学 位 論 文 の 要 旨

論文題目 **Microalgal communities on mangrove forest sediments of East Java, Indonesia**
(インドネシア東ジャワのマングローブ林底泥に生息する微細藻コミュニティ)

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CHAPTER 1: General introduction

Indonesia is known as an archipelagic country that has been attributed as the third longest coastline globally. Diverse coastal ecosystems of Indonesia are considered to be rich in biodiversity, and one of the vital ecosystems is the mangrove forests, those accounting for 23% (3 million ha) of the worldwide mangrove areas. In this ecosystem, benthic or epiphytic microalgae (herein abbreviated as MPB) have been regarded as the second largest carbon stocks after the litterfall of mangrove trees; however, there has been still scanty knowledge about MPB flora especially in the Indonesian waters. While MPB in mangrove ecosystems were primarily composed of diatoms, other algal taxa occasionally occur in specific area or season. Dinoflagellates blooming were found at mangrove estuary in Thailand during a period of scant occurrence of diatoms in dry season. Actually, during the survey for MPBs in East Java coasts in this study, a harmful raphydophycean alga was found on the mangrove sediments at Probolinggo.

This study was conducted aiming to increase an awareness and to provide new insights on the importance of mangrove ecosystems in Indonesia, at where the highest annual deforestation rate of mangrove forests in the world has been reported. For this general purpose, the investigation on MPB in the sediment of mangroves are of interests because their compositions are expected to be indices of fertility. Also, there has been no research particularly for harmful algal species in the Indonesian mangrove areas.

CHAPTER 2: Cryptic occurrence of *Chattonella marina* var. *marina* in mangrove sediments in Probolinggo, East Java, Indonesia

During the MPB flora investigation on mangrove swamps of Probolinggo, a mud sample unexpectedly included large numbers of a notorious fish killer, Chattonella-like motile cells, and its resting cysts. These motile cells were established as clonal cultures for further identification and physiological tests. The cysts were examined through palynological and molecular biological techniques. Based on light microscopy and ribosomal RNA gene sequences, these cells and

cysts were identified as Chattonella marina var. marina. While the strains were genetically identical to the temperate strains isolated from Japan and China, temperature experiments showed that the Indonesian strains possessed a high maximum quantum yield of photosystem II even after exposure to 34 °C, a temperature at which the Japanese strain could not survive. Salinity experiments showed adaptation of the strains to a salinity of 15. These findings, together with the discovery of populations of cysts in the mangrove sediment, highlight the tough and unique nature of the Indonesian strains, which are likely adapted to wide fluctuations of temperature and salinity in mangrove swamps, and pose a potential risk to fisheries in Indonesia.

CHAPTER 3: Microphytobenthos florae in different mangrove sediments along the East Java coast; Probolinggo, Situbondo, Banyuwangi

It has been reported that MPB composition tends to exhibit unique locality and unique to adjacent environments. If so, those in the Indonesian mangrove forests could be an indicator of the forests' status, those now suffering intensive activities by human. Therefore, for the first time in Indonesia, this study aims to determine the taxonomic composition of MPB in the sediment of mangrove forests those ranging in geographically and environmentally variations in the East Java. The study locations were stretch along the northern coast of East Java, Probolinggo, Situbondo and Banyuwangi, each representing artificially replanted forest, well-conserved forest and adjacent to shrimp ponds, respectively.

A total of 27 species were identified morphologically in the sediment samples. The cell density and biovolume based on the cell length, width and height measured under the microscope revealed the highest biovolume (183,495 x 10^3 ± 66,965 x 10^3 μm^3 cm^{-3}) at Situbondo, followed by the replanted site, Probolinggo (107,092 x 10^3 ± 26,021 x 10^3 μm^3 cm^{-3}) and the nearby shrimp pond site, Banyuwangi (66,962 x 10^3 ± 19,920 x 10^3 μm^3 cm^{-3}), insisting MPB assemblages and their biomass might differ depend on the fertility status of mangrove ecosystems.

