## Development of Biodiversity Monitoring Methods for the Management of Deep-sea Marine Protected Areas

Principal Investigator: Katsunori FUJIKURA

Institution: 2-15 Natsushima-cho, Yokosuka City, Kanagawa, JAPAN

Tel: 046-867-9555 / Fax: 046-867-9525

E-mail: fujikura@jamstec.go.jp

Cooperated by: National Institute for Environmental Studies, Japan, The University of Tokyo, Natural History Museum and Institute, Chiba, Okinawa Churashima Foundation, Kobe University, Kyoto University, Kumamoto University

## [Abstract]

Key Words: Deep-sea ecosystem monitoring, Biodiversity, In-situ Free-Fall Deep-Sea Ecosystem Observatory (Lander), Marine protected areas, Metagenome, Environmental DNA, Image analysis

Based on the Aichi Targets of the Convention on Biological Diversity, marine protected areas were established on the deep-sea floor in Japanese waters in 2020. However, the conditions for marine protected area designation should be inspected for conformity, as deep-sea ecosystems fluctuate due to geological phenomena, deep-sea fishing, and resource exploitation. Opportunities for deep-sea research are limited, as they typically require large oceanographic research vessels, manned submersible research vessels, humanoccupied vehicles, large deep-sea remotely operated vehicles (ROV), or large winches, which are expensive and specialized operators.

Fortunately, molecular biological methods, deep learning, and machine learning have made remarkable progress and make it possible to obtain data on biodiversity, functions, and abundance from seawater, sediments, and video images. Additionally, underwater environmental measurement sensors are also becoming smaller and more precise. Leveraging these advanced technologies, we aim to develop simple and inexpensive methods for deep-sea ecosystem monitoring.

Considering the depths targeted for deep-sea fisheries and resource exploitation, the monitoring methods to be developed for this project can be used at depths of up to 2000 m. Deep-sea marine protected areas are designated based on the distribution and biodiversity of microorganisms and animals, as well as the environmental characteristics of the marine ecosystem. Therefore, we aimed to use this monitoring method to obtain taxonomic data on prokaryotes, meiobenthos, mega invertebrates, and fish, along with information on their abundance, prokaryotic function, and environmental characteristics of the marine ecosystem.

We have developed an instrument called the In-situ Free-Fall Deep-Sea Ecosystem Observatory (Lander), which is deployed from a ship, lands on the deep-sea floor, and surfaces to conduct the observation. The lander is equipped to collect seawater, seawater filtrate, and sediments on the deep-sea floor. It can also acquire data on video, water temperature, salinity, water pressure, turbidity, current direction and velocity, and dissolved oxygen.

Data on prokaryotic biodiversity, abundance, and function were obtained from metagenomic analyses of seawater and sediment samples. We determined the biodiversity and abundance of meiobenthos by metagenomic and image analysis from sediment samples. Data on the biodiversity of fish and mega invertebrates were obtained by environmental DNA analysis using samples of seawater filtrate. In cases where remotely operated vehicles (ROV) acquired images, data on biodiversity and abundance of mega invertebrates were obtained through image analysis. A detailed manual on how to this deep-sea ecosystem monitoring method is available on the web page (see https://www.jamstec.go.jp/bioenv/j/mpa-monitoring-method/pdf/monitoring\_manual.pdf).

The free-fall deep-sea ecosystem observation lander will be smaller, lighter, and less expensive than previous deep-sea survey tools, and will also require no specialized operators. Deep-sea surveys with landers will be less expensive because they can be done with smaller vessels. Such a simple and inexpensive method of monitoring deep-sea ecosystems would increase opportunities for deep-sea research and allow management of deep-sea marine protected areas based on scientific data.

## [References]

- Aoki, K., Fujiwara Y, Tsuchida S (2022) Estimating deep-sea fish population density from the odour extension area: A theoretical basis and comparison with the conventional methods. Frontiers in Marine Science, doi.org/10.3389/fmars.2022.854958 (IF:5.247)
- Fujiwara., Y., Tsuchida S., Kawato M., Masuda K., Sakaguchi S.O., Sado T., Miya M. and Yoshida T. (2022) Detection of the Largest Deep-Sea-Endemic Teleost Fish at Depths of Over 2,000 m Through a Combination of eDNA Metabarcoding and Baited Camera Observations. Frontiers in Marine Science 9:945758. doi: 10.3389/fmars.2022.945758 (IF:5.247)
- Kawato, M., T. Yoshida, M. Miya, S. Tsuchida, Y. Nagano, M. Nomura, A. Yabuki, Y. Fujiwara, K. Fujikura (2021) Optimization of environmental DNA extraction and amplification methods for metabarcoding of deep-sea fish. Method X, https://doi.org/10.1016/j.mex.2021.101238 (IF: 1.837)
- Kobayashi I, Yamamoto M, Fujiwara Y, Tsuchida S, Fujita T (2022) First Record of the Family Myxasteridae (Asteroidea: Velatida) from Western North Pacific with Description of a New Species of Asthenactis. Species Diversity 27: 251-258, doi 10.12782/specdiv.27.251 (IF:0.336)
- 5) Koeda, K., S. Takashima, T. Yamakita, S. Tsuchida, Y. Fujiwara (2021) Deep-sea fish fauna on the seamounts of southern Japan with taxonomic notes on the observed species. Journal of Marine Science and Engineering, 9(11), 1294, https://doi.org/10.3390/jmse9111294 (IF:2.744)
- 6) Komai, T., Tsuchida, S., Fujiwara, Y. (2023) A new deep-sea palaemonid shrimp assigned to *Periclimenes* Costa, 1844 (Decapoda: Caridea) from the West Mariana Ridge, northwestern Pacific. Zootaxa 5231(4): 376-392. (IF: 1.091) テーマ1と共同
- Minamoto, T. (2022) Environmental DNA analysis for macro-organisms: Species distribution and more. DNA Research, 29: dsac018. (IF: 4.477)
- Miya, M. (2022). Environmental DNA metabarcoding: A novel method for biodiversity monitoring of marine fish communities. Annual Review of Marine Science 14: 161–185 (IF: 16.561)
- 9) Oka, S. I., Miya, M., Sado, T. (2022). Gravity filtration of environmental DNA: A simple, fast, and powerfree method. MethodsX, 9, 101838. (IF: 1.837)
- 10) Okanishi, M., Kohtsuka, H., Wu, Q., Shinji, J., Shibata, N., Tamada, T., Nakano, T., Minamoto, T. (2023) Development of two new sets of PCR primers for eDNA metabarcoding of brittle stars (Echinodermata, Ophiuroidea). Metabarcoding & Metagenomics 7: e94298. (h-index: 14)
- 11) Wu, Q., Sakata, M. K., Wu, D., Yamanaka, H., Minamoto, T. (2021) Application of environmental DNA metabarcoding in a lake with extensive algal blooms. Limnology 22: 363-370. (IF: 2.156)
- 12) Wu, Q., & Minamoto, T. (accepted). Improvement of recovery yield of macro-organismal environmental DNA from seawater samples. Analytical Sciences. (IF: 1.967)
- 13) Zhu, T., Sato, T., Sado, T. Miya, M. & Iwasaki, W. (2023). MitoFish, MitoAnnotator, and MiFish pipeline: updates in 10 years. Molecular Biology and Evolution 40(3): msad035. (IF: 8.800)