its basic formula and is used against the fungal pathogens *Fusarium*, *Pythium* and *Rhizoctonia*. *Burkholderia cepacia* is again found in Leone®, which is used against *Botrytis* and *Phytophthora* diseases of potatoes as well as *Septoria* on wheat (Driesche and Bellows 1996; Fravel *et al.* 1998; McPartland *et al.* 2000; Khetan 2001). Products that contain *Trichoderma* as well have been formulated for commercial purposes. An example is Trichodex™, which contains *T. harzianum* as the active ingredient, is used against post harvest rot of apple. Another product, Binab-T™, which is a combination of *T. harzianum* with *T. polysporum*, is used in the control of wood rots (Samuels 1996).

To date and to our best knowledge, no studies have yet been conducted where a bacterial strain is used against *G. boninense*; it is thus one aspect worth exploring. Another unexplored avenue is the use of a compatible bacterial strain as a co-inoculant to a known fungal biocontrol agent for enhanced synergistic performance.

Nagappan (2005) reported that *T. harzianum* (isolate FA 1132) performed best when applied individually to artificially-infected oil palm seedlings. However, when the isolate was applied as 1:1 combination with *T. longibrachiatum*, or 1:1 combination with *T. virens*, or 1:1:1 combination of all 3 species, the control performance declined significantly. Sundram (2005) also reported that strain FA 1132 exhibited excellent efficacy when used individually in greenhouse trials, but it performed poorly when used as a 1:1 combination with another strain of *T. harzianum* (FA 1166). Thus the possibility of using bacterial strains compatible with FA 1132 instead of in combination with members of the genus *Trichoderma*, is one avenue pursued in this investigation.

The first objective of this study was to isolate and identify resident bacteria from two sources, namely soils from several oil palm rhizospheres and samples from a *Trichoderma*–infused compost. The latter is the end-product of a pilot scale trial in the production of a *Trichoderma*-based biocontrol product, for application onto mature palms infected with *Ganoderma* (Abdullah *et al.* 2005). The second objective was to screen and select for bacterial isolates that showed antagonistic properties towards *G. boninense*. The third was to screen the selected antagonistic bacteria for compatibility with *T. harzianum*, strain FA 1132. It is expected that from this study, a bacterial species that simultaneously showed an acceptable degree of antagonistic intimidation towards *G. boninense* and good compatibility with *T. harzianum* will be found. A combination of these two characteristics in a bacterial strain may give added value to its possible use as a co-inoculant in consortium with FA1132 in the end-product, for use against *G. boninense*.

MATERIALS AND METHODS

Sampling Site

Bacteria were isolated from two sources, namely from oil palm rhizospheres and from 25 kg packed bags of *Trichoderma*-infused compost meant for biocontrol trials. Both sources were from Sedenak, Johor. A custom–made soil auger was used to collect the soil samples, taken at 15 cm depth and from four cardinal points per palm, for eight randomly-selected palms. About 200 g of each composite sample per source was obtained and put into labeled containers and brought back to the laboratory for processing.

Isolation of Bacteria

Composite samples for each source were prepared by pooling and mixing for homogenous distribution, after which a stock solution of 10:100 (w:v) sample to water was made. The solutions were maintained as shake cultures at 100 rpm, under ambient laboratory conditions of temperature (28° ± 2°C) and light (12 hours light, 12 hours darkness). An aliquot of 1ml solution was subjected to a ten-fold serial dilution, from which 0.5ml at dilution 10^{-8} was dispensed onto a Nutrient Agar (NA) culture plate. An L-shaped glass rod was used to spread the cultures evenly and the plates were then incubated for 48 hours under ambient conditions. The morphologies of each colony-forming unit (CFU) that emerged were recorded to aid species characterisation. The bacteria from each colony type was then streaked onto fresh NA and incubated for 48 hours to obtain pure single colonies.

Biolog® *Identification of Bacteria*

Single colonies obtained from the isolation process were subjected to a series of tests before placing on the Biolog® plate reader for species identification. Bacterial isolates were picked up with a sterile loop and stirred into 1 ml of KOH solution. Samples that agglutinated to the loop indicate Gram Negative (GN) bacteria, while those that did not, were Gram Positive (GP). The GN samples were next subjected to an oxidative test wherein a loopful of bacteria was mixed into a solution of oxidative agent and observed for colour change. The formation of a dark blue color was recorded as GN–NENT (Gram Negative–non enteric).

