# Postprint of Ecological Assessment of Oyster Reef Restoration Project in Li’ ershan, Haimen, Jiangsu

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## Abstract

Jiangsu Haimen Lijia Mountain oyster reef is currently the largest existing intertidal living oyster reef in China. However, due to disturbances from human activities and siltation, among other reasons, the area of this natural oyster reef has been continuously decreasing, affecting its ecological function and conservation management. The first-phase restoration project for the Jiangsu Haimen Lijia Mountain oyster reef was implemented during 2013-2014, and the ecological effectiveness of this oyster reef restoration project was evaluated based on ecological monitoring results. The abundance of living oysters on multi-layer reef structures was significantly higher than on single-layer reef structures ($P < 0.05$); with reef development, oyster abundance on restored reefs decreased significantly ($P < 0.05$). Non-parametric Kruskal-Wallis test results showed that oysters exhibited rapid growth during two growth periods (2013-09-2013-11, 2014-03-2014-05) (Q=10.519, Q=6.527, $P < 0.05$), while during the overwintering period (2013-11-2014-03), oyster growth almost ceased (Q=0.35, $P > 0.05$). With reef development, species richness of macrobenthic communities on restored oyster reefs did not increase, but both the average total density and total biomass showed significant increases ($P < 0.05$); one year after reef construction, the average total density and biomass of macrobenthos on restored oyster reefs were similar to those on natural oyster reefs ($P > 0.05$), but significantly higher than those in unrestored areas ($P < 0.05$); restored oyster reefs and natural oyster reefs had significantly different macrobenthic communities (ANOSIM, P=0.001). The research results indicate that this oyster reef restoration project achieved preliminary success.

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## Ecological Assessment of the Oyster Reef Restoration Project at Haimen Liayashan, Jiangsu Province

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### Abstract

Oyster reefs are important coastal habitats that provide essential ecosystem services, including water filtration, habitat provisioning, shoreline stabilization, and nutrient retention. The Liayashan oyster reef, located offshore of Dongzhao Port in Haimen County, Jiangsu Province, represents the largest extant intertidal living oyster reef in China. However, this natural reef has been rapidly degrading due to high sedimentation, habitat loss, and overfishing, with its area reduced by 30% during 2003–2013. To protect this critical biogenic habitat, a restoration project was initiated during 2013–2014. We recycled 100 tons of oyster shells (aged 6 months) from the oyster aquaculture zone in Xiangshan Bay as substrate (cultch) for larval settlement, filling 16,000 nylon bags (diameter: 20 cm, height: 50 cm, mesh: 2 cm) with these shells. In early July 2013, the bagged shells were deployed in the Liayashan waters, creating habitat for young oysters to attach and grow. We constructed 53 small reefs (24 single-layer reefs [SLR] and 29 multi-layer reefs [MLR]) at five restoration sites within the Liayashan area.

Oyster abundance was significantly higher in MLR than in SLR at each of the four sampling periods, but showed a significant decline with reef development. The mean shell height of oysters in the restored reefs significantly increased during two growth periods (from September to November 2013, and March to May 2014) (Kruskal-Wallis test, $H = 6.527$, $10.519$, $P < 0.05$), but oysters stopped growing during the overwintering period (November 2013 to March 2014) ($P > 0.05$). Forty-one species of resident benthic macrofauna were recorded at the restored reefs. Although species richness showed few changes, the mean density and biomass exhibited significant increases with reef development that were similar to the adjacent natural oyster reef, but significantly higher than at the non-restored zone. The mean total density and biomass of benthic communities were 5.1 and 3.1 times higher, respectively, at restored than at non-restored reefs. MDS plots showed that resident benthic communities differed significantly between the restored and adjacent natural reefs (ANOSIM, $r = 0.001$), which was mainly attributed to higher densities of sipunculid worms (*Phascolosoma esculenta*), rock snails (*Thais luteostoma*), polychaete worms (*Perinereis nuntia*), and Asian shore crabs (*Hemigrapsus sinensis*). Oyster reef restoration in Jiangsu Province was considered successful, improving the oyster population, reef area and density, and biomass of resident macrofauna.

