

Horizontal distribution of concentration and composition of microplastics in sediments of Otsuchi Bay, Japan

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INTRODUCTION

The amount of waste plastic discharged into the ocean is estimated between 4.8 and 12.7 million tons per year (Jambeck et al., 2015)¹. It was considered about 47.4% of these plastics were degraded into fine pieces (microplastics, hereinafter referred to as MPs) less than 5 mm in diameter (Lebreton et al., 2017)².

In particular, the number of MPs drifting in the Japanese coastal waters was estimated 1720000 pcs km⁻², which was 27 times higher than the global average (Isobe et al., 2015)³. Previous studies have indicated that 73% of missing MPs accumulated on the seabed as sediments (Matsuguma et al., 2017)⁴. However, our knowledge about MPs contamination in sediments is still insufficient in coastal areas of Japan. The influence of river water on the behavior of MPs is also unclear.

In order to solve the above problems, we observe the horizontal distribution of MPs concentrations and composition in sediments on the sea floor in Otsuchi Bay, one of small ria bays in Sanriku on the Pacific coast of Tohoku, Japan.

METHODS

The survey was conducted in March 2021 by a research vessel of boat of the International Coastal Research Center, AORI, Univ. Tokyo at the three stations in Otsuchi Bay. The seabed sediments were sampled by a G.S. type core sampler (ASYURA) at each station. The sediment cores were sliced onsite into 5-cm intervals and stored in a dark, cold chamber.

The MPs concentration was measured with reference to Masura et al. (2015)⁵. Approximately 10 g (wet weight) of the sample from 0-5 cm layer was separated into a beaker. Next, 300 mL of NaI solution adjusted to a density of 1.6 g cm⁻³ was poured into the beaker and stirred. The sample was left to stand for 24h and the supernatant was then collected. This density-separation operation was repeated three times. After that, the collected supernatant was passed through a sieve with mesh size of 10 µm, and the particles in the supernatant were collected. The particles remaining on the sieve were washed with distilled water. Next, 20 mL of 30% hydrogen peroxide was added to the sample, and the mixture was heated at 60 °C for 3 days to remove impurities. Finally, pure MPs were collected on a PTFE filter (25 mm diameter, 0.45 µm pore diameter) by vacuum filtration.

The sample was analyzed by FTIR-microscope (Thermo Fisher Scientific Nicolet iN10). The measured spectrum was compared with the spectrum of a standard plastic registered in advance (polyethylene, polypropylene, polystyrene, polyamide and polyvinyl chloride) to determine the composition of the particles in the sample (Matsuguma et al., 2017)⁴. The spectrum and size of the MPs particles was measured by an analytical software, OMNIC.

RESULTS AND DISCUSSION

The concentrations of seabed sediments at Sta.1, 2, and 3 in March 2021 were 13.6±9.8, 1.7±0.6, 2.6±0.3 pcs g⁻¹ DW, respectively. The MPs concentration at Sta.1 was significantly higher than that of Sta.2 (Kruskal-Wallis test, $p < 0.05$). The size ranges (average particles size ± standard deviation) of MPs were 353.1±161.5, 269.5±95.6, and 163.7±31.1 µm, respectively. There were no significant differences among stations in terms of the average MPs particle size (Kruskal-Wallis test, $p > 0.05$).

The MPs concentration in sediment in Otsuchi Bay indicated to be higher toward the inner part of the bay which is located on the estuary of the Otsuchi River. This result is consistent with Wang et al., (2021)⁶, who found that MPs concentration of 145 pcs g⁻¹DW on the estuary part of Tokyo Bay was higher than 53.3 pcs g⁻¹DW on the mouth side of the bay. The results might be explained by that the deposition of particles containing MPs is likely strongly affected by the enhanced outflow of the Otsuchi River in estuary, especially during the period of heavy rains.

Another possible explanation is influences of inflow of the offshore water into the bay. The bottom water originated from The Tsugaru Warm Current or the Oyashio Current flows counterclockwise along the coast of the bay during autumn to spring in Otsuchi Bay (Ishizu et al., 2016)⁷. It is necessary to study the seasonal change of MPs distribution in order to clarify the influence of the circulation in Otsuchi Bay.

CONCLUSION

We clarified the horizontal distribution of MPs concentration and composition in seafloor sediments in Otsuchi Bay. The MPs concentration of 13.6±9.8 g⁻¹ DW on estuary side was higher than the 1.7±0.6 g⁻¹ DW on coastal side. There was no difference in the average particle size of MPs at observation points. These results suggest that the settlement of MPs may affected by the river discharge or inflow of the offshore waters into Otsuchi Bay.

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Interdiurnal variation of chlorophyll-a and transition of dominant phytoplankton observed at the Yodo River estuary, JAPAN

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Key words; interdiurnal variation, diatom, dinoflagellate, mole ratio, Yodo River

INTRODUCTION

Osaka Bay located in West Japan is the semi-enclosed bay. Large amounts of nutrients are loaded from the Yodo River and are released from the bottom. Diatom dominates usually in the Yodo River estuary, but other various phytoplankton species also appear. The year-to-year variation of dominant phytoplankton species in 1990s was analysed¹. On the other hand, since the minimum interval of the field observations of red tide is one week, the transition of species at the shorter time and its reasons are indistinct. The field observation was carried out at the Yodo River estuary to clarify the interdiurnal variation of dominant phytoplankton species and the marine environment. And the reason of the transition was discussed.

OBSERVATION

The field observation was carried out from June 29 to July 14, 2004 for 16 days². CTD was installed at the Yodo River estuary. Water depth of the site is about 12 m, and the depth of CTD around 1.5 m. The time series of water temperature, salinity, chlorophyll-a (chl.a) concentration, dissolved oxygen saturation, and light intensity were obtained every 10 minutes. The vertical profile of the same parameter at the site were obtained every 0.1m by another CTD once a day at almost every day. Seawater was also sampled at the three depths, the sea surface, 7 m and 1m above the bottom, and dissolved inorganic nitrogen (DIN), phosphorous (DIP) and silicate (DSi) concentrations, chl.a concentration and cell density of phytoplankton were analysed. General meteorology parameters, air temperature, solar radiation and so on, and tidal height were recorded every minute at Fukae campus of Kobe University.

