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学位論文要旨 Dissertation Abstract

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学位論文題目: Studies on toxic benthic dinoflagellates from Thailand Title of Dissertation タイ産の底生性有毒渦鞭毛藻に関する研究

学位論文要旨: Dissertation Abstract

Benthic dinoflagellates of genera *Ostreopsis*, *Gambierdiscus* and *Coolia* are known as producers of biotoxins associated with human health problems worldwide. To date, none of the reports, however, have undertaken the detailed morphological and molecular studies to identify them from Thailand. Similarly, their toxicities and effects of environmental factors on their growth have not been elucidated. To obtain useful information for evaluating potential problem by benthic dinoflagellates in Thailand, studies on the species composition, distribution and toxicities of these genera from Thailand and on effects of temperature and salinity on growth of *Ostreopsis* and *Gambierdiscus* were conducted.

The cells of Ostreopsis, Gambierdiscus and Coolia were isolated from the Gulf of Thailand and the Andaman Sea and their strains were established. Based on morphological characteristics of the strains, Ostreopsis cf. ovata, Gambierdiscus caribaeus and Coolia malayensis were identified using a light microscope (LM) and a scanning electron microscope (SEM). The cells of O. cf. ovata displayed tear-like cell shape and a similar plate pattern of Po, 3', 7", 5''', 2''' and 1p. The cells of G. caribaeus from Thailand displayed plate formula: Po, 4', 0a, 6", 6c, ?s, 5''', 0p and 2''''. They showed round and anterior-posteriorly compressed cell shape, broad 2'''' plate, rectangular 2' plate, and symmetrical 3" plate. The thecal plate formula of C. malayensis from Thailand was Po, 4', 0a, 6", ?c, ?s, 5''', 0p and 2''''. The cells of C. malayensis showed the straight 4' plate adjoining the 5" plate, and the largest hypothecal 3''' plate.

Sequences of the large subunit rDNA (LSU rDNA D1/D2 and D8/D10) and internal transcribed spacer containing the 5.8S rDNA (ITS-5.8S rDNA) of the three genera from Thailand were determined. Phylogenetic analyses using the sequences resulted in some of the *Ostreopsis* strains being assigned to *O*. cf. *ovata* and *Ostreopsis* sp. 6, and revealed the existence of a novel clade named *Ostreopsis* sp. 7. Within the *O*. cf. *ovata* clade, several strains from Thailand formed a new subclade, the Thailand

subclade, whereas a few strains belonged to the South China Sea subclade. The LSU rDNA D8/D10 trees of *Gambierdiscus* from Thailand supported the results of the morphological identification and confirmed the existence of *G. caribaeus*. The LSU rDNA D1/D2 trees of Thai *Coolia* also confirmed the existence of *C. malayensis* and found *C.* cf. *tropicalis*. These results suggest that *Ostreopsis* sp. 7 is distributed in the Andaman Sea, whereas the two subclades of *O.* cf. *ovata*, *Ostreopsis* sp. 6 and *C. tropicalis* are distributed in the Gulf of Thailand. *Gambierdiscus caribaeus* and *C. malayensis* were found not only in the Gulf of Thailand but also in the Andaman Sea.

Toxicities of *Ostreopsis*, *Gambierdiscus* and *Coolia* from Thailand were evaluated using mouse bioassay. Strains of *Ostreopsis* sp. 6, *Ostreopsis* sp. 7 and *G. caribaeus* tested were highly toxic, while toxicities of the two subclades of *O*. cf. *ovata* and *C. malayensis* strains were below the limit of detection.

To discuss the bloom dynamics of Ostreopsis species/clade/subclades and G. caribaeus in Thailand, we clarified the effects of temperature, salinity and temperature-salinity interaction on their growth. First, we showed that the f/2 and IMK/2 media were suitable media which gave the highest cell yields for Ostreopsis species/clade/subclades and G. caribaeus strains, respectively. The semi-optimal temperature and salinity conditions (\geq 80% of the maximal growth rate) were 22.7–27.4 °C and 28.7–35.0 for O. cf. ovata Thailand subclade, 27.9–30.8 °C and 23.8–30.8 for O. cf. ovata South China Sea subclade, 23.5–26.4 °C and 29.8–36.0 for Ostreopsis sp. 6 and 23.0–27.2 °C, 28.0–36.0 for Ostreopsis sp. 7 and 20.5–30.5 °C and 25.0–37.5 for G. caribaeus, respectively. Considering these results, temperature-salinity interactions may play important roles in bloom dynamics and the distribution of the Ostreopsis species/clade/subclades and G. caribaeus in Thai coastal waters.

In this study, the benthic dinoflagellates O. cf. ovata, Ostreopsis sp. 6, Ostreopsis sp.7, G. caribaeus, C. malayensis and C. cf. tropicalis were identified from Thailand. The results clearly demonstrate that the toxicities and growth characteristics differ among the benthic dinoflagellates. The data obtained in this study may be useful for understanding the risks of problems caused by them in Thailand.