**Keywords:** habitat; biogenic reef; ecosystem engineer; macrobenthos; community; *Crassostrea sikamea*

## Introduction

Oyster reefs represent a special marine habitat widely distributed in temperate estuaries and coastal zones, providing important ecological functions such as water purification, habitat provision for fish, shoreline erosion prevention, and carbon sequestration [1–5]. The global distribution area of oyster reefs has declined substantially, altering the structure and function of nearshore ecosystems. Environmental pollution and disease infestation threaten the population maintenance and resource replenishment of important economic fish species in estuaries and coastal areas [6–7]. To revitalize oyster industries and restore the ecological service values of oyster reef habitats, the U.S. federal government and many states initiated a series of oyster reef restoration projects along the Atlantic and Gulf of Mexico coasts beginning in the 1980s [1, 6, 8]. Artificially restored oyster reefs have played important ecological roles in improving water quality, providing fish habitat, and maintaining biodiversity.

The Haimen Liayashan oyster reef in Jiangsu Province is currently the largest intertidal living oyster reef in China. Research results indicate that this reef maintains high biodiversity and provides habitat for numerous aquatic organisms [12–14]. However, due to human disturbance and sediment deposition, the reef area has been continuously shrinking, and individual oysters within the reef have become progressively smaller, affecting the reef's ecological functions and conservation management. In 2013, the Haimen Liayashan National Marine Park Management Office and the East China Sea Fisheries Research Institute of the Chinese Academy of Fishery Sciences formed a professional team to launch the first phase of the Haimen Liayashan oyster reef restoration project, with the goal of expanding the area of living oyster reefs, proliferating oyster populations, increasing fishery resources and economic shellfish production, and providing ecological service value.

Evaluation indicators for oyster reef restoration effects typically include oyster density, individual size, and resident animal communities [5, 9]. Although the United States has conducted numerous oyster reef restoration projects, most have not implemented follow-up monitoring and assessment of restoration outcomes [10–11]. This project represents the first intertidal oyster reef restoration effort in China, and we evaluated its ecological effectiveness based on follow-up monitoring results.

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### Study Site

The Jiangsu Haimen oyster reef (32°08 22.6 –32°09 22.5 N, 121°32 33.2 –121°33 22.8 E) is located in the Xiaomiaohong waters along the southern Yellow Sea coast of Haimen City, Jiangsu Province. The total reef area is approximately 20 $km^2$, with a dense coverage area of about 1.5 $km^2$ [13]. The reef consists of intertidal patches of varying sizes. During spring tides, the reef is exposed for 4–6 hours per tidal cycle; during moderate tides for 2–4 hours; and during neap tides for 1–2 hours. Two species of living oysters are distributed within the reef, with *Crassostrea sikamea* being the primary reef-building species [13].

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### Restoration Engineering

The substrate material for reef construction consisted of *Crassostrea sikamea* shells collected from the oyster aquaculture zone in Xiangshan Bay, Zhejiang Province. The collected shells were rinsed with high-pressure water guns, disinfected by direct sunlight exposure, and then placed in mesh bags (specifications: 25 cm diameter, 50 cm height, 2.5 cm mesh). Following methods described in references [1, 8], we employed two reef types: (1) Single-layer reef (SLR): bags were arranged side-by-side according to water flow direction without gaps; (2) Multi-layer reef (MLR): after constructing the base layer as described above, an additional layer was placed on top to form a multi-layer structure. This restoration project was conducted during July 2013 at five intertidal restoration sites (AOR1–AOR5) within the Haimen Liayashan area, with reef bags deployed during spring tide exposure periods. A total of 23,350 shell bags were constructed.

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### Ecological Monitoring and Assessment

From September 2013 to May 2014, we conducted monthly ecological monitoring of oyster abundance, individual size, and reef benthic animal communities. During each sampling event, we randomly selected three surface shell bags at each restoration site, immediately placed them in mesh bags, and transported them to shore. All materials were poured into stainless steel sieves, rinsed with tap water, and live oysters on each shell were counted and measured for shell height (SH, mm) using vernier calipers (0.5 mm precision). All resident macrobenthic animals in the sieves were simultaneously collected, fixed and preserved in ethanol, and brought back to the laboratory for analysis. Each animal specimen was identified to species level, counted, and weighed (0.5 mm precision). During the final ecological survey in May 2014, we simultaneously sampled macrobenthic animals from natural oyster reefs and non-restored areas. At natural reef sites and non-restored locations, we randomly placed 0.3 m × 0.3 m quadrats (six replicates per habitat type), with specific sampling methods following reference [13].