RESULTS AND DISCUSSION

Figure 1 shows the transition of the cell density of each species and total in the sea surface of the site. Variation of the total cell density agreed with chl.a concentration. *Thalassiosira spp.* dominated throughout the period. On the other hand, other diatom species decreased, and dinoflagellate and other species increased in later period. Figure 2 shows the temporal variation of nutrient concentrations and the ratios. All nutrient decreased toward July 5, but DIN and DIP concentrations recovered. DSi concentration continued the low condition after July 7. From the results based on nutrient concentrations and those mole ratio, the growth of all species was limited by DIP before 2 July. Diatom has advantage in this case because it can grow under the lower DIP concentration. After that, nutrient which limits the growth of diatom changed to DSi due to the decrease of DSi concentration. Then DIP and DIN

concentration increased in favour of dinoflagellate. This is the reason of increase of dinoflagellate in the later period.

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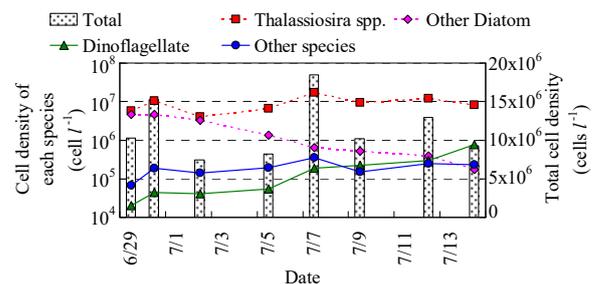


Fig.1 The transition of the cell density in the sea surface of the site.

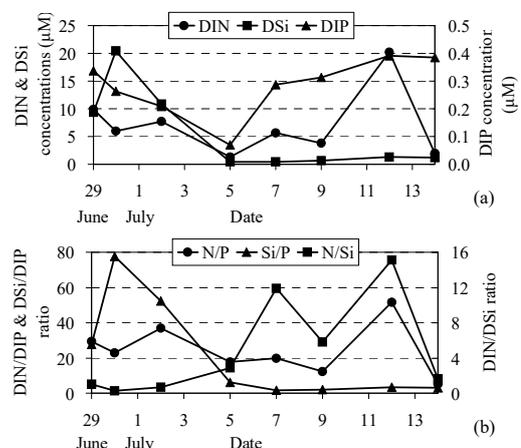


Fig.2 The temporal variation of nutrient concentrations(a) and the ratios(b).

Impact of basin soil on river water quality in salmon breeding rivers in the Nemuro region of Hokkaido

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INTRODUCTION

Salmon (*Oncorhynchus keta*) that live in the Hokkaido-Nemuro region spawn in the upper reaches of rivers. When the fry hatch, they descend to the Bering Sea and grow. After growing, it has the property of returning to the upper reaches of the river.

Salmon fishing has been practiced in the Nemuro region since the 1700s. Even today, salmon fishing is a major industry in the Nemuro region.

The Nemuro region of Hokkaido was a deciduous broad-leaved forest zone until the 1800s. After World War II, the region was developed as a grassland dairy area. As a result, most of the river basin became grassland. On the other hand, deciduous broadleaf forest remains around the river.

It has been pointed out that such changes in the river environment have reduced the forest rate in the river basin and increased the artificial nitrogen input. It has been pointed out that the river water quality is changing due to such changes in the river basin environment.

It has been pointed out that the concentration of nitrate nitrogen, potassium, calcium, and aluminum in river water increased due to the fact that most of the river basin became grassland. It has been pointed out that aluminum is toxic to juvenile salmon.

Therefore, the purpose of this study was to investigate the factors that affect the aluminum concentration in river water.

METHODS

The site where the test was conducted is near the Toyama Bridge on the Jinhuuren River (latitude 43.3 ° north, longitude 145.2 ° east). The basin area is 1543.1 ha. Every year, the Betsukai Fisheries Cooperative releases juvenile salmon at the Toyama Bridge. In May and August 2019, water was sampled at two locations on the Toyama Bridge and the main stream of the Jinhuuren River, and two tributaries, and water quality was analyzed. In October 2019, soil was collected at 35 locations in the grassland upstream of the Toyama Bridge, and soil chemistry was analyzed.

RESULTS AND DISCUSSION

As the exchangeable calcium content of basin grassland soil increased, the aluminum concentration in river water tended to decrease (Table 2). In addition, a negative correlation was found between soil exchangeable calcium and the aluminum concentration in river water. These things have the same results as previously reported. This suggests that the soil exchangeable calcium concentration may be a factor influencing the aluminum concentration in river water at the site where the test was conducted.

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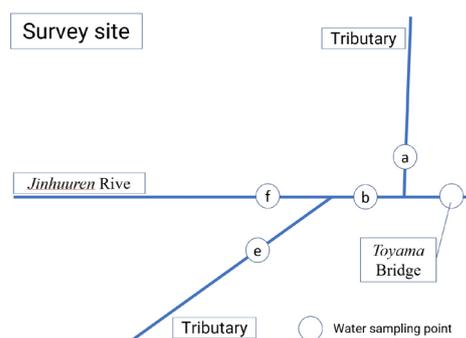


Table 1 Land use of survey site

point	Basin area	Basin grassland ratio
	ha	%
Toyama Bridge	1543.1	60.0
a	226.7	68.1
b	1534.2	60.2
f	-	-
e	35.7	60.5

Table 2 Results of river water quality and soil chemistry

point	river	soil	
	T-Al mg/l	ex-Ca mg/100gsoil	T-C %
Toyama Bridge	71.0	-	-
a	22.5	204.8	10.0
b	123.4	-	-
f	69.2	198.7	9.4
e	214.1	150.0	10.0

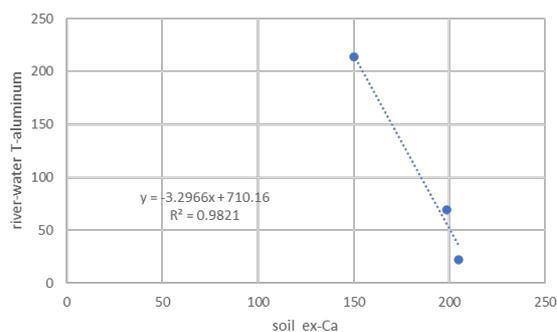


Fig 1 Relationship between soil exchangeable calcium and total aluminum in river water



Contamination of marine Echinoderms by radiocesium released during the Fukushima Daiichi Nuclear Power Plant accident.

