

## Oyster aquaculture using seagrass beds as a climate change countermeasure

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**Abstract:** In the framework of the Sustainable Development Goals (SDGs) led by the United Nations, coastal management methods are required to achieve both sustainable food production and environmental conservation as a climate change countermeasure. Oyster farming is an important food production method now being developed in coastal areas around the world. Recently, climate change has caused several negative effects on oyster aquaculture such as poor spat collection due to oligotrophication, ocean acidification, and poor spat growth and survival due to frequent anoxic events derived from high seawater temperature. The oysters cultivated in many regions of the world are intertidal species inhabiting intertidal zones such as sandy/muddy tidal flats and estuaries, where seagrass beds are often distributed in adjacent lower intertidal and subtidal areas. Seagrass vegetation is one of the most important ecosystems functioning as a countermeasure for global climate change. Not only does it mitigate greenhouse gas emissions by sequestration and storage of blue carbon derived from atmospheric CO<sub>2</sub>, but it also functions as an adaptation measure providing a buffering function against ocean acidification and water quality improvement.

Based on the concept of aquaculture supported by natural ecosystem interactions between oysters and seagrass beds, our project examined whether aquaculture techniques that take into account both mitigation and adaptation to climate change are effective for both sustainable use of coastal areas and environmental conservation. We conducted field experiments in both the French Mediterranean Sea and the Seto Inland Sea of Japan to clarify the effect of eelgrass beds on (1) natural oyster spat collection and (2) growth and survival of oyster spat. The results of our experiments revealed that spat recruitment was significantly higher in areas without eelgrass distribution, while spat growth and survival rate after the settlement were significantly higher in eelgrass beds even when anoxic events occurred in the study areas. Therefore, our results indicate a possibility that seagrass vegetation contributes to sustainability of oyster aquaculture by mitigating environmental degradation during cultivation.

**Keywords:** oyster aquaculture, *Zostera*, *Crassostrea gigas*, blue carbon ecosystem, integrated coastal management

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## Introduction

As the impacts of climate change on human societies and ecosystems intensify, various measures to address climate change are being implemented in various fields across the world. The Paris Agreement adopted in 2015 by the UN Conference of the Parties to the United Nations Framework Convention on Climate Change (UNFCCC) states that efforts will be made to keep global average temperature rise well below 1.5°C compared to before the Industrial Revolution. To achieve the climate target, it is recommended that the balance between greenhouse gas emissions and absorption be reduced to zero. However, the CO<sub>2</sub> emissions that are currently in operation and the CO<sub>2</sub> emissions that are committed by fossil-fuel energy infrastructure were estimated at more than 800 Gt in 2018, which is already far beyond the targets of the Paris Agreement to keep the temperature rise below 1.5°C (Tong *et al.*, 2019). Therefore, in order to meet the Paris Agreement climate goals, it will be necessary to retire these infrastructures as soon as possible, as well as increase CO<sub>2</sub> sequestration sinks.

Moreover, climate change is triggering food crises in various regions of the world, and the measures for sustainable food production are also urgently needed. It is generally recognized that there is a trade-off between current food production systems and climate change mitigation measures (Elmqvist *et al.*, 2013). For example, deforestation in terrestrial ecosystems is accelerating to create pastures and other agricultural fields to increase food production (Schiermeier 2019). Therefore, some countermeasures are needed to harmonize sustainable food production with climate change mitigation (Bommarco *et al.*, 2013).

Shallow coastal ecosystems are now highlighted as one of the measures to mitigate the effects of climate change (Hoegh-Guldberg *et al.*, 2019). In particular, blue carbon ecosystems such as mangroves, salt marshes and seagrass vegetation are important ecosystems currently being considered as a new and effective atmospheric CO<sub>2</sub> sink (Kuwae and Hori, 2018). In addition, the blue carbon ecosystem is known as one of the most productive ecosystems with high biodiversity resulting in higher food

production. Therefore, the blue carbon ecosystem is a typical ecosystem providing co-benefits to both sustainable food production and climate change mitigation.

Oyster farming is an important food production method now being developed in coastal areas around the world. Recently, climate change has caused several negative effects on oyster aquaculture such as poor spat collection due to oligotrophication, ocean acidification, and poor spat growth and survival due to frequent high seawater temperature and anoxic events (Hori *et al.*, 2018; Lagarde *et al.*, 2018, 2020). The oysters are originally intertidal species inhabiting intertidal zones such as sandy/muddy tidal flats and estuaries, where seagrass beds are often distributed in adjacent lower intertidal and subtidal areas. Not only can seagrass vegetation mitigate greenhouse gas emissions through sequestration and storage of blue carbon derived from atmospheric CO<sub>2</sub>, but it also functions as an adaptation measure buffering against ocean acidification and improving water quality (Larkum *et al.*, 2006; Duarte *et al.*, 2013; Groner *et al.*, 2018).