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## Statistical Analysis

Data were log-transformed and analyzed using Statistic 6.0 software. Two-way ANOVA was used to examine the effects of sampling time and reef type on oyster abundance. Non-parametric Kruskal-Wallis tests were employed to analyze seasonal variation in oyster shell height. One-way ANOVA was used to test seasonal changes in total density and biomass of resident macrobenthos on restored reefs, with multiple comparisons using LSD tests and letter-based marking. Multivariate statistical analysis (PRIMER v6) was used to compare macrobenthic communities between restored and natural oyster reefs. We first calculated ranked similarity matrices between habitat types to generate non-metric multidimensional scaling (MDS) ordinations, and used analysis of similarity (ANOSIM) to test for significant differences in benthic communities between restored and natural reefs. Similarity percentage analysis (SIMPER) was used to examine dissimilarities between restored reefs at different time points and between restored and natural reefs, identifying species that contributed most to these dissimilarities.

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## Results

**Oyster Abundance and Growth** Two-way ANOVA results indicated that both time and reef type significantly affected oyster abundance on restored reefs ($F_{,} = 4.371$, $P = 0.006$; $F_{,} = 21.665$, $P < 0.001$), with no significant interaction between the two factors ($F_{,} = 1.847$, $P = 0.143$). LSD multiple comparisons showed that oyster abundance was significantly higher in MLR than in SLR ($P < 0.05$), but showed no significant temporal variation ($P > 0.05$). However, abundance in May 2014 was significantly lower than in September 2013 ($P < 0.05$).

Kruskal-Wallis test results showed that mean shell height of oysters exhibited significant increases during two growth periods (September–November 2013 and March–May 2014) ($H = 10.519$, $P < 0.05$), increasing by 3.5 mm and 5.1 mm, respectively. During the overwintering period (November 2013–March 2014), oysters showed no significant growth ($H = 0.35$, $P > 0.05$).

A total of 41 macrobenthic species were recorded on the restored reefs across the four sampling periods, including 13 crustacean species, 11 mollusk species, 10 annelid species, 3 echinoderm species, 2 sipunculid species, and 1 each of coelenterate and chordate species. As the reefs developed, species richness of the macrobenthic community did not change significantly from September 2013 to May 2014. However, mean total density and biomass showed rapid increases ($P < 0.05$).

**Benthic Community Development** One-way ANOVA results revealed that after approximately 20 months of reef development (by May 2014), the mean total density and biomass of macrobenthos on restored reefs approached those of natural oyster reefs ($P > 0.05$) but were significantly higher than in non-restored areas ($P < 0.05$). Specifically, the mean total density on restored reefs was 5.1 times that of non-restored areas, and the mean total biomass was 3.1 times higher.

MDS ordination showed that restored and natural oyster reefs supported significantly different resident macrobenthic communities (ANOSIM, $r = 0.001$). The primary species contributing to these community differences were the sipunculid worm *Phascolosoma esculenta*, the rock snail *Thais luteostoma*, the polychaete worm *Perinereis nuntia*, and the Asian shore crab *Hemigrapsus sinensis*. Macroinvertebrate communities on restored reefs also exhibited significant temporal variation during the 8-month study period (ANOSIM, $P < 0.05$), with key discriminating species including the brachyuran crabs *Macroeodeus distinguendus*, *Hemigrapsus penicillatus*, *Metopograpsus quadridentatus*, the polychaete *Diopatra chiliensis*, and the gastropod *Mitrella bella*.

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## Discussion

**Oyster Population and Benthic Community Development** The most important goal of oyster reef restoration is to establish a self-sustaining oyster population that provides ecological services [11]. Oyster population formation and maintenance depend on suitable environmental conditions such as temperature and salinity. Following reef construction in July 2013, oyster densities in multi-layer reefs reached (10,350.9 $\pm$ 1,948.8) individuals per reef bag by September 2013, indicating high larval recruitment during summer. This success is primarily attributed to the high *Crassostrea sikamea* population in the Haimen Liayashan oyster reef. After approximately 20 months of reef development (by May 2014), mean oyster densities in multi-layer reefs were (3,884.0 $\pm$ 558.6) individuals per bag, comparable to adjacent natural oyster reefs [13] but significantly higher than restored reefs in Louisiana (609–3,119 individuals/$m^2$) and the Yangtze River estuary (180–920 individuals/$m^2$) [5]. These results demonstrate that the restoration site environmental conditions are highly suitable for oyster growth, reproduction, and reef development.