CHAPTER 4: Microphytobenthos florae in mangrove sediments of Pang-Pang Bay; a comparison between a conserved mangrove area and human-affected swamps of the bay

Further field survey was conducted in the unique areas that is Pang-Pang Bay, where the human-affected swamps and the well-conserved national forest (Alas Purwo National Park) are coexisting. At this bay, we aimed to clarify MPB communities of both sides and further proof the above hypothesis stating MPB assemblages might differ depend on the fertility status of mangrove ecosystems. Also, in this study, the water columns during high tide were observed to further estimating the primary production rate of the bay that could be driven by suspended MPB. A total of 56 species of MPB were observed in the sediment samples. The highest cell density (448,380 cells cm^{-3}), as well as biovolume (1,666 x 10^6 μm^3

cm^{-3}) was found at St 1 that located in the conserved side at the east. A dendrogram using the cell density data of each station showed an interesting result where two major clusters determined with ca. 48-50% similarities separated the west (human-affected swamps) and east side (well-conserved swamps). During high tide sampling, about half of microalgal community in the water was consisted of MPB species, including *Navicula agnita* (124 x 10^6 μm^3 cm^{-3}) that was also dominant in sediment samples during low tide sampling. An estimation of primary production in Pang-Pang Bay resulting the highest value of 1.28 gC m^{-2} day^{-1} at the offshore waters (St E).

CHAPTER 5: Physiological responses of microphytobenthos towards environmental changes

Benthic communities of intertidal area, including mangrove swamps, usually suffered to harsh environmental drifts, e.g. drastic salinity and temperature changes within a single day, exposure to direct sunlight at low tide, etc. The unique physiological responses of MPB have been reported to enable them to tolerate these harsh environmental conditions. In this study, the physiological characters of the candidate MPB species were tested against some environmental fluctuations; i.e. extremely high temperature, wide fluctuation of salinity and intensive light exposure those are characteristic environments of the sites. *Tryblionella cocconeiformis*, *Navicula gregaria*, *N. agnita* and *Nitzschia sigma* were selected as the candidate Indonesian strains. For the comparison, two planktonic strains were subjected, *Biddulphia* sp. (Myanmar strain) and *Melosira* sp. (Japan strain). Against the rises of temperature, *Fv/Fm* values of *N. sigma* was recognized to be highest even under the highest temperature (36 °C). Among other strains, Indonesian strains was also able to grow in the broader range of salinity (10 to 30 PSU) with *N. sigma* as the most tolerate species with 1.43 ± 0.14 division day^{-1} at the lowest salinity of 10. The photosynthetic parameters, ΦPSII, NPQ, and qP, were measured to investigate the tolerances on repeated alteration between low light and high light. During the exposure of high light, the ability to quench an excess energy as heat (NPQ) was limiting among the Indonesian strains, especially in *T. cocconeiformis* and *Navicula* species. Even without this dissipating function, at least *N. agnita* could utilize high light without photoinhibition, as indicated by higher photochemical quenching (qP) during high light, indicating the species could adapt high light exposure on the sediments.

CHAPTER 6: General discussion

The ultimate goal of this study is to provide new insights on the importance of mangrove ecosystems, particularly in Indonesia, where the wide mangrove forest is existing but at the same time facing the highest annual deforestation rate. For this purpose, investigation of microalgal communities on mangrove forest sediments

was done revealing their abundance and diversity which was indicating the fertility of mangrove forests in East Java, Indonesia. Beside MPB that primarily consisted of diatoms, *Chattonella marina* var. *marina* was also found in this study, wherein when they bloom, it can cause severe economic losses in fisheries production for adjacent waters.

This study reported a linear regression between the chlorophyll *a* of sediment surface and biovolume of the MPB which showed almost all MPB florae consisted of benthic diatom species. Such linear regression was also obtained in Chapter 4 (Fig. 4-13) which further showed the *Fv/Fm* values and the species diversity index were correlated, which implied environments suitable for the diatoms' photosynthetic performance led high MPB diversity and biomass. The physiology of candidate MPB species were evaluated against some environmental parameters, revealing their high abilities in adapting to temperature increase and wide salinity range, and also their unique mechanisms to respond high light exposure. MPB also played an important role in the primary production in the water columns because of their high occurrence among other planktonic species. The occurrence of dense MPB may contribute as an important primary producer for the swamps as well as the adjacent coastal waters. In the future, these secret function of secret garden in the mangrove forest should be clarified to elucidate high biological production of mangrove forest.