Based on the concept of returning to traditional aquaculture using the natural ecosystem interactions between oysters and seagrass beds (Hori *et al.*, 2018), our project is now demonstrating whether aquaculture techniques that take into account both mitigation and adaptation to climate change are effective for both sustainable use of coastal areas and environmental conservation. In this study, we present the results from field experiments to clarify the effect of eelgrass beds on the sustainability of oyster aquaculture in both the French Mediterranean Sea and the Seto Inland Sea of Japan. The seagrass contribution to oyster production was divided into two processes in relation to oyster life cycle stages: recruitment processes from the larval stage to spat settlement, and post-recruitment processes with spat growth. A series of *in situ* experiments were conducted in Japan and France (1) to demonstrate the larval recruitment variability in shellfish farming areas in the presence or absence of eelgrass beds, and (2) to reveal the effects of eelgrass vegetation on the survival and growth of oyster spats. To our knowledge, there has been no prior case study directly demonstrating the effect of oyster-seagrass

interactions on ecosystem functioning, although there has been some prior modelling research on material cycling in a coastal ecosystem including oyster and seagrass beds (*e.g.* Kishi and Oshima, 2008).

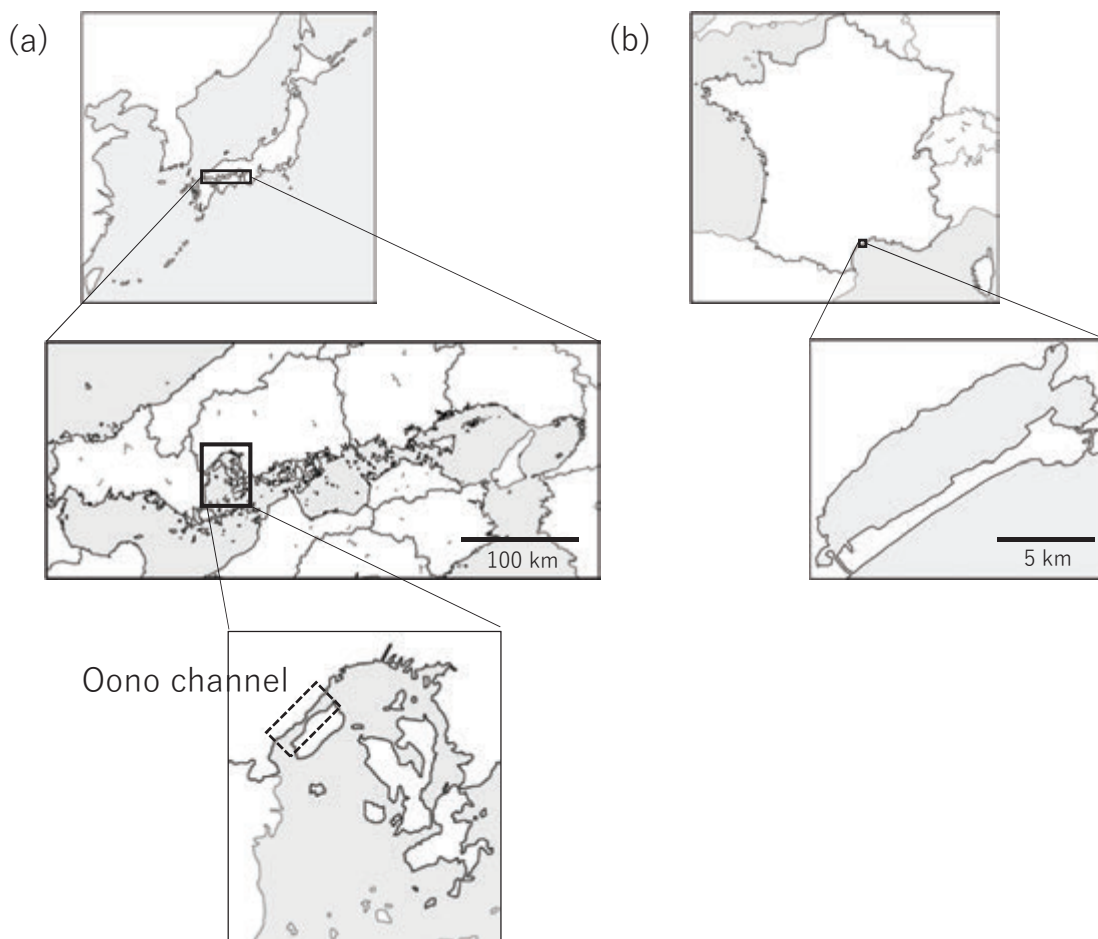
## Methods

### Study sites

The Seto Inland Sea (coordinates at its centre: 34.1667 N, 133.3333 E) in Japan and the Thau lagoon (coordinates at its centre: 43.41 N, 3.6241 E) in France were chosen as study areas for this research (**Fig. 1**). The Seto Inland Sea is located at the southwestern part of the main island of the Japanese archipelago. Rafted aquaculture using natural spats of the native Pacific oyster *Crassostrea gigas* is