Sediment deposition is a critical environmental factor affecting oyster reef restoration success. Many studies have shown that reefs with vertical structure, such as concrete modules, are more conducive to larval settlement, growth, and reef development. Schulte et al. [19] found that high-relief reefs had significantly higher oyster densities than low-relief reefs. In this study, multi-layer reefs supported significantly higher live oyster densities than single-layer reefs, primarily because the low-relief single-layer reefs were easily buried by sediment. Due to their own weight, single-layer reefs settled to only 10–15 cm above the seabed, whereas multi-layer reefs provided 30–40 cm of vertical structure, offering more attachment and growth space for oysters.

Complex three-dimensional habitat structure increases species richness and diversity in marine ecosystems and enhances predator survival rates. Consequently, biogenic reefs typically support high densities and diversities of benthic and nektonic communities [20–22]. This study found that after reef construction, the mean total density and biomass of resident macrobenthos on restored reefs were similar to natural reefs but significantly higher than non-restored areas. The vertical structure of multi-layer reefs increased habitat complexity, providing more refuge and foraging sites for resident animals. However, restored reefs supported macrobenthic communities that were significantly different from natural reefs, consistent with findings from many other studies. Brown et al. [23] compared nekton and benthic macroinvertebrate assemblages among different reef types on the northern Gulf of Mexico coast, finding that restored and natural reefs supported similar animal communities. Dillon et al. [24] evaluated functional equivalence between restored and natural oyster reefs in Mississippi Bay, northern Gulf of Mexico, and found similar animal communities and food web structures. Our study period was only 8 months, indicating the restored reefs are still in rapid successional stages. Typically, 3–5 years are required for restored reefs to develop benthic communities similar to natural reefs.

**Evaluation of Restoration Success** Internationally, evaluation criteria for oyster reef restoration success remain controversial. Commonly used indicators include oyster density, reef height, and resident fauna [1, 16, 25–26]. Powers et al. [25] proposed the following success criteria based on monitoring in North Carolina: >20 cm reef height, >50 oysters/m², >15 cm mean shell height, and larval recruitment within 3 years. Harwell et al. [26] established evaluation standards for 11 restored reefs in Chesapeake Bay. La Peyre et al. [27] defined complete success as the presence of live oysters on the reef and partial success as the presence of hard substrate when evaluating restored reefs on the northern Gulf of Mexico coast. The monitoring results from our restored reefs fully satisfy these criteria, indicating that the oyster reef restoration project has achieved initial success.

The restored reefs support high-density oyster populations and rich resident macrobenthic communities, demonstrating their capacity to simultaneously provide ecological functions such as water purification and habitat provision. However, the resident macrobenthic community composition differs significantly from natural reefs. Whether these differences affect ecological functions requires further investigation. Continuous monitoring and assessment are needed to elucidate the development and successional processes of oyster populations and reef fauna and to evaluate functional equivalence between restored and natural oyster reefs.

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**References**

[1] Coen LD, Luckenbach MW. Developing success criteria and goals for evaluating oyster reef restoration: ecological function or resource exploitation? *Ecological Engineering*, 2000, 15(3): 323–343.

[2] Cressman KA, Posey MH, Mallin MA, Leonard LA, Alphin TD. Effects of oyster reefs on water quality in a tidal creek estuary. *Journal of Shellfish Research*, 2003, 22(3): 753–762.

[3] [Reference text appears incomplete in original]

[4] [Reference text appears incomplete in original]

[5] Quan WM, Zhu JX, Ni Y, Shi LY, Chen YQ. Faunal utilization of constructed intertidal oyster (*Crassostrea rivularis*) reef in the Yangtze River estuary, China. *Ecological Engineering*, 2009, 35(10): 1466–1475.

[6] Beck MW, Brumbaugh RD, Airoldi L, Carranza A, Coen LD, Crawford C, Defeo O, Edgar GJ, Hancock B, Kay MC, Lenihan HS, Luckenbach MW, Toropova CL, Zhang GF, Guo XM. Oyster reefs at risk and recommendations for conservation, restoration, and management. *BioScience*, 2011, 61(2): 107–116.