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INTRODUCTION

The Great East Japan Earthquake and tsunami that struck on 11 March 2011 caused a severe accident at Fukushima Dai-ichi Nuclear Power Plant (FDNPP), owned by the Tokyo Electric Power Company (TEPCO)¹. The total amount of ¹³⁷Cs directly discharged into the sea was estimated to be approximately 3.5 PBq². Radioactive cesium was detected from marine biota collected in the waters off Fukushima Prefecture and its vicinity immediately after the FDNPP accident³. Our study reports on regional dispersion trends of radiocesium in marine echinoderms caught from coasts of Fukushima Prefecture and ecological half-lives (T_{eco}) in selected species. The purpose of this study was to determine how the radiocesium contamination of echinoderms from the FDNPP accident changed over time. To achieve this, T_{eco} of radioactive cesium in echinoderms were investigated over the period from 426–2726 days after the accident.

METHODS

Six species (*Mesocentrotus nudus*, *Echinocardium cordatum*, *Luidia quinarian*, *Distolasterias nipon*, *Astrocladus coniferus* and *Asterias amurensis*) of Echinoderm samples were collected from three locations (Yotsukura, Ena and Hirono Thermal Power Plant) in Fukushima near FDNPP. After collecting samples, brought to the laboratory and processed. Then Germanium Semiconductor spectrometer (GMX Series Coaxial HP Ge Detector) was used to measure the radiocesium in echinoderms. The measurement time was 7200 sec for each sample. Concentrations of radiocesium in different species of echinoderms were measured and then the T_{eco} values of radiocesium were calculated. The following formulas were used to calculate T_{eco} of radiocesium in echinoderms.

$$1/T_{eco} = 1/T_{eff} \text{ (in situ)} - 1/T_p$$

Where, T_{eff} and T_p denote effective half-life and physical half-life of radiocesium, respectively.

RESULTS AND DISCUSSION

As shown in Fig. 1, the ¹³⁷Cs and ¹³⁴Cs concentrations (converted to log scale) in echinoderm samples tended to decrease over time (426–1175 days after the FDNPP accident). Both radionuclides (¹³⁷Cs and ¹³⁴Cs) concentrations were much higher in *E. cordatum* (318 and 165 Bq/kg-ww, respectively) at Yotsukura station approximately 500 days after the FDNPP accident. In contrary, radiocesium concentrations were much lower in *A. coniferus* (4.25 Bq/kg-WW) and *L. quinaria* (4.45 Bq/kg-WW) at Yotsukura station.

In the case of Ena station, both of ¹³⁷Cs and ¹³⁴Cs concentrations were higher in *E. cordatum* (289 Bq/kg-

WW and 161 Bq/kg-WW, respectively) and lower in *M. nudus* (respectively, 3.55 Bq/kg-WW and 2.08 Bq/kg-WW). At Hirono Thermal Power Plant station, ¹³⁷Cs concentrations were much higher in *M. nudus* (63.7Bq/kg-WW) and lower concentrations were observed in *A. amurensis* (4.21 Bq/kg-WW).

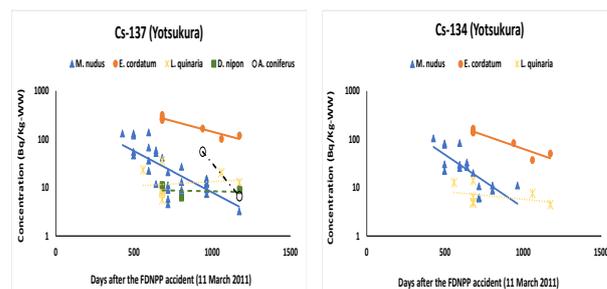


Figure 1: Spatial and temporal changes in the ¹³⁷Cs and ¹³⁴Cs concentrations in different echinoderms collected from Yotsukura station in Fukushima Prefecture after the FDNPP accident. Solid and dashed lines represent statistically significant and insignificant regression slopes, respectively. Different trend lines indicate different samples for ¹³⁷Cs and ¹³⁴Cs (— *M. nudus*, — *E. cordatum*, ... *L. quinaria*, - - - *D. nipon* and - · - *A. coniferus*), respectively. Data below the detection limit are excluded.

Ecological half-lives of ¹³⁷Cs (T_{eco}) in different echinoderms were also estimated using those collected samples from three areas. T_{eco} of ¹³⁷Cs in *M. nudus* and *E. cordatum* were 176 days and 358 days, respectively, and T_{eco} values of ¹³⁴Cs in *M. nudus* and *E. cordatum* were respectively 170 days and 333 days at Yotsukura station. At Ena station, T_{eco} of ¹³⁷Cs and ¹³⁴Cs in *M. nudus* were 358 days and 136 days respectively.

CONCLUSION

It is concluded that variations of spatial environment and food habit may effect on the ecological half-life (T_{eco}) of radiocesium but the mechanism is not clear. So, it would be useful to carry out further analysis of concentration factors and food uptake amounts of various species to improve our understanding of how echinoderms ingest and eliminates radioactive cesium.

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Shell pigments in cultured abalone

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INTRODUCTION

Pigments in the algae upon which abalone feed are thought to color the abalone shell, so the food on which abalone feed can be deduced from analysis of the pigments present in the shell. In this study, pigments were analyzed in the shells of cultured Ezo abalone (*Haliotis nipponensis*), whose feeding environment was known during rearing, to investigate the relationship between the pigment composition detected and the food on which the abalone had fed.

METHODS

The experiments were performed on *H. nipponensis* of mean shell length 30 mm. They were reared firstly on nursery boards covered predominantly by the green alga *Ulva lens*, and then by feeding with artificial feed. These shells had a characteristic green colour (referred to in the industry as 'green mark') derived from the artificial feed. In preparation for pigment analysis, any matter adhering to the shells was removed by brushing the surface. The shells were then ground in a blender (Wonder blender, Osaka Chemical, Osaka). Lipid-soluble and water-soluble pigments were extracted from the ground shells and analysed by high performance liquid chromatography (HPLC; Shimadzu, Kyoto). Lipid-soluble pigments, such as chlorophyll and xanthophyll, were analysed by the method of Zata et al. (2000); and water-soluble pigments, such as phycobiliprotein, by the method of Zolla et al. (1999).