flourishing in many areas of the Seto Inland Sea. The production in the Hiroshima Bay and all areas of the Seto Inland Sea accounts for about 60% and 80% of the national production of oysters in Japan, respectively. Recent oligotrophication due to legal restrictions on nutrient input from the watershed has resulted in eelgrass recovery in the Seto Inland Sea over the last decade. It has been estimated that the area of seagrass meadows has increased from 6000 ha to about 10,000 ha in 2011 (Hori and Tarutani, 2015).

The Thau lagoon is the largest lagoon located on the southern French coast in the Mediterranean Sea. The lagoon is famous for oyster farming using non-native Pacific oyster spats attached on ropes containing a specific cement. The ropes with the spats are hung from oyster tables established in



**Fig. 1.** The two comparative study sites, including a) Seto Inland Sea, including the Hiroshima Bay and Oono channel, and b) Thau Lagoon near the Gulf of Lion, Mediterranean Sea. These maps were revised from Hori *et al.* (2018).

the nearshore zone. About 10% of the French national production of oysters is cultivated there, representing the largest oyster farming area in the Mediterranean Sea. It has been suggested that the recovery of eelgrass beds is still proceeding, and that now the area of seagrass distribution extends up from 2 ha to 800 ha (Hori, personal communication with Syndicat mixte du bassin de Thau). The expansion of eelgrass meadows was observed even within oyster farming areas in June 2016.

### 1. Larval recruitment variability in shellfish farming area

In Thau lagoon, settlement and recruitment of oysters were monitored at six contrasting stations in August 2017 from east to west, in the absence or presence of *Zostera* meadow and shellfish farming in the Thau Lagoon (Fig. 1). The oyster collectors were deployed as described by Lagarde *et al.* (2017; 2019) (Fig. 2). The collectors were deployed in the water column (Fig. 3a) inside/outside shellfish farm sites and also hung from the oyster tables above and below the canopy in *Zostera* meadows inside

*Zostera* spp. sites (Fig. 3b) (Lagarde *et al.*, 2020). All collectors were sampled after 2 weeks of immersion to assess pediveligers and postlarvae abundance and after 4 weeks of immersion to assess oyster spat abundance.

In Japan, oyster spat collectors were deployed on the seagrass beds and also hung from the oyster rafts in the Oono channel of the Hiroshima Bay in July 2017 (Fig. 1). Only the collectors in seagrass beds were exposed daily to the atmosphere due to tidal movement. All collectors were sampled after 4 weeks of immersion to assess oyster spat abundance.

### 2. Effect of eelgrass vegetation on oyster survival and growth

Our hypothesis was that seagrass beds could maintain or improve the safety and environmental sanitary conditions for oysters, especially reduction of harmful microbiomes (*e.g.* Lamb *et al.*, 2017; Groner *et al.*, 2018). To clarify the effects of seagrass vegetation on microbiome in both oyster and ambient sea waters, a series of census and experiments were carried out in the Thau Lagoon

