Ⅳ．英文 Abstract

Development of Biodiversity Monitoring Methods for Deep-sea Macro-organisms using Environmental DNA Metabarcoding

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The conservation of marine ecosystems is a matter of global concern, and the Convention on Biological Diversity has set a target of designating 10% of coastal and marine areas as marine protected areas (MPAs) by 2020. While the Japanese government has recently designated MPAs in deep offshore waters, there are still many challenges regarding monitoring biodiversity that need to be addressed. The primary objective of this project is to develop a method for detecting deep-sea macro-organisms using environmental DNA (eDNA) metabarcoding, which allows for the simultaneous detection of multiple species from seawater samples. To achieve this objective, we have identified four criteria: 1) uniqueness or rarity, 2) endangered or declining species and/or their habitats, 3) vulnerability, and 4) biological diversity. The project is divided into two subgroups — fish and invertebrates — and both have 1) established experimental methods, 2) collected reference sequences associated with specimens for accurate species identification, and 3) tested the detectability of deep-sea macro-organisms using deep-sea water pumped from coastal areas and seawater taken from MPAs. For fishes, we optimized the MiFish metabarcoding methods and various experimental protocols to detect deep-sea fish. As a result, we detected a deep-sea-endemic teleost fish (Yokozunaiwashi), the largest of its kind, at depths of over 2,000 meters in an MPA. This species had previously been described from Suruga Bay, 400–600 kilometers away from the area, and has been unknown from the other areas. Additionally, we captured the biodiversity patterns and spatio-temporal dynamics of deep-sea fish communities using pumped deep-sea water and actual seawater from MPAs. For invertebrates, we designed new PCR primers for almost all major taxa in various mitochondrial gene regions and optimized experimental protocols accordingly. As a result, we detected the “giant squid,” with an unknown life history, at multiple MPA sites. Also, we captured biodiversity patterns and spatial dynamics of deep-sea invertebrate communities using pumped deep-sea water and actual seawater from MPAs. We conclude that the newly developed eDNA metabarcoding methods for fish and invertebrates provide relevant information that meets the above four criteria through future continuous monitoring. However, collecting more reference sequences for both groups is necessary to improve species identification accuracy. We also argue that biodiversity monitoring using eDNA metabarcoding and conventional methods, such as nettings and visual observations, is desirable to provide complementary biological information on the target organisms. Finally, we have created a manual of experimental protocols to ensure the reproducibility of experiments.