RESULTS AND DISCUSSION

Many pigments detected in the shells were also detected in abalone feed. The major pigments detected in the shells were 19'-butanoyloxyfucoxanthin (But), Chlorophyll *a* (Chl *a*), Chlorophyll *b* (Chl *b*), β -carotene (β -Car), Fucoxanthin (Fuco), 9'-cis-neoxanthin (Neo), Siphonaxanthin (Siphx), Violaxanthin (Viola), and Zeaxanthin (Zea).

Chl *a* and β -Car are pigments found naturally in most plants, including green algae. The nursery boards (to which *U. lens* was attached) contained (in addition to Chl *a* and β -car) Chl *b*, Neo, Siphx, Viola, and also Antheraxanthin (Anthera), Lutein (Lut) and Prasinoxanthin (Proasino). These pigments are contained in green algae. It is considered that Neo, Siphx, and Viola detected in the shells were derived from the pigments of *U. lens*. Viola, Anthera, and Zea are pigments related to protecting plants from photooxidative damage (Havaux and Niyogi 1999), changing from Viola to Anthera to Zea, or Zea to Anthera to Viola, depending on the light conditions. Therefore, it is deduced that Zea detected in the shells was derived from *U. lens*.

From the artificial feed fed to the abalone, derivatives of both Chl *a* and Chl *c* were detected and are considered to be degradation products arising during the thermal drying process prior to measurement (See, for example, Nosan Corporation, <https://www.nosan.co.jp/business/fodder/progress.htm>). In addition to these Chl derivatives, Fuco and Lut were detected and it is considered that Fuco detected in the shell is derived from pigments in the artificial feed.

In addition to Chl *a* and β -car, Chlorophyll *c*₂, Chlorophyll *c*₁, Fuco, and Diadinoxanthin were detected in the culture seawater. These are pigments contained in Bacillariophyceae (diatoms). Alloxanthin, present in Cryptophyta, was also detected in the seawater, suggesting the presence of both diatoms and Cryptophyta in the ambient seawater. Diatoms (which are planktonic and unable to swim) are common in seabed deposits. Therefore, some of the Fuco detected in the seawater may be derived from diatoms deposited to the bottom of water tank and attached to the nursery boards.

Prasino from the nursery boards, Lut from the nursery boards and in artificial feed, and Chl *a* and Chl *c* derivatives found in artificial feed were not detected in the shells. It may be that the latter derivatives were not detected as pigments in the shells because of further degradation. Prasino and Lut were found in lower concentrations than those of chlorophyll pigments, so they may have been beyond the level of detection in the shells, or for some reason do not accumulate in the shell.

The maximum absorption wavelengths of the water-soluble pigments analysed by HPLC were in the range 550-610 nm, so they were considered to be phycocyanin and phycoerythrin-like pigments from the spectrum. These pigments were retained by cyanobacteria and red algae and appear to be derived from the artificial feed.

Overall, it is considered that these results confirm the hypothesis that the "green mark" of abalone shells is indeed derived from coloured pigments in the algal feed, which include various lipid-soluble and water-soluble pigments.

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Effect of naphthalene on phytoplankton

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INTRODUCTION

Chemical substances, such as aromatic hydrocarbons, are known to become more toxic to aquatic organisms when their structure is altered by light (mainly ultraviolet light) and their water solubility increases. This phenomenon is called phototoxicity. While photosynthetic organisms are known to have various photoprotective mechanisms, such as the xanthophyll cycle, to protect them from photooxidative damage in response to excessive light exposure, the combination of high light intensity and chemicals may affect the toxicity or the response of phytoplankton. In this study, three phytoplankton species of differing pigment composition were exposed to naphthalene under different light conditions and investigated for effects on growth and the xanthophyll cycle.

METHODS

Three concentrations (0.05, 0.5, and 5.0 ppm) of naphthalene were administered to three species of phytoplankton: the diatom *Phaeodactylum tricorutum* (Phylum Bacillariophyta, Class Bacillariophyceae), *Pycnococcus provasoli* (Phylum Chlorophyta, Class Prasinophyceae), and *Phormidium* sp. (Phylum Cyanobacteria, Family Oscillatoriaceae). Control preparations contained no naphthalene. All preparations were exposed to white ($200 \mu \text{mol m}^{-2} \text{s}^{-1}$), red ($200 \mu \text{mol m}^{-2} \text{s}^{-1}$, wavelength 625 nm), green ($200 \mu \text{mol m}^{-2} \text{s}^{-1}$, wavelength 530 nm), or intense white light ($2000 \mu \text{mol m}^{-2} \text{s}^{-1}$). Growth was measured by Chl *a* analysis with high performance liquid chromatography (HPLC; Shimazu, Kyoto, using a short column) every 24 h for 72h (14 light :10 dark cycle).

To test for short term reactions, naphthalene (15 ppm to *Phaeodactylum*, and 30 ppm to *Pycnococcus*) was administered under dark conditions, followed rapidly by exposure to white light ($200 \mu \text{mol m}^{-2} \text{s}^{-1}$), or intense white light ($2000 \mu \text{mol m}^{-2} \text{s}^{-1}$). Control preparations contained no naphthalene. After 0 min (dark), 5 min, 15 min, 30 min, and 1 h, changes were measured in the composition of pigments

involved in the xanthophyll cycle. Pigment profiles were measured by HPLC (Zapata et al. 2000).

RESULTS AND DISCUSSION

Phytoplankton growth was inhibited with increasing naphthalene concentration under all light conditions. Growth of the diatom and the prasinophyte showed the largest inhibition under strong white light, while that of the cyanobacterium was most strongly inhibited under red light. The wavelengths affecting these three phytoplankton species differed presumably because of their differing pigment profiles. Unlike diatoms and prasinophytes, cyanobacteria contain phycocyanin, which has a wavelength of maximum absorption in the red (620 nm): this may explain the observed susceptibility to damage by red light in *Phormidium* sp.

