

Patchareewan Maneesakorn 2010: Discovery, Identification and Molecular Phylogeny of Entomopathogenic Nematodes and Their Symbiotic Bacteria. Doctor of Philosophy (Entomology), Major Field: Entomology, Department of Entomology. Thesis Advisor: Professor Angsumarn Chandrapatya, Ph.D. 125 pages.

Characterization and molecular phylogeny analyses of five *Steinernema* entomopathogenic nematodes based on Internal Transcribed Spacer (ITS) sequences and their obligate symbiotic bacteria based on 16S rDNA sequence suggested that four USA nematode strains (D60, D90, D98, and FC48) were closely related to *Steinernema carpocapsae* and the symbiotic bacteria of D60, D90, and D98 were closely related to *Xenorhabdus nematophila* whereas one associated with FC48 was suggested to be a new species of *Xenorhabdus*. The *Steinernema* strain (MP10) from Thailand was described as *Steinernema minuta* sp. nov. and its symbiotic bacteria was closely related to *Xenorhabdus stockiae*.

Phylogenetic relationships of sixty-seven *Heterorhabditis* entomopathogenic nematodes and their symbiotic bacteria were investigated based on ITS sequences and gyrase B gene sequence, respectively. The MP68 was sister to *H. amazonensis* clade, strains MP17 and MP111 were in *H. indica* clade. Other sixty-four strains from USA belonged to clades of four known species; *H. bacteriophora*, *H. georgiana*, *H. indica* and *H. megidis*. The symbiotic bacteria isolated from all nematode strains belonged to clades of either subspecies *Photorhabdus temperata* or *Photorhabdus luminescens*. Two bacteria strains from Thailand MP68 and MP17, were sister to each other. They are paraphyletic to strain MP111 with respect to the monophyletic sister group of *P. luminescens* subsp. *akhurstii* strain D1. Cophylogenetic tests between *Photorhabdus* and *Heterorhabditis* species using ParaFit detected a significant correlation between the nematode and bacteria trees (ParaFitGlobal = 0.001).

The virulence of four Thai entomopathogenic nematode strains was evaluated against the second instar larva of Japanese beetle, *Popillia japonica*, in laboratory conditions. *H. indica* (MP111) was the most virulent with only 136 IJs/larva were required for 50% larval mortality within 5 days whereas 199, 254 and 501 IJs/larva were required for *Heterorhabditis* sp. (MP68), *H. indica* (MP17) and *S. minuta* (MP10), respectively. The LT₅₀ values at 100 IJs/larva revealed that MP111 killed larvae faster (7.4 days) than MP68 (9.4 days) followed by MP17 (10.5 days) and MP10 (15.7 days), respectively. At a concentration of 1,000 IJs/larva the MP111 strain caused the highest larval mortality (84.81%) compared with MP17 (72.15%), MP68 (72.15%), and MP10 (36.71%) at 5 days after treatment.

Student's signature

Thesis Advisor's signature