A bacterial suspension was then made, from which a loopful was mixed into an inoculant solution and then put onto a turbidimeter. The Biolog® turbidimeter percentage of transmission was pre-set to accommodate the GN or GP status. The cultures were next inoculated into GN or GP microplates (based on the status determined from previous tests) at 145 µl per well of the 96 well micro plates. These were then covered and incubated at 28° to 30°C for 24 hours. Finally, the microplates were placed into the automated plate reader installed with the Biolog® identification software, which identifies bacteria up to species level based on percentage similarity. Details of the protocol and procedures for bacterial identification were in accordance with that outlined in the Biolog® identification system.

Antagonistic Studies Against Ganoderma boninense *(FA 5011)*

Strain FA 5011 was previously isolated from an infected oil palm in Banting, Selangor, which was the infecting agent used in the nursery trials of Abdullah *et al.* (2003), Nagappan (2005) and Sundram (2005). Two single bacterial streaks were made on Potato Dextrose Agar (PDA) on the upper and lower part of the plate, after which a 6-mm mycelial plug of FA 5011 cut from a freshly-growing colony and was placed centrally in between the streaks. The cultures were incubated under ambient laboratory conditions for 8 days. The linear growth of the fungus was observed and compared to its growth on the unchallenged control plate. Bacterial colonies which exhibited antagonistic properties towards *G. boninense* were selected for subsequent quantitative assessment by dual culture bioassays.

For the bioassay, a single bacterial streak was made at 5 cm away on one side from the edge of the petri plate, after which one 5 mm-diameter culture plug of *G. boninense* (isolate FA 5011) was centrally placed on the plate. Growth measurements were made of the linear mycelial growth away from the bacteria (R1) and that facing the bacteria (R2). Readings were recorded at 8, 16, 24 and 32 days after incubation (i.e. until static fungal growth was observed).

The percentage inhibition of radial growth (PIRG) of *G. boninense* by the bacteria was determined based on the formula by Skidmore and Dickinson (1976), given as:

$$PIRG = \frac{R1 - R2}{R1} \times 100$$

where R1 = radius of the colony growing away from the bacteria.
R2 = radius of the colony growing towards the bacteria.

At 32 days, a 6 mm-diameter plug of the challenged *G. boninense* on the side facing the bacteria was cut and replated onto a fresh PDA plate. A resultant fungal regrowth would indicate a fungistatic property while a non-growth would indicate a fungicidal property exerted by the bacteria.

Compatibility Tests with T. harzianum *(FA 1132)*

Strain FA 1132 was originally isolated from soils of an oil palm plantation in Gemencheh, Negeri Sembilan. It proved to be a good biocontrol agent based on previous nursery trials and is the fungus being produced on a large scale for field applications (Abdullah *et al.* 2005).

Bacterial isolates that inhibited the mycelial growth of *G. boninense* were selected and subjected to a dual culture bioassay against *T. harzianum*. A single linear streak of each of the selected bacterial strain was made on PDA and a culture disc of *T. harzianum* (FA 1132) was plated centrally on the agar. The cultures were incubated for seven days after which radial extensions were measured and the PIRG calculated. At the same time, the challenged bacterial colony was picked up with a sterile loop, streaked onto NA and observed for subsequent bacterial growth. A normal regrowth would indicate compatibility whereas an abnormal or a non-growth would indicate lysis of the bacterial colony by the *Trichoderma* isolate.

RESULTS

Bacterial Identification

Four species of bacteria were isolated from the oil palm rhizospheres. They were identified as *Corynebacterium urealyticum* with a population density of 1.27 x 10^9 cfu/ml, *Pseudomonas spinosa* (8.7 x 10^8 cfu/ml), *Chromobacterium violaceum* (5.6 x 10^8 cfu/ml) and *Burkholderia cepacia* (1.07 x 10^8 cfu/ml). With the excpection of *C. urealyticum*, all species were Gram Negative (GN).

Three bacterial species were obtained from the *Trichoderma*-infused compost. These were *Corynebacterium urealyticum* with a population density of 5.8 x 10^8 cfu/ml, *Corynebacterium nitrilophilus* (2.1 x 10^8 cfu/ml) and *Actinomyces radingae* (3.2 x 10^8 cfu/ml). All the three bacterial species above were Gram Positive (GP).

Antagonistic Reactions towards G. boninense *(FA 5011)*

Three out of six bacterial isolates were antagonistic towards FA 5011. They were *Corynebacterium urealyticum, Chromobacterium violaceum* and *B. cepacia*. The non-antagonistic bacteria were *Corynebacterium nitrophilus* and *A. radingae* from the *Trichoderma*-infused compost and *P. spinosa* from the oil palm rhizospheres. Dual culture bioassays showed that except for the *P. spinosa* control isolate, the three other species inhibited the growth of FA 5011 by more than 56% on the eighth day (Table 1).

