

Chakrit Bunyoo 2010: Characterization and Monitoring of Endophytic Actinomycetes Isolated from Wattle tree (*Acacia auriculiformis* A. Cunn. ex Benth.) Using Molecular Techniques.

Master of Science (Genetics), Major Field: Genetics, Department of Genetics. Thesis Advisor: Associate Professor Arinthip Thamchaipenet, Ph.D. 90 pages.

Eleven strains of endophytic actinomycetes were isolated from healthy roots of wattle tree (*Acacia auriculiformis* A. Cunn. ex Benth.) collected from Bangkok and Nakhonprathom provinces, Thailand. Analysis of 16S rRNA gene sequences of those strains revealed that they belong to members of the genera *Streptomyces*, *Actinoallomurus*, *Amycolatopsis*, *Kribbella* and *Microbispora*. Five strains showed antimicrobial activities against test microorganisms. Strain GMKU 932 showed activity against *Bacillus cereus* while strains GMKU 937 and GMKU 938 were active against *Aspergillus niger*. Strain GMKU 940 showed activity against *B. cereus*, *Staphylococcus aureus*, *Escherichia coli*, *Fusarium proliferatum*, *F. moniliforme* and *A. niger*. Strain GMKU 944 was active against *B. cereus*, *S. aureus*, *Ralstonia solanacearum* and *A. niger*. The *egfp* gene was introduced into *Streptomyces* sp. GMKU 944 and GMKU 937 by intergeneric conjugation and was successfully transferred and expressed in *Streptomyces* sp. GMKU 944 only. The residing property of both endophytes in the roots of wattle tree was verified by inoculation of *Streptomyces* sp. GMKU 937 or GMKU 944/*egfp* alone, and the mixture with *Rhizobium* sp. to germinated seeds of wattle tree. The results showed that both strains revealed no effect to nodulation. *Streptomyces* sp. GMKU 937 could be re-isolated from the seedlings, but it was unsuccessful with strain GMKU 944/*egfp*. However, *Streptomyces* sp. GMKU 944/*egfp* could be visualized through EGFP expression by confocal laser scanning microscope and found mainly colonizing at phloem and some at xylem parenchyma cells. SEM micrographs indicated mycelia of *Streptomyces* sp. GMKU 944/*egfp* covering root and nodule surfaces. These results suggested that *Streptomyces* sp. GMKU 944/*egfp* was closely associated with wattle tree and is a true endophyte.

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Thesis Advisor's signature