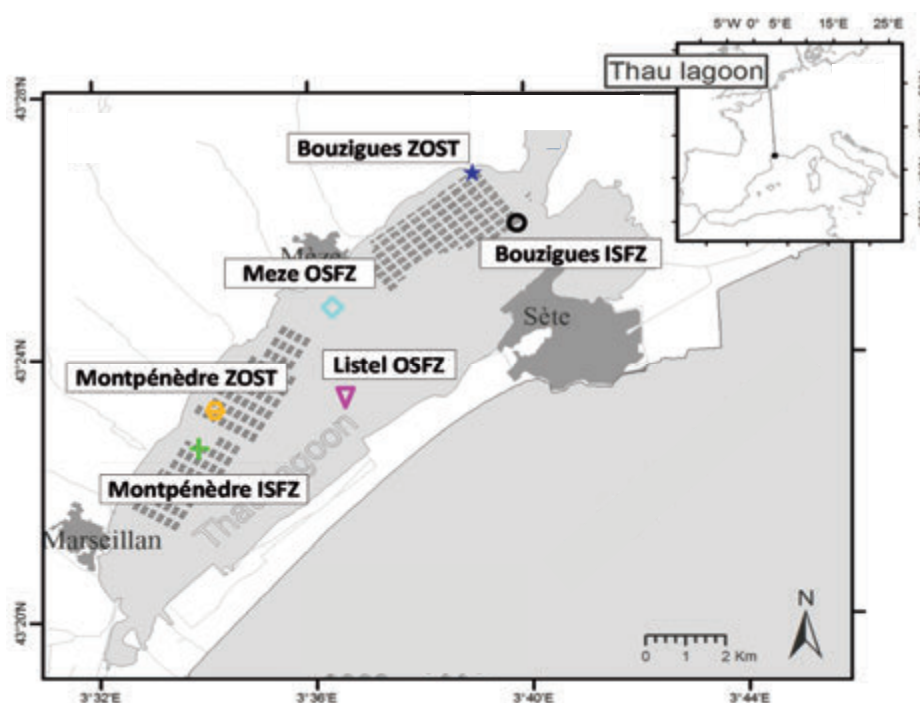


Fig. 2. The six sampling sites scattered from east to west inside Thau Lagoon with three conditions (ISFZ; Inside Shellfish Farming Zone, OSFZ: Outside Shellfish Farming zone, Zost: *Zostera* spp. beds). Shaded squares located at northwest side of the lagoon show oyster tables. This map was revised from Lagarde *et al.* (2020).

in 2017 and 2018 in the presence and absence of *Zostera* meadows (Fig. 2: Bouzigues ISFZ, Bouzigues ZOST, Montpénèdre ZOST, Montpénèdre ISFZ) for analyzing environmental DNA and oyster microbiome. The analyses are still ongoing, therefore the results are not shown in this paper. In parallel, the effect of *Zostera* meadows on growth and survival of juvenile oysters after 3 months of growing (September-December) was tested inside and outside eelgrass beds at Bouzigues and Montpénèdre in the Thau Lagoon in 2018 (Fig. 2).

In Japan, we established a field experiment in Hiroshima Bay to clarify the contribution of eelgrass beds to the survival and growth of oyster spats as a feasibility study of oyster-seagrass interactions (Hori *et al.*, 2018). We established an experimental area (5 m×5 m) in the lower intertidal area on the tidal flat with seagrass vegetation, and set a raft floating on the sea surface 200 m offshore from the tidal flat, hanging a replicate of three cages at a depth of 2 m from the sea surface using vinylon ropes. Thirty spats of each of three native species (*C. gigas*, *C. nippona* and *C. sikamea*), which were hatched from the same lot, were put into the cages on the tidal flat, and the other half of the spats of each species were put into the cages hanging from the raft. The experiment was conducted for two months from November 2016 to January 2017. In addition, we took environmental DNA and microbiome samples from the oysters and ambient waters in both tidal flat with seagrass and the raft to clarify the effects of

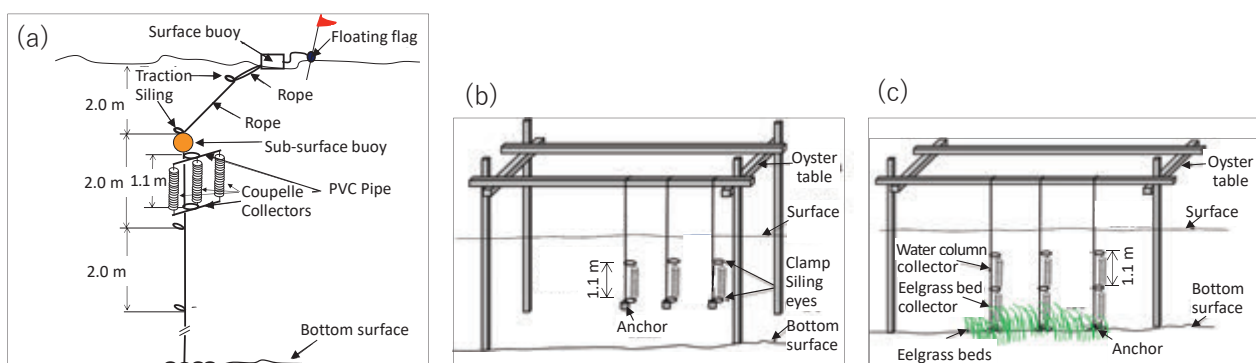
seagrass vegetation on the sanitary condition of the oysters. Analyses of this data and the results from Thau lagoon are ongoing.

## Result and Discussion

### 1. Larval recruitment variability in shellfish farming area

In France, the abundance of young settlers was lower within the canopy of eelgrass beds than above the eelgrass canopy at both *Zostera* sites (Fig. 4). The best sites for settlement were the OSFZ sites (Listel and Meze: outside of the oyster farming and seagrass areas), which confirm those of Lagarde *et al.* (2017; 2018; 2019), where there is a combination of high level hydrodynamic connectivity and favorable trophic supply. These results indicate that eelgrass beds are unlikely to be a preferred site for oyster settlement over other nearby sites in term of abundance. However, between the sites with seagrass vegetation, the abundance of young settlers was much higher in the Bouzigues site where the abundance of eelgrass vegetation was also higher than that in the Montpénèdre site. (Hori, unpublished data). Although eelgrass vegetation does not increase natural spat recruitment, there may be no negative effect on the recruitment.