The mycelial growth of FA 5011 on the control plate reached its maximum at eight days after incubation and started to show declined growth thereafter. For the others, the PIRG of FA 5011 peaked and levelled off at day 24 with no further increases by day 32 (*Fig. 1*). The highest and most significant PIRG at day 24 was shown by *Corynebacterium urealyticum* at 88.20%, followed by *Chromobacterium violaceum* (83.45%) and *B. cepacia* (76.92%), each being significantly different from each other. When the challenged *G. boninense* cultures were replated on PDA, two out of the three cultures grew back at a rate similar to the control plate. The exception was *Corynebacterium urealyticum*, which exhibited retarded growth on PDA. Thus, all the bacterial antagonists were deduced to be fungistatic towards *G. boninense*, with *C. urealyticum* showing very strong fungistasis, possibly fungicidal.

TABLE 1
Percentage inhibition of radial growth (PIRG) of *G. boninense* by selected bacterial isolates at 8, 16, 24 and 32 days of bioassay

Bacterial species / Days of Incubation	8	16	24	32
1. *Corynebacterium urealyticum*	68.42^{aC}	87.50^{aB}	88.20^{aA}	88.20^{aA}
2. *Choromobacterium violaceum*	63.33^{bC}	82.50^{bB}	83.45^{bA}	83.45^{bA}
3. *Burkholderia cepacia*	56.52^{cC}	71.79^{cB}	76.92^{cA}	76.92^{cA}
4. *Pseudomonas spinosa*	7.50^{dA}	0.00^{dB}	0.00^{dB}	0.00^{dB}

Mean separation was done using Tukey HSD. Different letters in the lowercase denote a significant difference ($P \leq 0.05$) between isolates while different letters in the uppercase denote a significant difference ($P \leq 0.05$) between days of incubation.

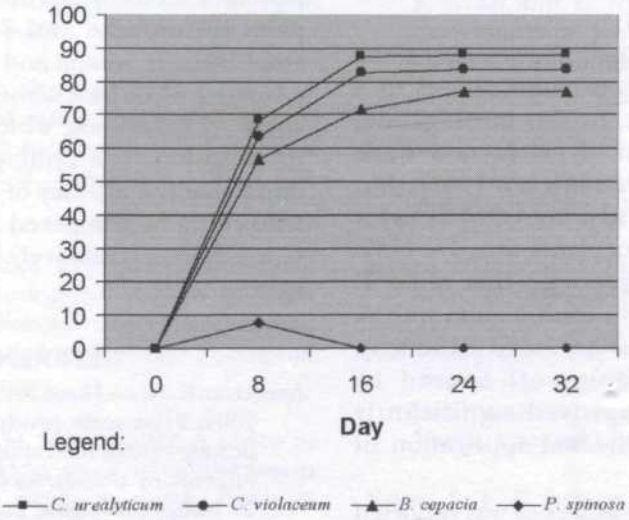

Fig. 1: Percentage inhibition of radial growth (PIRG) of G. boninense *by four bacterial species*

Compatibility with FA 1132

When tested against *T. harzianum*, only *Corynebacterium urealyticum* showed incompatibility towards FA1132. When all of the *Trichoderma*-challenged bacteria were re-isolated and recultured on NA, each of the cultures grew back normally, at a rate similar to the controls.

Of the bacteria isolated from the *Trichoderma*-compost, only *Corynebacterium urealyticum* showed a strongly antagonistic reaction towards FA 5011. *A. radingae* and *Corynebacterium nitrilophilus* both showed good compatibility with FA 1132; however they did not inhibit FA 5011. Thus, these two isolates, together with *Corynebacterium urealyticum* were not good candidates for the selection of potential *Trichoderma* co-inoculants.

DISCUSSION AND CONCLUSION

Bacteria are a good source of microbes for use as biocontrol agents of plant diseases and insect pests. This study successfully isolated and identified a total of seven strains belonging to six genera of bacteria, from two sources closely associated with the oil palm environment. *Pseudomonas spinosa, Actinomyces radingae* and *Corynebacterium nitrilophilus* showed no

antagonistic reaction towards *G. boninense*. The antagonistic isolates were *Corynebacterium urealyticum*, *Chromobacterium violaceum* and *Burkholderia cepacia*; with all three species showing significant differences between each other and a highly significant difference from the control, *P. spinosa*. The PIRG of these three strains increased with time and started to level off at day 24 onwards. However, their antagonistic interactions were fungistatic although *C. urealyticum* showed a reaction that was close to fungicidal. Nevertheless, all the interactions could not match the fungicidal property recorded for *T. harzanium* (strain FA 1132) against *G. boninense* by Abdullah *et al.* (2003) and Ilias (2000).

