

**BIOLOGICAL CONTROL OF *SCHIZOPHYLLUM COMMUNE*
FR. THE SEEDBORNE PATHOGEN OF OIL PALM WITH
ANTAGONISTIC BACTERIA**

By

ANTARJO DIKIN

**Thesis Submitted to the School of Graduate Studies,
Universiti Putra Malaysia, in Fulfilment of the Requirement
for the Degree of Master of Science**

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DEDICATION

Thanks to Allah SWT for his blessing to
my family,

To my wife Ir. Zakiah,

To my son Nicky Rahmana Putra and
my daughter Nanda Marizky

for their undying support.

To my parents who always pray for me.

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

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March 2004

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Schizophyllum commune Fr. is one of the most important seedborne pathogens of oil palm. This pathogen can be isolated from fermented fruits, germinated seeds, rotten fruits and infected seeds. The fungus produced papery and leathery compact white mycelia on both sides of the agar plates and often produced basidiocarps (diameter less than 2 cm) with gills on culture plates. The mycelia produced clamp connections with some hyphae having spinulose projections. The width of the hyphae was 2 to 4 μ . Spores produced from basidiocarp were hyaline, cylindrical and single-celled. *S. commune* grew on PDA medium added 50 g/L of NaCl at 30-35°C optimum temperature but the mycelia became dormant at 45°C. The fungus grew on PDA medium at pH 5 to 6 and was resistant to 1% sodium hypochloride.

Inoculation of *S. commune* Fr. on non germinating oil palm seeds by contact was found to cause a significant decrease of seed germination to 64.3%. Mycelia covered the germ pores of seeds and penetrated the germ pore to reach the surface of seed kernel. Inoculation of non germinating seeds produced abnormal seedlings, inhibition of germ tube elongation and brown discoloration of plumule and radicle. Inoculation of germinating seeds resulted in stunted growth of seedlings, decreased root growth and reduction in both fresh and dry weights.

Histological study of the infected seeds indicated that the mycelia penetrated the rotted fruit to reach the testa through germ pores. Mycelium was unable to directly penetrate the endocarp. In advanced seed infection, white mycelia colonized the surface of seed kernel. Mycelia infected the surface of the testa without formation of 'appressorium' to support absorption to the surface of the kernel nor produced 'haustorium' for absorption of nutrient from the host.

Eight out of 40 bacterial isolates from rotten fruits and infected seeds were found to inhibit the radial growth of *S. commune* in the range 42.9–79.8% and spore germination. The 8 antagonistic bacterial isolates were clustered into 5 species by Biolog® Identification System and they were *Bacillus*

thermoglucoasidarius, *Burkholderia cepacia*, *Pseudomonas aeruginosa*, *Serratia marcescens* and *Serratia* sp.

B. cepacia and *Serratia* sp. grew on NA medium containing 4% and 6% salt concentration respectively. Both species were able to grow on Nutrient Agar (NA) medium that were incubated at 40°C. Both species grew on NA medium containing 0.5% and 2% sodium hypochloride respectively. Both *B. cepacia* and *Serratia* sp. grew on NA medium with pH range 4-8.

Dipped vacuum treatment of antagonistic bacteria at 400 mm Hg vac. for 2 minutes significantly reduced the internal infection of the inoculated non germinating oil palm seeds. *B. cepacia* and *Serratia* sp. significantly increased percentage of seed germination. Dipped vacuum treatment was also used to treat inoculated germinating seeds at 150 mm Hg Vac. for 2 minutes. The results showed that *B. cepacia* reduced seedling infection.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia
sebagai memenuhi keperluan untuk ijazah Master Sains