[7] Jackson JBC, Kirby MX, Berger WH, Bjorndal KA, Botsford LW, Bourque BJ, Bradbury RH, Cooke R, Erlandson J, Estes JA, Hughes TP, Kidwell S, Lange CB, Lenihan HS, Pandolfi JM, Peterson CH, Steneck RS, Tegner MJ, Warner RR. Historical overfishing and the recent collapse of coastal ecosystems. *Science*, 2001, 293(5530): 629–637.

[8] Brumbaugh RD, Coen LD. Contemporary approaches for small-scale oyster reef restoration to address substrate versus recruitment limitation: a review and comments relevant for the Olympia oyster, *Ostrea lurida* (Carpenter, 1864). *Journal of Shellfish Research*, 2009, 28(1): 147–161.

[9] Quan WM, Humphries AT, Shen XQ, Chen YQ. Oyster and associated benthic macrofaunal development on a created intertidal oyster (*Crassostrea ariak-*

*ensis*) reef in the Yangtze River Estuary, China. *Journal of Shellfish Research*, 2012, 31(3): 599–610.

[10] Rodney WS, Paynter KT. Comparisons of macrofaunal assemblages on restored and non-restored oyster reefs in mesohaline regions of Chesapeake Bay in Maryland. *Journal of Experimental Marine Biology and Ecology*, 2006, 335(1): 39–51.

[11] Volety AK, Savarese M, Tolley SG, Arnold WS, Sime P, Goodman P, Chamberlain RH, Doering PH. Eastern oysters (*Crassostrea virginica*) as an indicator for restoration of Everglades ecosystems. *Ecological Indicator*, 2009, 9(6): S120–S136.

[12] [Reference appears to be about geomorphology of Xiaomiaohong oyster reef in Jiangsu - incomplete in original]

[13] [Reference appears to be about macrobenthic diversity and community structure - incomplete in original]

[14] [Reference appears to be about breeding habits of Songjiang perch - incomplete in original]

[15] Hadley NH, Hodges M, Wilber DH, Coen LD. Evaluating intertidal oyster reef development in South Carolina using associated faunal indicators. *Restoration Ecology*, 2010, 18(5): 691–701.

[16] Baggott LP, Powers SP, Brumbaugh R, Coen LD, DeAngelis B, Greene J, Hancock B, Morlock S. *Oyster Habitat Restoration Monitoring and Assessment Handbook*. Arlington, VA, USA: The Nature Conservancy, 2014: 96.

[17] La Peyre MK, Humphries AT, Casas SM, La Peyre JF. Temporal variation in development of ecosystem services from oyster reef restoration. *Ecological Engineering*, 2014, 63(1): 34–44.

[18] Nestlerode JA, Luckenbach MW, O' Beirn FX. Settlement and survival of the oyster *Crassostrea virginica* on created oyster reef habitats in Chesapeake Bay. *Restoration Ecology*, 2007, 15(2): 273–283.

[19] Schulte DM, Burke RP, Lipcius RN. Unprecedented restoration of a native oyster metapopulation. *Science*, 2009, 325(5944): 1124–1128.

[20] Drexler M, Parker ML, Geiger SP, Arnold WS, Hallock P. Biological assessment of eastern oysters (*Crassostrea virginica*) inhabiting reef, mangrove, seawall, and restoration substrates. *Estuaries and Coasts*, 2014, 37(4): 962–972.

[21] Scyphers SB, Powers SP, Heck KL Jr, Byron D. Oyster reefs as natural breakwaters mitigate shoreline loss and facilitate fisheries. *PLoS One*, 2011, 6(8): e22396.

[22] Quan WM, Humphries AT, Shi LY, Chen YQ. Determination of trophic transfer at a created intertidal oyster (*Crassostrea ariakensis*) reef in the Yangtze

River estuary using stable isotope analyses. *Estuaries and Coasts*, 2011, 35(1): 109–120.

[23] Brown LA, Furlong JN, Brown KM, La Peyre MK. Oyster reef restoration in the northern Gulf of Mexico: Effect of artificial substrate and age on nekton and benthic macroinvertebrate assemblage use. *Restoration Ecology*, 2014, 22(2): 214–222.

[24] Dillon KS, Peterson MS, May CA. Functional equivalence of constructed and natural intertidal eastern oyster reef habitats in a northern Gulf of Mexico estuary. *Marine Ecology Progress Series*, 2015, 528: 187–203.