Besides its individual application, some bacteria are effective as co-inoculants in a microbial consortium. In this investigation, only *B. cepacia* and *C. violaceum* were compatible with *T. harzianum* (FA 1132), thus presenting themselves as prime candidates for use as potential co-inoculants with FA 1132. Roberts *et al.* (2004) reported that when *T. virens* was formulated in combination with *B. cepacia* and *B. ambifaria* as a seed treatment, the control of damping-off caused by *Rhizoctonia solani* improved significantly compared to the individual application of these microbes.

Chromobacterium violaceum was reported to have shown good properties as a biocontrol agent of the soilborne *Fusarium solani* against damping off of aubergine by Park *et al.* (1995). However, this violet-pigmented bacteria was also recorded as a human pathogen (Betts *et al.* 2004; Chattopadhyay *et al.* 2002). *C. violaceum* was reported by Duran and Menck (2002) as an opportunistic pathogen but of a type which can cause extreme virulence to humans and animals when infected. Further studies will need to be conducted to determine its biosafety as a biological control agent. Similarly, *B. cepacia* has also been recorded as a human pathogen but studies by Richardson *et al.* (2002) confirmed that the *B. cepacia* used for the control of plant pathogens was of a different strain to that which caused cystis fibrosis in humans. In another study, Alias and Tan (2005) reported that *B. cepacia* has the ability to biodegrade polyhydroxyalkanoate (PHA).

Bacteria is also known to exert its influence in the rhizosphere competency of other biocontrol agents. Kredics *et al.* (2003) reported that strains of soil bacteria can actually suppress the activity of *Trichoderma* biofungicides in agricultural soils. Thus the success of *Trichoderma* biocontrol agents applied to soil is highly dependent on the resident soil bacteria for rhizosphere competency.

In conclusion, this study was able to isolate a total of seven strains of bacteria from an oil palm rhizosphere and *Trichoderma*-compost combined. *B. cepacia* and *C. violaceum* have a potential as co-inoculants to the biocontrol agent *T. harzanium*, which warrants further investigation. The findings of this study also imply that the efficacy of *T. harzanium* in the field could be hampered if the population of *C. urealyticum* is relatively high in the oil palm rhizospheres.

REFERENCES

ABDULLAH, F., NOOR HAIDA SEBRAN and STEVEN CHONG. 2004. Pilot scale production of *Trichoderma* propagules for field application onto oil palms infected by *Ganoderma boninense*. In Abstracts, *2nd Biology Colloquium. Biology: Unity and Diversity 2004*, organized by the Department of Biology, Universiti Putra Malaysia.

ABDULLAH, F., M. Z. N. A. IZZATI, G. N. M. ILIAS, M. NELSON and U.K. YUSUF. 2003. Disease assessment and efficacy of *Trichoderma* as a biological control agent of basal stem rot of oil palm. *Science Putra Research Bulletin* **11(2):** 31–33.

ALIAS, Z. and I. K. P. TAN. 2005. Isolation of palm oil-utilising, polyhydroxyalkanoate (PHA)-producing bacteria by an enrichment technique. *Bioresource Technology* **96(1):** 1229-1234.

BENNET, A. J., C. LEIFERT and J. M. WHIPPS. 2003. Survival of the biocontrol agents *Coniothyrium minitans* and *Bacillus subtilis* MBI600 introduced into pasteurized, sterilized and

non-sterile soils. *Soil Biology and Biochemistry* **31(3):** 422-427.

BETTS, H. J., R. R. CHAUDRI and M. J. PALLEN. 2004. An analysis of type-III secretion gene clusters in *Chromobacterium violaceum*. *Trends in Microbiology* **12(11):** 476-482.

CHATTOPADHYAY, A., V. KUMAR, N. BHAT and P. L. N. G. RAO. 2002. A rare but frequently fatal disease in *Chromobacterium violaceum* infection. *Journal of Pediatric Surgery* **37(1):** 108-110.

COLLINS, D. P. and B. J. JACOBSEN. 2003. Optimizing a *Bacillus subtilis* isolate for biological control of sugar beet Cercospora leaf spot. *Biological Control* **26(2):** 153-161.

DE JENSEN, C. V., J. A. PERCICH and P. H. GRAHAM. 2002. Integrated management strategies of bean root rot with *Bacillus subtilis* and *Rhizobium* in Minnesota. *Field Crops Research* **74(2):** 107-115.