The phytoplankton xanthophyll cycle responded fastest where naphthalene addition and intense light coincided.

CONCLUSION

Previous studies have focussed on structural changes to chemical substances during phytoplankton toxicity tests. However, the experiments conducted here focussed on photodamage within the phytoplankton itself. The results showed clearly that the combined effects of photodamage caused by strong light in the presence of a chemical substance increased the toxic effect. In addition, because different taxonomic groups of phytoplankton possess different pigments, the wavelength of maximum photodamage differed accordingly.

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Length-weight relationships for 17 fish species in the Luanhe River Estuary, Bohai Sea, northern China

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INTRODUCTION

Estuarine areas are extremely important areas in the life cycle of some fish species. These ecosystems provide food, shelter, and spawning grounds for varieties of marine organisms. The Luanhe River is a sediment-laden water course on the northern shore of Bohai Bay, China¹. The estuary of the Luanhe River is a famous fishing ground and nursery area for marine organisms within Bohai Bay. This area is recognized as an important feeding and breeding location for migratory species²⁻⁵.

Length-weight regressions (LWR) are an important tool for the proper exploitation and management of fish populations⁶. Length and weight data for fish are needed to estimate growth rates, age structure, and other population dynamics⁷. This information is commonly used in the ecosystem modeling approach⁸ to calculate the production to biomass ratio (P/B) of different functional groups, taking into account that for more precise weight estimates it is advisable to make use of local values. In addition, LWR allow life history and morphological comparisons between different fish species, or between fish populations from different habitats and/or regions⁹. Biological scientists often estimate fish weight in the field using LWR¹⁰.

Prior to this study there was LWR data available for fish species in the Luanhe River Estuary and this study provides the first LWR references for 17 fish species from this area. This study aimed to provide information that could be used for the management of the Luanhe River fishery grounds. The LWR data will be made available through the Fishbase Database¹¹, so that they can be used by other researchers.

METHODS

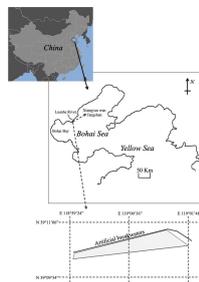


Fig. 1 Schematic map showing the survey area in the northernmost part of Bohai Bay, the Bohai Sea, China.

This study was carried out in the Luanhe River Estuary between longitude E 118°59'24"-119°01'48" and latitude N 39°11'06"-39°09'54" (Fig. 1). Samples were collected at monthly intervals from December 2016 to August 2017 and at bimonthly intervals from July 2016 to November 2017. The fishing gear used for sampling included a crab pot, trammel net of various inner mesh sizes, and a bottom trawl. The standard

length (L) of each specimen was measured to the nearest 0.1 cm using a 30-cm ruler. Fish body weight for all specimens was weighed to the nearest 0.01 g using an electric balance (CR-5000WP, Custom, Japan).

RESULTS AND DISCUSSION

A total of 7354 individuals belonging to 17 species (11 families) were recorded in this study. Linear regressions of log transformed data were highly significant ($P < 0.05$) for all analyzed species. The most abundant species sampled was *Chaeturichthys stigmatias* (N=2483). The best represented family was Gobiidae with 4 species recorded.

The coefficients of determination (R^2) ranged from 0.95 to 1.00 for *Mugil cephalus*, *Sebastes schlegelii*, *Engraulis japonicus*, *Paralichthys olivaceus*, *Tridentiger barbatus*, *Sardinella zunasi*, *Acanthogobius ommaturus*, *Thryssa kammalensis*, *Hexagrammos otakii*, *Chaeturichthys stigmatias*, *Platycephalus indicus*.

LWR slope (b) values ranged from 2.572 for *Acanthogobius ommaturus* to 3.6581 for *Engraulis japonicus*. The median value was 3.114 for *Platycephalus indicus*, although 50% of the values ranged from 2.9451 to 3.2965 for the complete data set.

CONCLUSION

The data collected during this study represents an important contribution of base line data on the LWR of a number of fish species that were previously unavailable. It is important to point out that these LWR should be strictly limited to the length ranges used in the estimation of the linear regression parameters¹⁴. The results obtained in the current study will contribute to the knowledge of fish populations in the important Luanhe River Estuary and also assist fisheries scientists and managers in the future.

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Shipboard observations of the physical marine environment at the mouth of Otsuchi -Funakoshi Bay, Sanriku Coast

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INTRODUCTION

Otsuchi Bay is one of rias bays of Sanriku Coast, Pacific coast ranging 600 km in the northern Japan, and is known for intrusion of cold bottom water from the open ocean¹. It is also known that baroclinic current structures composed of inflow at lower (upper) layer and outflow at upper (lower) layer occur at flood (ebb) tide². Nutrients and dissolved oxygen that are necessary for active aquaculture in the bay are maintained by the inflow of bottom water from open ocean. We surveyed the physical marine environment, that is, water property and current, to reveal the unknown movement of open-ocean water between the bay and the open ocean.

METHODS

We conducted shipboard observations at the mouth of both Otsuchi and Funakoshi Bays by R/V Yayoi (12 tonnage) of International Coastal Research Center, AORI, on 20 July 2015, 12 July 2016, and 29 June 2018 (Fig. 1). In every observation, we operated towed ADCP (V-fin) for sufficient measurement of the bottom currents³ (Photo 1) as well as CTD and shipboard ADCP of R/V Yayoi. Tidal phases at observations were flood tide in 2015, ebb tide in 2016, low tide in 2018.

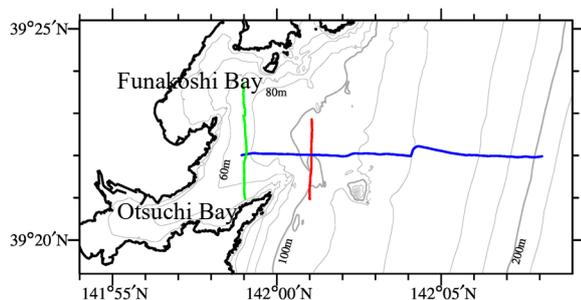


Fig. 1. Observation area and V-fin lines in 2015 (red), 2016 (blue), 2018 (green).



