Doctoral Thesis

Taxonomic and phylogenetic studies on coastal marine and brackish-water calanoid copepods in the tropical Indo-West Pacific, especially in territorial waters of Thailand

(summary)

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December 2015
Chapter 1. Introduction

In Thailand, taxonomic studies of marine calanoid copepods have been carried out by several researchers. These copepods are composed mainly of planktonic taxa, and have so far accommodated over 120 species recorded from Thailand. Their habitats broadly range from freshwaters to deep oceans. Since these copepods have been considered to be main prey components of planktivorous fish, taxonomic studies have been carried out since the 20th century. Recently, morphology- and molecular-based taxonomic and phylogenetic studies on calanoids have been intensively performed to disclose the speciation and evolutionary patterns of these ecologically important crustaceans. These have unveiled the presence of many sibling and cryptic species of calanoids. However molecular studies on copepods have never been done in Thailand. Since each species of congeneric copepods has its own unique behavior, habitat, distribution and ecology, molecular taxonomy is highly recommended to detect their fine speciation. I have studied the molecular taxonomy of three common Indo-West Pacific species of calanoids occurring in Thailand, using two mitochondrial genes (cytochrome oxidase I (COI) and 16S rRNA (16S)): *Acartia* (*Odontacartia*) *pacific* Steuer, 1915, *Tortanus* (*Tortanus*) *forcipatus* Giesbrecht, 1889, and *Labidocera rotunda* Mori, 1929.

Thailand is bordered by the Gulf of Thailand on the West Pacific side and the Andaman Sea on the Indian Ocean side. These water bodies are separate subunits of the Indo-West Pacific, and have been greatly influenced by geological events such as the existence of Sundaland during the Pleistocene. Hence I have given attention to the physical differences in calanoid copepod habitats occurring in Thailand. Genetic variation of the above-mentioned Indo-West Pacific species of calanoids was also examined.

Chapter 2. Taxonomy of coastal and brackish copepods in Thailand: discoveries of undescribed and first recorded species

In my faunal surveys of copepods, I succeeded in collecting samples broadly from East Asia (Japan, Korea, China) through the Philippines and the Gulf of Thailand to the Andaman Sea. During my surveys in Thailand, I found one new and three first-recorded species of calanoids: *Pseudodiaptomus siamensis* Srinui, Nishida & Ohtsuka, 2013; *Acartiella kempi* Sewell, 1914; *A. nicolae* Dussart, 1985; and *Centropages brevifurcus* Shen & Lee, 1963. All genera are major components of coastal and brackish waters of the Indo-West Pacific region. *Acartiella sinensis* Shen & Lee, 1963 was also redescribed from Thailand, because the original description was so poor. These species are carefully described or redescribed herein, following the Huys & Boxshall’s (1991) strict homology.

This study separated these five species of copepods into two distribution patterns, zoogeographically restricted by water temperature and salinity to either the West Pacific Ocean or the Indian Ocean. Four species were restricted to the West Pacific Ocean: *P. siamensis* occurred in brackish waters of temperature of 28.0 °C and salinity ranging from 0.7 to 23.3; *A. sinensis* occurred in waters around 31.3 °C and 5.0; *A. nicolae* was found in temperature and salinity ranges of 28–29.9 °C and 1.0–4.0, respectively; and *C. brevifurcus* occurred with water temperature and salinity of 31°C and 32.0. The species restricted to the Indian Ocean was *A. kempi*, which appeared in water temperature and
salinity ranges of 31.4–32.1 °C and 13.0–22.0, respectively.

I have been revising the taxonomy of *A. (O.) pacifica sensu lato* collected broadly from Asian waters with modern techniques. The population from the Philippines was revealed to be undescribed, and tentatively described as *A. (O.) sp.* herein.

Chapter 3. Molecular analyses of Indo-West Pacific calanoids: three cases

To elucidate the presence or absence of sibling or cryptic species within three species complexes of marine calanoid copepods in the Indo-West Pacific waters, I analyzed nucleotide sequences of mitochondrial genes of *A. (O.) pacifica* from six populations in East and Southeast Asia, *T. (T.) forcipatus* and *L. rotunda* from various locations in Asian waters. For the species *A. (O.) pacifica s.l.*, two genes detected two species groups consisting of brackish-water (China, Gulf of Thailand, Andaman Sea) and coastal water (Japan, Korea, Philippines) groups. The phylogenetic trees can be further separated into four subgroups, (1) Gulf of Thailand and China, (2) Andaman Sea, (3) Philippines, and (4) Japan, Korea, and *A. (O.) pacifica* (DQ071177, DQ071175 in GenBank). A 636 bp COI fragment of 28 individual sequences revealed 23 haplotypes, and a 163 bp 16S region of 29 individual sequences revealed 17 haplotypes. The maximum genetic divergence between these subgroups was found between the Andaman Sea (Indian) and the Philippines (West-Pacific) (divergence values = 19–20 % and 33–34 % for COI and 16S genes, respectively). These four subgroups most likely correspond to separate species, one of which has been evidently revealed to be an undescribed species based on both morphological and molecular data. In addition to *A. (O.) pacifica s.l.*, *T. (T.) forcipatus* and *L. rotunda* showed moderate genetic differences in COI and 16S between the West Pacific and Indian Oceans. In *T. (T.) forcipatus*, genetic differences were 10–11 % and 7–8 %, respectively, while in *L. rotunda* 11–12 % and 3–4 %, respectively. These suggest that the Indian and West-Pacific populations of coastal or brackish calanoid copepods exhibit great to moderate genetic divergence.

Chapter 4. General discussion

This study is the first to use two genes for the taxonomic and phylogenetic analyses of calanoid copepods from coastal and brackish waters in Thailand. I suggest using morphological and molecular data to continue investigating common species of calanoid copepods throughout coastal and brackish habitats of the Gulf of Thailand and the Andaman Sea. The present data on calanoid copepods have clearly indicated the distinct zoogeographical separation between the Indian and West-Pacific Oceans. Hence even a common species that is broadly distributed in the Indo-West Pacific region needs a taxonomic revision with modern techniques. It is likely that it is composed of several sibling/cryptic species. The next step is to clarify what kinds of vicariant events have caused speciation.