[25] Powers SP, Peterson CH, Grabowski JH, Lenihan HS. Success of constructed oyster reefs in no-harvest sanctuaries: implications for restoration. *Marine Ecology Progress Series*, 2009, 389: 159–170.

[26] Harwell HD, Kingsley-Smith PR, Kellogg ML, Allen SM, Allen SK Jr, Merritt DW, Paynter KT Jr, Luckenbach MW. A comparison of *Crassostrea virginica* and *C. ariakensis* in Chesapeake Bay: does oyster species affect habitat function? *Journal of Shellfish Research*, 2010, 29(2): 253–269.

[27] La Peyre M, Furlong J, Brown LA, Piazza BP, Brown K. Oyster reef restoration in the northern Gulf of Mexico: Extent, methods and outcomes. *Ocean & Coastal Management*, 2014, 89(2): 20–28.

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**Note:** Figure and table references

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## Figures

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Figure 1: Figure 1

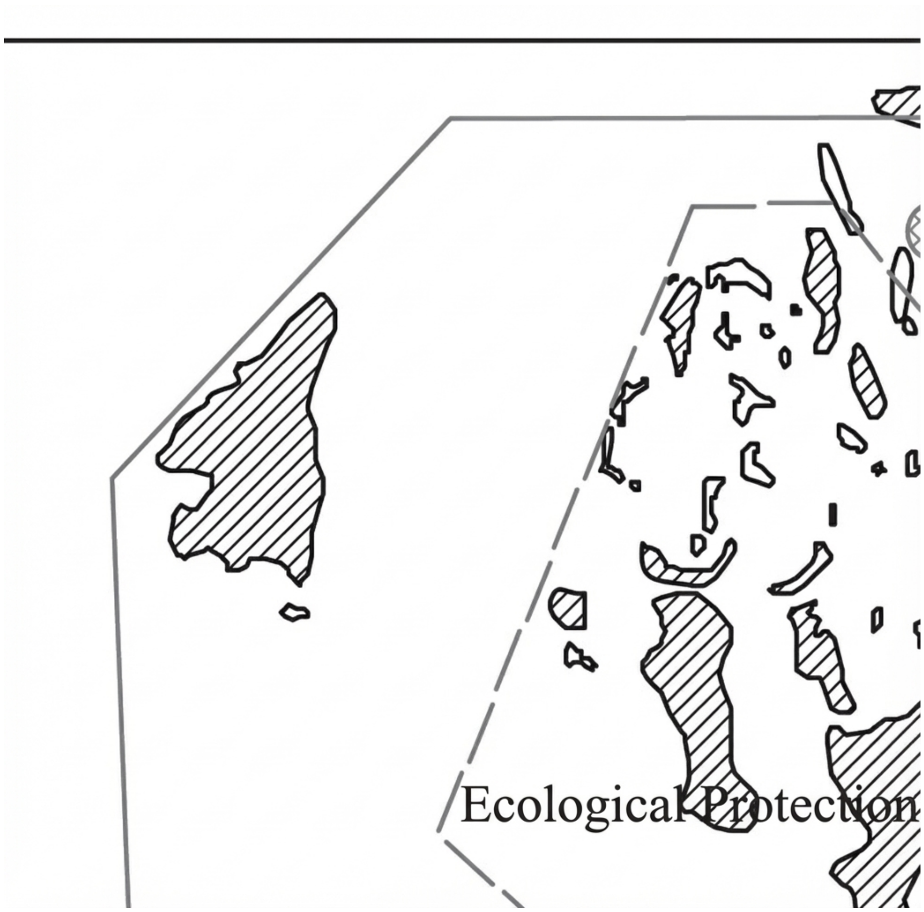
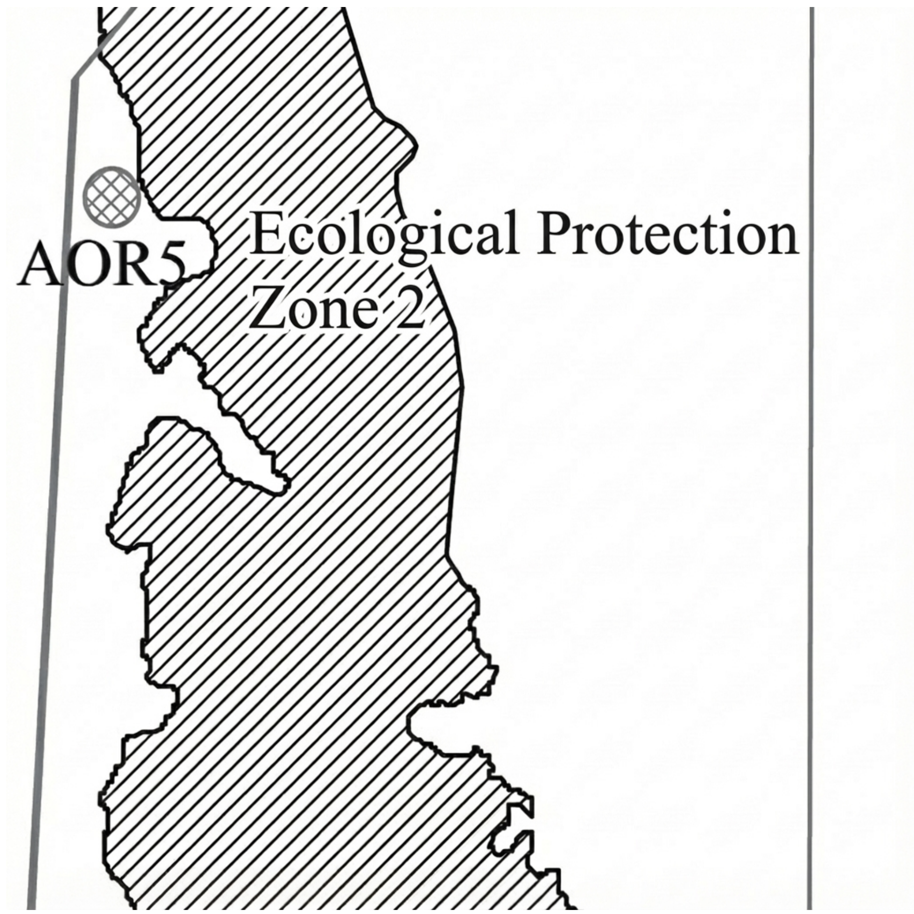