Photo 1. V-fin at the start of towing.

RESULTS AND DISCUSSION

Eastward velocity component (u) at the red line of Fig. 1 in 2015 shows inflow below 30-m depth and outflow in the upper particularly at the north at flood tide. Before end of flood tide, the inflow below 60-m depth weakens. It implies the tidal change has time lag between above

and below 60-m depth. ADCP results at the green line of Fig. 1 in 2018 was too complex to interpret due to low tide as well as low echo intensity that implies low concentration of tiny particles in water (not shown).

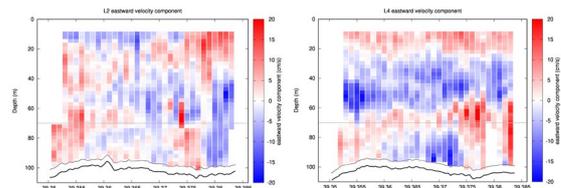


Fig. 2. Eastward velocity component (u) sections at the first half (left) and just before the end (right) of flood tide on 30 July 2015. U above and below 70-m depth are measured by shipboard ADCP and V-fin, respectively.

We tried east-west observation across isobaths towing V-fin along the blue line of Fig. 1 in 2016 and succeeded in measuring offshore bottom current over the continental shelf during ebb tide (Fig. 3).

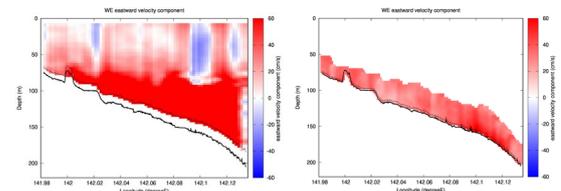


Fig. 3. Eastward velocity component (u) sections during ebb tide on 12 July 2016. Left and right panels are from shipboard ADCP and V-fin, respectively.

CONCLUSION

We confirmed the circulation pattern of the outflow and inflow in the lower layer at ebb and flood tides, respectively. However, the circulation pattern is more complex below 60-m depth at the bay mouth probably due to the time lag between the bottom and the middle depth.

We plan further accumulation of water-property and current data while considering the possibility of mooring observation for investigating temporal variability.

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Changes in thyroxine (T₄) concentrations in the larva and juvenile of marbled flounder *Pseudopleuronectes yokohamae*

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INTRODUCTION

The metamorphosis of flatfish is regulated by thyroid hormone (Inui *et al.*, 1994). Thyroid hormone levels rise during the climax of metamorphosis (Tagawa *et al.*, 1990; de Jesus *et al.*, 1991). The morphological development of marbled flounder *Pseudopleuronectes yokohamae* continues after the climax stage of metamorphosis (G-H Stage), and the K stage, where the lateral line is identified, occurs 70 days post hatching (dph) (Fukuhara, 1988). The levels of thyroxine (T₄), one of the thyroid hormones, fluctuates notably during the metamorphosis of flatfish (Tagawa *et al.*, 1990). This notable fluctuation is known as the T₄ surge. There are limited reports of changes in T₄ levels during the juvenile stage since the post-climax of flatfish. This study investigates the dynamics of T₄ from the larval to juvenile stages of marbled flounders.

METHODS

Marbled flounder artificially fertilized and reared between 2015 and 2019 (except 2017) at the Seed Production Res. Lab., Futtsu Sea Farming Section, Chiba Prefectural Fisheries Research Center, were used as the experimental animals. In 2015, we sampled fish at different developmental stages: F stage (24 dph; pelagic larva), G-H stage (24 dph), I stage (35 dph), J stage (48 dph), and K stage (122 dph) based on Fukuhara (1988). After 2016, samples were differentiated based on dph: at intervals of 10 days for 20-70 dph and 10-30 days for 80-120 dph. The total number of specimens for 4 years were 30, 83, 84, 82, respectively. The whole-body T₄ concentrations in larvae and juveniles were measured and analyzed at the Nikko Field Station, Fisheries Technology Institute. Frozen fish were minced and extracted with the ethanol method (Kobuke *et al.*, 1987). The extracted sample was treated according to Tagawa and Hirano (1987), and the T₄ concentration was measured by ELISA.

RESULTS AND DISCUSSION

The changes in the mean whole-body T₄ concentrations during the larval and juvenile stages are shown in Fig. 1. Approximate T₄ concentrations in different stages were F stage, 0.71 ng/g; G-H stage, 0.85 ng/g; I stage, 1.01 ng/g; and J and K stage, 1.42 and 0.18 ng/g respectively. The highest T₄ concentration was observed in the J stage and not in the G-H stage. The fluctuation was a slight increase. In Fig. 1, the observed increase at 70

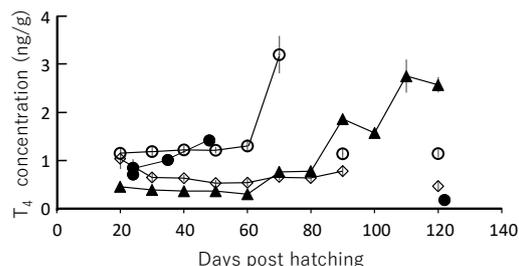


Fig. 1. Changes in the mean whole-body T₄ concentrations from larva to juvenile of marbled flounder *Pseudopleuronectes yokohamae*. (●, 2015; ○, 2016; ◇, 2018; ▲, 2019). Error bars indicate SEM.

dph (2016) and 110 dph (2019) is the T₄ surge. In 2018, the T₄ concentration increased at 90 dph but decreased at 120 dph. We also observed that T₄ dynamics show large annual fluctuations. Tagawa and Kimura (1991) reported that the T₄ concentration of marbled flounder during the metamorphosis (up to 45 dph) was maximum at 30 dph. In this study, the T₄ concentration increased during the metamorphosis period, but the fluctuation was smaller than the T₄ surges in the juvenile stage. The results suggest that the T₄ surge of marbled flounder appears at the final stage of development, rather than at the climax of metamorphosis, and the variation in the appearance time is large.