In Japan, the mean abundance of recruited spats was lower in the eelgrass beds than that hung from the raft without eelgrass vegetation (Fig. 5), although the difference was not statistically



**Fig. 3.** Oyster spats collectors deployed in each site of Thau lagoon. In the site of Outside Shellfish Farming Zone (OSFZ), mooring system (a: left) was used. In the site of Inside Shellfish Farming Zone (ISFZ) without eelgrass beds (b: middle) and the site of inside shellfish farming zone with eelgrass beds (ZOST, c: right), collectors were hung from the oyster table, and deployed above eelgrass canopy (water column) and below the eelgrass canopy. These illustrations were revised from Lagarde *et al.* (2020).

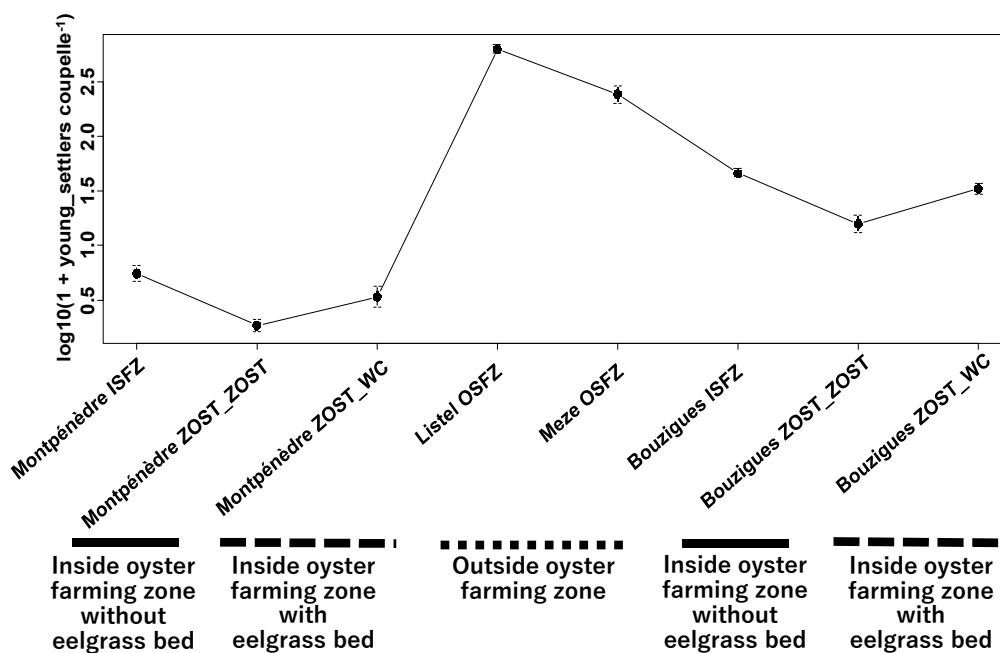


Fig. 4. The abundance of young settlers (pediveligers and postlarvae) on each coupelle collector (mean  $\pm$  SE) in each site. ISFZ: Inside Shellfish Farming Zone, OSFZ: Outside Shellfish Farming Zone, ZOST\_ZOST: collectors inside eelgrass bed, ZOST\_WC: collectors in water column above eelgrass bed.

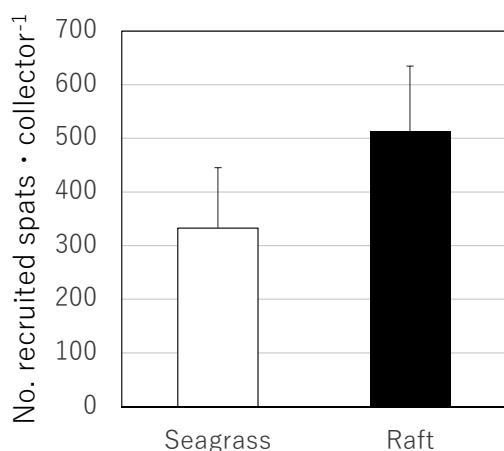


Fig. 5. The abundance of recruited spats on each collector (mean  $\pm$  SD) on both eelgrass beds and oyster raft.

significant (ANOVA:  $F=3.573$ ,  $p>0.05$ ). Natural oyster spats are generally collected in the intertidal zone where native spats of *C. gigas* most frequently settle (FRA, 2016), and young settlers reach the intertidal zone during high tide. The depth of the collectors hung from the raft is similar to the depth of the intertidal zone during high tide.