Figure 2: Figure 7

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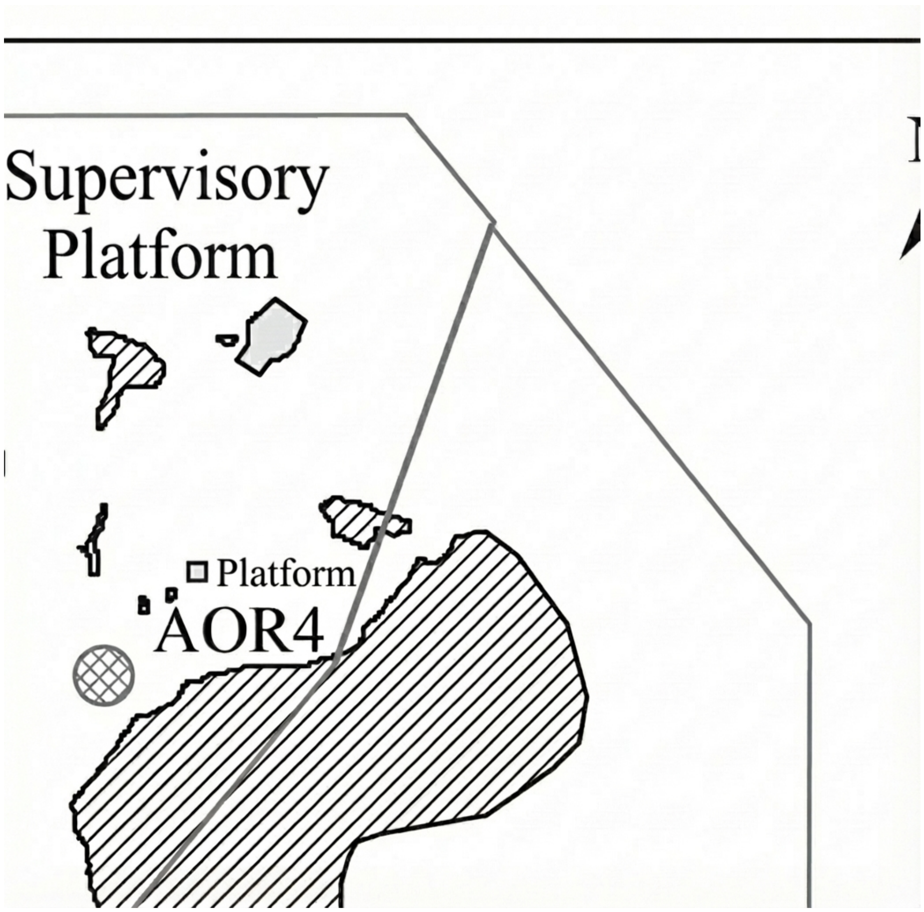