In the seedling production of marbled flounder, caudal fin loss by nipping is observed during the juvenile stage. Plasma T₄ levels of cherry salmon are negatively correlated with the frequency of nipping behavior, and T₄-treated cherry salmon and brown trout show a reduction in nipping behavior (Hutchison and Iwata, 1998). If T₄ is negatively correlated with nipping behavior, a decrease in T₄ surge should increase juvenile nipping. However, the nipping behavior continues into the juvenile stage. Our data show large fluctuations in post-metamorphosis T₄ concentrations of flatfish. Future studies on the post-metamorphosis T₄ dynamics in other flatfish are needed.

CONCLUSION

The present study investigated the T₄ concentration dynamics in marbled flounder, and it was suggested that T₄ surges appear after 70 dph of the final stages of development. Furthermore, it was observed that T₄ dynamics have large annual fluctuations.

Eustress (Good Stress) and Distress (Bad Stress) in Fish

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Keywords: thermal stressor, oxidative stress, eustress, distress, fish

INTRODUCTION

The production of farmed fish has increased due to the worldwide decline of ocean fisheries stocks. It is known that almost half of the world's fishery production is currently based on aquaculture¹. Thus, appropriate monitoring and evaluation methods for impact and risk of aquaculture on the environment need to be considered^{1,2}. Fish are exposed to various local and global environmental stressors, such as pollutants and acute changes in temperature, and the chances of succumbing to infectious diseases may be increased as a result^{3,4}. Accordingly, the stress induced by environmental stimuli in fish is thought to influence their fitness, productivity, and health. In the present study, we examined the changing patterns of stress and growth-related biomarkers in response to a thermal stress in tropical rabbitfish (*Siganus guttatus*). The results on rabbitfish were compared with those obtained previously in temperate coho salmon *Oncorhynchus kisutch*⁵.

METHODS

Animal experiment: The fish (approx. body weight, 43 g) which were reared at the facility of Sesoko station, TBRC, Univ. Ryukyus, were divided into groups. One group was undisturbed used as a control; the others were exposed to heat shock (+10°C for 2 h). **Measurements:** Lipid peroxides (LPO) were determined as thiobarbituric acid reactive substances. Total glutathione (GSH) levels were measured by a glutathione reductase-recycling method. Heat shock protein 70 (HSP70) levels were determined by immunoblotting. The levels of growth hormone (*gh*) and insulin-like growth factor (*igf1*) mRNA expressions in the tissues were determined by real-time quantitative PCR (qPCR).

RESULTS AND DISCUSSION

Although the plasma LPO levels in rabbitfish were almost the same for all sampling periods, they increased significantly in coho salmon at both 17 h and 48 h post stress⁵. In rabbitfish, GSH decreased gradually and reached its lowest value at 48 h post stress. On the other hand, GSH in coho salmon decreased tentatively at 2 h post stress after which it then increased⁵. HSP70 expression in the liver of both fish species increased at about 17 h post stress⁵.

Pituitary *gh* mRNA expression in rabbitfish gradually increased following heat stress treatment. On the other hand, *gh* mRNA expression in coho salmon pituitary increased at 2 h post stress but returned to control levels at 17 h and 48 h post stress. Hepatic *igf1* mRNA in rabbitfish increased, reaching its highest value at 17 h post stress before decreasing. On the other hand, *igf1*

mRNA in coho salmon liver gradually decreased following thermal stress treatment.

The level of stress-related markers in coho salmon have been changed by stress at initial stage, compared with rabbitfish. These results suggest that temperate fish species such as coho salmon is subject to thermal stress. The changing patterns of markers also suggest that severe thermal stressors can induce oxidative stress in fish. It is known that oxidative stress leads to oxidative damage *in vivo*. However, a moderate level of oxidative stress might modulate important cellular functions⁶. The possibilities of using eustress (good stress) and avoiding distress (bad stress) for animals has been discussed^{7,8}. When the organism is exposed to stimuli that induce distress, a functional physiological state is no longer maintained. However, when the organism is exposed to stimuli that induce eustress, it enters a qualitatively different physiological state, but still maintains homeostasis⁸. Hence, manipulation of appropriate stressor such as moderate thermal treatment and handling stress might be useful to control and improve the health of fish as a eustress⁹. Further studies are needed to reveal the relationships between the oxidative stress, fitness, and health of farmed fish.

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Characteristics of fish species distribution revealed by the surveys of vertical longline and echosounder in Hachirigase Sea Hill, Japan Sea

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INTRODUCTION

The Hachirigase Sea Hill off Mishima Island, Japan, is located in the continental shelf of Japan Sea and is an important spawning and nursery ground to support coastal fisheries. However, fishers concern unplanned/illegal fishing activities are devastating fish resources. Thus, accurate estimates of fisheries resources are needed to use them as a sustainable way. The undulating reefs of Hachirigase Sea Hill prevent us from fish stock surveys using trawl. We applied an acoustic monitoring method to assess the distribution and stocks of fish in this area. This method requires to identify the species of the fishes on the echogram by in situ survey for accurate estimation of the fish abundance. This study examines characteristics of fish species distribution using vertical longline sampling to identify fish species corresponding to fish echoes observed in the Hachirigase Sea Hill.

METHODS

On 21 and 22 June 2006, we conducted vertical longline sampling of fish in Hachirigase Sea Hill on board a fishing boat (6.08 tons). We used the 50 kHz echosounder (FCV-291, Furuno Electric Co., Ltd.) equipped with the ship to measure fish echoes and depths. We set 8 sampling stations over the hill and allocated 10 minutes to catch fish with a vertical longline at a station. If a fish was caught, the sampling time for this station was extended by 10 minutes each time until no more fish were caught. In vertical longline fishing, fish were caught by hanging a line by hand, without the use of floats and fishing rods. To investigate the fish species distributions by depth, we attached 10 branch lines (length: 0.375 m; diameter: 0.285 mm; load capacity: 5 kg) to the main line (diameter: 0.375mm) at intervals of 1.5 m. A 187.5 g weight was attached to the end of the main line. Circle hooks (called as “nemuri-bari” or “mutsu-bari” in Japan) with a curved part (gap: 7 mm; height: 16 mm) were used as fishing hooks attached to the branch lines. Fishing lures or boiled mysids were used as fishing baits. Catch per unit effort (CPUE) of vertical longline fishing was defined as the number of catches divided by the number of sampling times.