## 2. Effect of eelgrass vegetation on oyster survival and growth

Although eelgrass vegetation has no significant positive effect on oyster recruitment, the results of spats cultivation experiments exhibited that eelgrass beds seemed to develop a better environment for spat survival and growth in both France and Japan. In France, the survival rate of oyster spats after three months of cultivation was higher in the sites with eelgrass beds (Fig. 6a). Moreover, the survival rate was higher in the Bouzigues site with dense eelgrass beds than the Montpénèdre site with sparse eelgrass beds, suggesting that more abundant eelgrass vegetation could be a better environment for oyster spats. The spats also exhibited better growth in shell length and fresh weight in the site with eelgrass beds than the site without eelgrass beds after three-months cultivation (Fig. 7a).

In the experiment conducted in Hiroshima Bay, the survival rate of oyster spats was higher in the tidal ground with eelgrass beds than that hung from the raft, except for *C. nippona (iwagaki)* spats (Fig. 6b). The tidal flat may not be a good habitat for the *iwagaki* oyster because this species naturally

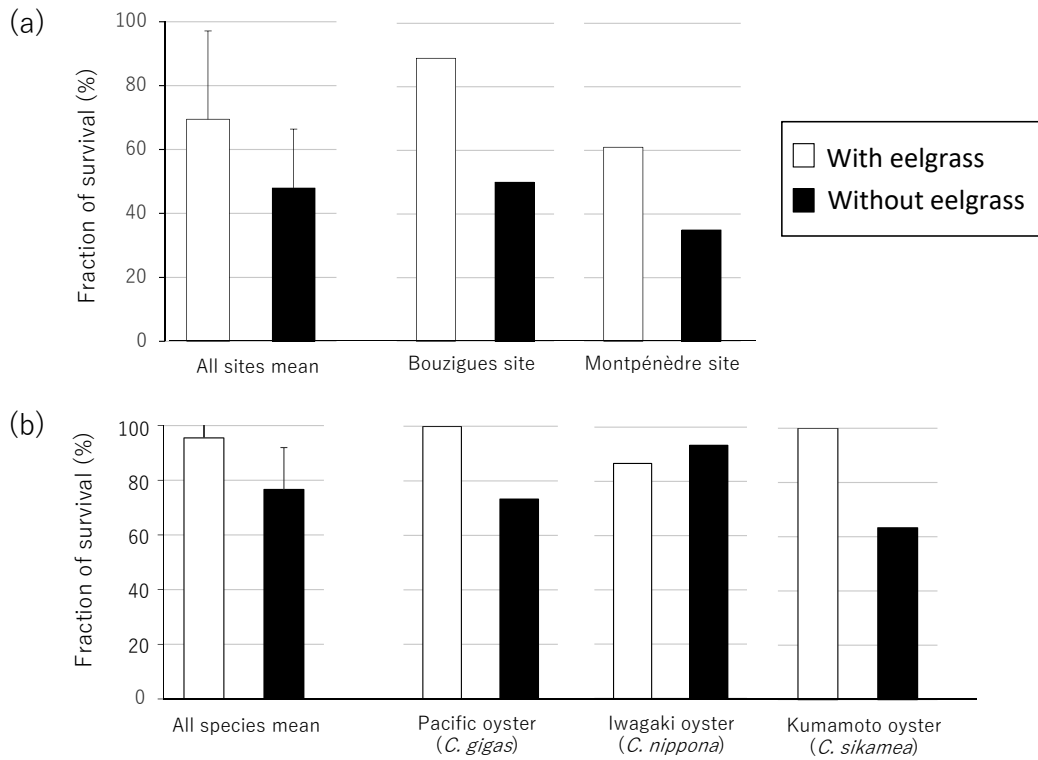


Fig. 6. The survival rate of oyster spats in a) Thau lagoon and b) Hiroshima Bay. In Thau lagoon, the biomass (shoot density x shoot height) of eelgrass in Bouzigues site was much larger than that in Montpénèdre site.