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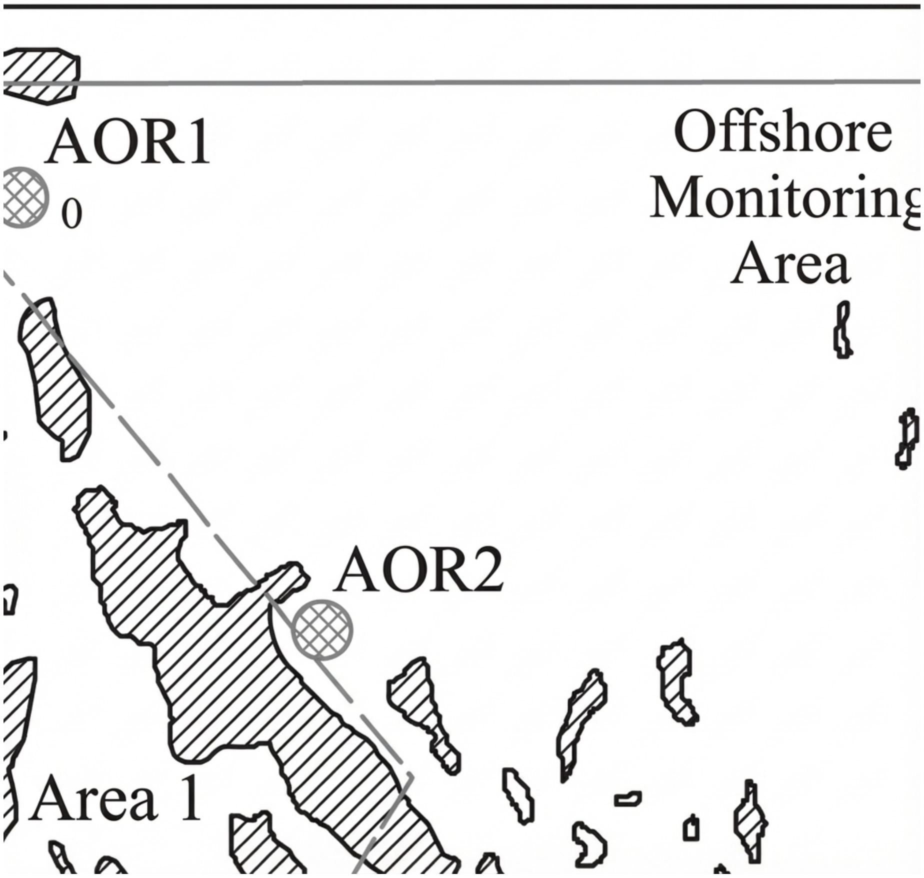


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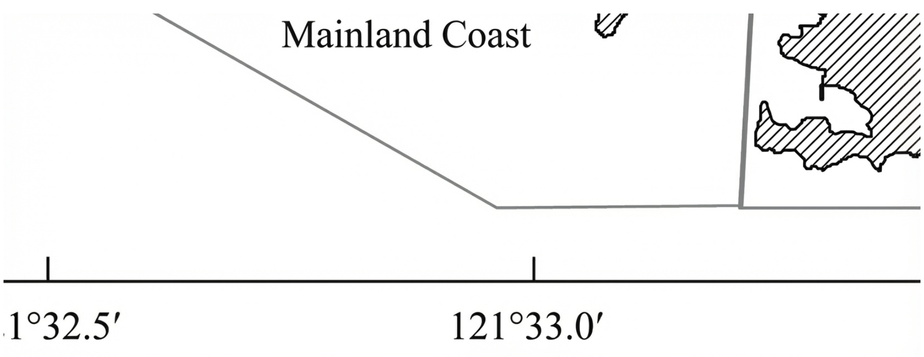


Figure 6: Figure 10