RESULTS AND DISCUSSION

We caught 7 species at 7 stations out of a total of 8 sampling stations and in 18 times out of a total of 27 sampling times, resulting in 1.15 CPUE. The ratios of the fish species in CPUE were 43.7 % of threeline grunt

(*Parapristipoma trilineatum*), 37.0% of red lizard fish (*Synodus ulae*) and less than 5% of species, cherry bass (*Sacura margaritacea*), bottom perch (*Apogon semilineatus*), filefish (*Thamnaconus modestus*), John dory (*Zeus faber*) and rockfish (*Sebastes inermis*), respectively. Fork length of the grunt (n=13) caught at six sampling stations (rocky area) of Stn. 2 to Stn. 7 (less than 60 m in bottom depth) were 28.3 ± 4.7 cm (mean \pm SD) and body weight were 440.8 ± 191.9 g. Fork Length of red lizard fishes (n=12) at five stations (rocky and sandy area) of Stn. 4 to Stn. 8 were 17.0 ± 2.5 cm and body weigh were 47.5 ± 31.0 g. For the grunts and red lizard fishes, the sampling bottom depths were 43.3 ± 19.3 m and 55.5 ± 10.0 m, respectively. There was no significant difference in the average bottom depth of the sampling areas where these fishes caught (Mann-Whitney U, $P > 0.05$). Grunts, benthopelagic fish, were caught in the layer between the sea bottom and 9 m above the sea bottom, whereas red lizard fishes, demersal fish, were caught in the narrower layer only between the sea bottom and 3 m above the sea bottom. The mean heights from the sea bottom where grunts (n=13) and red lizard fishes (n=12) were caught were 5.5 ± 2.0 m and 2.0 ± 0.7 m, respectively. There was a significant difference in the average height of the two fish species (Mann-Whitney U, $P < 0.01$).

CONCLUSION

Our surveys revealed that the dominant species distributed below the height of 9 m above the sea bottom in the rocky area shallower than a depth of 60 m, where distributions of fish echo was observed in Hachirigase Sea Hill, was the grunt, which the most important commercial fish there. From the above, it is expected that the method, which assigns fish species to the fish echoes according to the standard set based on the ratio of CPUE by height, will lead to improvement in the accuracy of fish abundance estimation. This study shows that the combination of echosounder and vertical longline is effective in determining the abundance of fish in the sea hill where trawl surveys are difficult.

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New technologies to improve bycatch mitigation in industrial fisheries

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INTRODUCTION

For many years, tremendous effort has been dedicated to developing new industrial tuna fisheries while their adverse impacts on threatened marine species has received relatively little attention. In tuna fisheries bycatch is its major anthropogenic threat to marine megafauna in general, particularly sharks. Research on the development of gear technology for bycatch reduction and potential mitigation measures helped tuna Regional Fisheries Management Organizations adopt bycatch reduction management measures. Despite the urgency in investigating and implementing mitigation measures in fisheries, there are still a number of burgeoning questions inherent in the process: (1) Do we know enough about the capture process? (2) How to better assess the efficacy of mitigation measures in practice? (3) Why mitigation measures are not always transferable? (4) What should we study to innovate effective mitigation measures? and (5) Which tools should we develop in the future? This study presents a history of the development of the techniques and facilities for studying tuna and marine megafauna behaviour and reviews research on the development of mitigation measures for pelagic longline and tropical purse seine, and proposes new perspectives integrating recent technological breakthroughs based on investigations on bycatch behaviours especially sharks and fishing gear dynamics. Lastly, this study highlights new discoveries on fish sentience and their other capabilities recently revealed.

METHODS

By reviewing the literature on the development of mitigation measures it appears that effective methods or promising concepts to avoid capture of unwanted species or/and methods to reduce bycatch mortality arose from four main types of approach: a) shark biology and sensory physiology, b) aggregating behaviour of various species under natural or man-made FADs, c) fishing gear behaviour and marine creatures interactions during the fishing process and d) habitat use.

RESULTS AND DISCUSSION

What is missing in our understanding of fish capture and escape processes? Basically, the efficiency of a measure is evaluated based on data collected by scientists. However, various interactions with fishing gears, different from capture, are generally not observed and remain cryptic. Moreover, no study has tested the responses of sharks and other prey using the same experimental procedures. In other words, results of the experiments were never fully duplicated or extended on other species. This could be the reason why the transfer of a successful mitigation measure from one region to another did not always occur. Understanding species' behaviours especially during capture is essential for formulating further bycatch reduction approaches and to assess the effectiveness of mitigation measures.

New tools to investigate bycatch behaviour and fishing gear dynamics

•Longline and mini AUVs: platform of observation

Instrumented pelagic longline gear could be used to investigate specific animal behaviours, to evaluate a suite of prognostic environmental and operational factors determining the effectiveness of candidate mitigation measures. Furthermore, it requires designing a comprehensive system of "monitored" fishing gear to assess gear dynamics while fishing and the effectiveness of candidate mitigation measures during specific experiments (e.g., hook tests of a different shape or size, bait test, etc...). •*Visual inspection of DFADs and in the purse seine*: The mini AUV could be used to approach and inspect deployed DFADs and to observe at different spatial and temporal scales the occurrence, density and location of pelagic organisms and to monitor the free swimming schools of pelagic species during purse seining operation. Visual inspection could be done using its embedded camera but multiple cameras can be added to record the whole environment. Sonar could be used to assist the drone to track tunas (large swim bladder) to analyze their movements near the DFAD. •*Computer simulation and analysis of fishing gear geometry and dynamics*: The recent development of numerical simulations of the three-dimensional dynamics of fishing nets has brought new insights on purse seine capture processes and perspectives for the development of technical mitigation measures.

CONCLUSION

All the technological innovations proposed will help scientists exploring the mechanisms of interactions of all animals with fishing gears, the causes of differing responses among individuals and species, allowing an understanding of potential adverse effects of the fishing gear on unwanted individuals and how these could be reduced. In the light of the results, simplified technological transfers to fishers could be envisaged giving them the ability to monitor their fishing operations and to make adjustments to maximise catch and avoid or reduce mortality of bycatch.

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