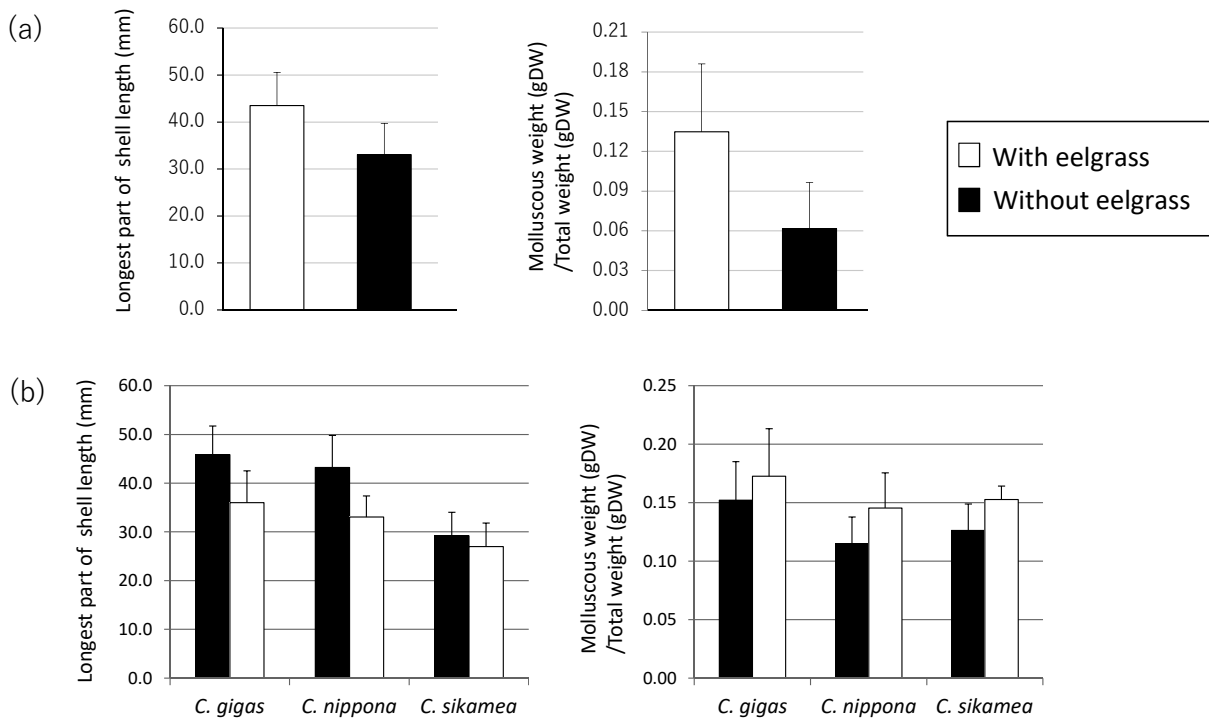


Fig. 7. The difference in shell length and relative meat weight between the site with eelgrass beds and the site without eelgrass beds as an index of growth rate in a) Thau lagoon and b) Hiroshima Bay. The results in Hiroshima Bay was originally referred from Horii *et al.* (2018).

inhabits subtidal rocky shore without exposure to the atmosphere. The growth rate based on relative fresh weight per total weight after two months was also better in the tidal flat with seagrass beds (Fig. 7b), although the shell length of all three species were shorter in the tidal flat with eelgrass. Oyster spats in the cages deployed on the tidal flat were frequently exposed to heavy wave action, such as fishing boat wake during the low tide period, and the tip of the shells were often broken by clashing each other in the cage (Hori, personal observation). This may be a reason that the shell length was shortened in all spats in the tidal flat site.

At least two possible hypotheses can be raised for seagrass vegetation providing better environment to oyster spats: trophic support and the improvement of the sanitary conditions. As a trophic support hypothesis, Hori *et al.* (2018) suggested that oyster spats cultivated in the tidal flat could be used for pelagic production and benthic production in eelgrass beds based on the result of carbon and nitrogen stable isotope analyses. Even *iwagaki* oyster spats with poor survival exhibited better growth in relative fresh weight, thus trophic function of eelgrass beds may have some contribution to the spat growth. These results are also supported in study of Barbier *et al.* (2017) showing that bivalve recruitment abundance is lower in *Zostera marina* beds than others benthic habitat, like subtidal coarse sand or maerl beds (Corallinophycidae, Rhodophyta). However, they observed higher accumulation of fatty acids in the digestive glands of juveniles settled in *Z. marina*, suggesting higher food availability. However, *Crepidula fornicata* banks appear to be the favorable habitat for bivalve recruitment (higher recruit abundance and diversity) with individuals having fatter digestive glands.

The other hypothesis is that seagrass beds could maintain or improve the safety and environmental sanitary conditions for oysters (*e.g.* Lamb *et al.*, 2017; Groner *et al.*, 2018). There are some known negative environmental factors. Some lethal viruses and bacterial pathogens capable of causing mortality of oysters are well known, and anoxia events are a growing threat related to seawater temperature rise during the summer. Lamb *et al.* (2017) reported that the presence of seagrass vegetation reduced

50% of the relative abundance of potential bacterial pathogens capable of causing disease in humans and marine organisms. In our ongoing analyses of the environmental DNA and microbiome from the oysters and ambient waters, some of the results support the previous report by Lamb *et al.* (2017), although more detailed analyses are needed.