**KAWALAN SECARA BIOLOGI PATOGEN BIJI BENIH KELAPA SAWIT,
SCHIZOPHYLLUM COMMUNE FR. DENGAN BAKTERIA ANTAGONIS**

Oleh

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Schizophyllum commune Fr. adalah sejenis kulat penting pada jangkitan biji benih kelapa sawit. Kulat ini boleh diasingkan dari pada buah yang terfermentasi, biji bercambah, buah reput dan jangkitan biji. Kulat ini menghasilkan miselia berwarna putih seperti kertas, dan miselia padat pada permukaan kedua-dua piring petri, ia juga menghasilkan basidiokarpa (diameter kurang dari 2 cm) dengan insang. Miselia mempunyai klem dan berduri. Lebar hifa 2 hingga 4 μ . Spora dihasilkan daripada basidiokarpa, putih, silinder dan bersel tunggal. *S. commune* boleh hidup pada media PDA yang mengandungi NaCl 50 g/L pada suhu optimum 30-35°C, tetapi miselia menjadi dorman pada suhu 45°C. Kulat boleh tumbuh di atas

media PDA pada pH 5 hingga 6 dan tahan hidup pada media PDA yang mengandung natrium hipoklorida 1%.

Inokulasi *S. commune* Fr. pada biji yang belum bercambah dengan kontak telah menyebabkan berkurangan percambahan sehingga 64.3%. Miselia menutupi liang biji dan menembusi hingga ke permukaan endosperma. Inokulasi biji yang belum bercambah menghasilkan percambahan abnormal, menahan pemanjangan saluran percambahan, perubahan warna perang pada plumula dan radikel. Inokulasi pada biji yang telah bercambah menyebabkan percambahan bantut, pertumbuhan akar yang lambat dan berat basah dan berat kering percambahan berkurangan.

Kajian histologi pada biji yang dijangkiti menunjukkan miselia menembusi buah yang reput hingga ke testa melalui liang percambahan. Miselia gagal menembusi endokarpa biji sawit secara langsung. Jangkitan selanjutnya menunjukkan miselia menutupi permukaan endosperma. Miselia menjangkiti permukaan endosperma tanpa membentuk apresorium untuk menopang pelekatan pada permukaan endosperma dan menghasilkan haustorium untuk menyerap makanan dari perumah.

Daripada 40 pemencilan bakteria, lapan daripadanya diperolehi daripada buah yang busuk dan biji benih yang dijangkiti menahan pertumbuhan

miselia pada 42.9-79.8%, percambahan spora dan menghasilkan bahan antikulat. Lapan pemencilan bakteria antagonis dalam kumpulan 5 jenis menggunakan Sistem Identifikasi Biolog[®] adalah *Bacillus thermoglucosidasius*, *Burkholderia cepacia*, *Pseudomonas aeruginosa*, *Serratia marcescens* dan *Serratia* sp.

B. cepacia dan *Serratia* sp. boleh hidup pada media Agar Nutrien (AN) yang mengandungi garam klorida 4% dan 6 % berturut-turut. Kedua jenis bakteria boleh hidup pada media AN pada 40°C. Kedua jenis boleh hidup pada media AN yang mengandungi natrium hipoklorida 0.5% dan 2% berturut-turut. Kedua jenis boleh hidup pada media AN pada pH 4-8.

Pengawalan rendaman bakteria antagonis tanpa udara pada 400 mm Hg vac. selama 2 minit bermakna mengurangkan jangkitan pada biji benih sawit yang belum bercambah. *B. cepacia* dan *Serratia* sp. bermakna menaikkan peratus percambahan. Rendaman tanpa udara juga boleh mengawal jangkitan biji benih yang telah bercambah pada 150 mm Hg vac. selama 2 minit. Keputusan *B. cepacia* mengurangkan jangkitan pada anak benih.

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I certify that an Examination Committee met on March 30th 2004 to conduct the final examination of Antarjo Dikin on his Master of Science thesis title “Biological Control of Seedborne Pathogen of Oil Palm, *Schizophyllum commune* Fr. with Antagonistic Bacteria” in accordance with Universiti Putra Malaysia (Higher Degree) Act 1980 and Universiti Putra Malaysia (Higher Degree) Regulation 1981. The Committee recommends that the candidate be awarded the relevant degree. Member of the Examination Committee are as follows:

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DECLARATION

I hereby declare that the thesis is based on my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously or currently submitted for any other degree at UPM or other institutions.

ANTARJO DIKIN

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