DRIESCHE, V. R. G. and T. S. J. BELLOWS. 1996. Methods for biological control of plant pathogens. In *Biological Control*. Washington: Chapman & Hall. International Thompson Publishing Company.

DURAN, N. and C. F. M. MENCK. 2002. A review of pharmacological and industrial perspectives in *Chromobacterium violaceum*. *Critical Review in Microbiology* **27(3):** 201-222.

FARIDAH ABDULLAH, NOR HAIDA SEBRAN and MOHD. HEFNI RUSLI. 2005. Production and field application of *Trichoderma harzianum* on oil palms. In *Proceedings of the 27th Symposium of the Malaysian Society for Microbiology: Innovation through Microbes*, pp. 77 – 79. Organised by the Malaysian Society for Microbiology.

FRAVEL, D. R., W. J. JR. CONNICK and J. A. LEWIS. 1998. Formulation of microorganisms. In *Formulation of Microbial Biopesticides*, ed. H.D. Burges, pp. 198-202. Dordrecht: Kluwer Academic Publishers.

ILIAS, G. N. M. 2000. *Trichoderma (Pers. Ex Fr.)* and its efficacy as a biological control agent of basal stem rot of oil palm *(E. guineensis Jacq.)*. Ph.D. Thesis, Universiti Putra Malaysia.

ILIAS, G. N. M. and F. ABDULLAH. 1998. Diversity of *Trichoderma* from its natural habitat and its *in vitro* studies against *G. boninense*. Abstracts of *International Science Conference (ISC 98)*, 7-9 May 1999.

KHETAN, S. K. 2001. *Biofungicides*. New York: Marcel Dekker Inc.

KNOX, O. G. G., K. KILHAM and C. LEIFERT. 2000. Effects of increased nitrate availability on the control of plant pathogenic fungi by the soil bacterium *Bacillus subtilis*. *Journal of Bioscience and Bioengineering* **91(2):** 173-177.

KREDICS, L., Z. ANTAL, L. MANCZINGER, A. SZEKERES, F. KEREI and E. NAGY. 2003. Influence of environmental parameters on *Trichoderma* strains with biocontrol potential. *Food Technology and Biotechnology* **41(1):** 37-42.

MCPARTLAND, J. M., R. C. CLARKE and D. P. WATSON. 2000. *Hemp Diseases and Pests: Management and Biological Control*. UK: CABI Publishing.

MPOB. 2003. *Annual Research Report 2002*. Bangi, Selangor, Malaysia: Malaysian Palm Oil Board.

NAGAPPAN, J. 2005. The efficacy of three species of *Trichoderma* as a biological control agent of basal stem rot on oil palm seedlings. MSc. Thesis, Universiti Putra Malaysia.

NEMEC, S., L. E. DATNOFF and J. STRANDBERG. 1996. Efficacy of biocontrol agents in planting mixes to colonize plant roots and control root diseases of vegetables and citrus. *Crop Protection* **15(8):** 735-742.

PARK, S. K., YEON and K. K. CHUNG. 1995. Antagonistic effect of chitinolytic bacteria on soil borne plant pathogens. *Korean Journal of Plant Pathology* **11(1):** 47-52.

RICHARDSON, J., J. ELPHINSTONE, D. STEAD and R. COUTTS. 2002. Differentiation of *Burkholderia cepacia* isolates by fatty acids profiling and PCR for biocontrol of *Ralstonia solanacearum*. In *Proceedings of the 7th International Congress of Plant Pathology*, Scotland.

ROBERTS, D. P., S. M. LORHKE, S. L. F. MEYER, J. S. BUYER, J. H. BOWERS, C. J. BAKER, J. T. DE SOUZA, J. A. LEWIS and S. CHUNG. 2004. Biocontrol agents applied individually and in

combination for suppression of soilborne diseases of cucumber. *Crop Protection* **24(2):** 141-155.

SAMUELS, G. J. 1996. Centenary review. *Trichoderma*: A review of biology and systematic of the genus. *Mycological Research* **100**: 923-935.

SKIDMORE, A. M. and C. H. DICKINSON. 1976. Colony interaction and hyphal interference between *Septoria nodorum* and phylloplane fungi. *Transactions of The British Mycological Society* **66(1):** 57-64.

SUNDRAM, S. 2005. Performance of *Trichoderma harzianum Rifai* as biological control agent of basal stem rot of oil palm *(Elaeis guineensis Jacq.)* caused by *Ganoderma boninense* on oil palm seedlings. MSc. Thesis, Universiti Putra Malaysia.

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