Anoxia decreases the growth of oysters even when it is not lethal, because oysters keep their shells closed in an anoxic environment. In fact, serious anoxia occurred around oyster farming areas in Thau lagoon in August 2018 (Author's personal observation). The exceptional climatic conditions in 2018 led to a heat wave and summer anoxia in Thau lagoon, causing massive mortality of oysters and mussels (4000 t, corresponding to 41% of the annual production). Even in this condition, the survival rate of oyster spats in the site with more abundant eelgrass vegetation was approximately 90%, suggesting a possibility that eelgrass vegetation mitigated anoxic environment in the study site. To unravel this mitigative function, more surveys and analyses are needed, such as the relationship between mortality and dissolved oxygen concentration in detail.

In conclusion, seagrass vegetation has potentially positive effects on oysters as reported in Morimoto *et al.* (2017). Our results directly revealed that eelgrass beds can contribute to some processes of oyster aquaculture such as spat cultivation. Therefore, oyster aquaculture using oyster-seagrass interactions could be an effective coastal management option to achieve both sustainable food production and environmental conservation as a climate change countermeasure. For example, oyster-eelgrass interactions may support high water transparency and better sanitary conditions, which are also beneficial for recreational uses. Larger eelgrass beds can absorb more carbon dioxide from the atmosphere and store them as organic carbon (Kuwae and Hori, 2018), which can mitigate ocean acidification and, moreover, offset the carbon emissions from oyster aquaculture and recreational activities. Such local offset systems for carbon emissions can contribute towards solutions for climate change in relation to the framework of the Sustainable Development Goals (SDGs) led by the



United Nations.

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- integrated management for sustainable coastal fisheries under oligotrophication. *Fish. Sci.*, **84**(2), 283–292.
- Harmonizing coastal fisheries with water quality improvement has become an essential factor for the sustainable use of coastal ecosystem services. Here, we present the scope of our study based on an interdisciplinary approach including ecological, socio-economic and socio-psychological actions. We chose to focus on the interaction between oyster aquaculture and seagrass vegetation as a typical ecological action using the coastal ecosystem complex (CEC) concept. Coastal organisms have adapted their traits to the environment over a long period of time, so that restoration of the CEC represents reconstruction of the original process of coastal production. Subtidal seagrass vegetation with intertidal oyster reefs is the original CEC in Japan, which would be expected to enhance coastal production by improving the production efficiency without adding nutrients. A simple field experiment examining carbon and nitrogen contents and stable isotope ratios revealed that oyster spats cultivated on a tidal flat adjacent to seagrass beds had higher nitrogen contents and higher  $\delta^{13}\text{C}$  ratios than spats cultivated in an offshore area using only pelagic production. This result suggests that utilization of the CEC, which enables oysters to use both pelagic and benthic production, has potential to sustain a food provisioning service for humans, even in oligotrophic conditions.
- (2) Lagarde F., Richard M., Bec B., Roques C., Mortreux S., Bernard I., Chiantell C., Messiaen G., Nadalini J., Hori M., Hamaguchi M., Pouvreau S., d’Orbcastel E. R., and Tremblay R., 2018: Trophic environments influence size at metamorphosis and recruitment performance of Pacific oysters. *Mar. Ecol. Prog. Ser.*, **602**, 135–153.
- Reproduction and recruitment of benthic invertebrates are influenced by climate and by the ecological structure of marine ecosystems, along with local anthropogenic pressures such as eutrophication or oligotrophication. Using the Pacific oyster, *Crassostrea gigas*, as a biological model, we tested the hypothesis that the variability in prodissoconch II (PII) size (*i.e.* size at metamorphosis) depends on

#### Annotated Bibliography of Key Works

- (1) Hori M., Hamaoka H., Hirota M., Lagarde F., Vaz S., Hamaguchi M., Hori J., Makino M., 2018: Application of the coastal ecosystem complex concept toward

ecological functioning. Settlement and recruitment were assessed at 5 sampling sites in the French Mediterranean shellfish farmed Thau lagoon during the main summer recruitment events in 3 consecutive years (2012-2014). Hydrobiological and planktonic analyses were conducted at 3 sampling sites. Our results showed that recruitment was extremely heterogeneous, ranging from 0 to  $260 \pm 27$  SE ind.  $\text{dm}^{-2}$  throughout the ecosystem and was linked with variability in PII size, which ranged from 180 to 296  $\mu\text{m}$ . The annual temporal pattern of PII sizes appeared to be controlled by temperature during the settlement period, whereas the spatial pattern depended on phytoplankton biomass and on

the trophic functioning of the ecosystem. Smaller PII sizes were significantly correlated with the highest phytoplankton biomass, while larger PII sizes were positively correlated with mixotrophic cryptophyte abundance. We found an inverse relationship between PII size and survival after metamorphosis, showing that recruitment success was associated with smaller PII sizes. Regional climate conditions and local trophic functioning appear to be key factors in metamorphosis and consequently contribute to recruitment heterogeneity. Further studies should be performed in other ecosystems following an oligotrophication trajectory to generalize this result.