Original article

Diatom resting stage cells and benthic diatoms in the sediment of the Japanese coastal waters

Nami Ako^{1,*}, Kazuyoshi Miayamura², Hideki Iwano³ and Ichiro Imai¹

¹ Graduate School of Fisheries Sciences, Hokkaido University, Hakodate, Hokkaido, 041-8611, Japan

² Fisheries Research Division, Oita Prefectural Agriculture, Forestry and Fisheries Research Center, Saiki, Oita, 879–2602, Japan
³ Fisheries Research Division, Oita Prefectural Agriculture, Forestry and Fisheries Research Center, Bungotakada, Oita, 879–0608, Japan

* Correspondence: anami-0313@eis.hokudai.ac.jp; Tel.: +81-138-40-5543

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Introduction

Many species of planktonic centric diatoms form resting stage cells to endure unsuitable environments for growth, and survive in bottom sediments. The centric diatom resting stage cells in bottom sediments are occasionally resuspended into the water column, and they can germinate and resume vegetative growth [1]. Therefore, seed banks (sediment accumulations of resting stage cells) can be a major factor in plankton dynamics in coastal sea [2]. The morphology and physiology of centric diatom resting stage cells are rather well investigated [3-6]. However, there is a paucity of findings on benthic diatoms in bottom sediments of coastal areas. In the present study, the abundance and taxonomic composition of viable resting stage cells of centric diatoms and vegetative cells of benthic diatoms were investigated using freshly collected bottom sediments. Relative abundance of benthic diatoms are discussed referring environmental conditions.

Materials and methods

The bottom sediments were collected at 5 stations in Suo-Nada and at 11 stations in Saiki Bay, Oita Prefecture, on June 3, 2016. The centric diatom resting stage cells and benthic diatoms (mainly pennate diatoms) in bottom sediments were enumerated by the most probable number method (MPN method) [5] and direct count method within 5 days after the each samplings.

The direct count method was conducted by the following procedure. Aliquots (1 g wet weight) of sediment sample were suspended in the filtered and sterilized sea water at a concentration of 0.1 g wet weight ml^{-1} (10⁰ dilution). Serial tenfold dilutions (10⁻¹ and 10⁻²) were made with the filtered sea water. Sediment suspensions of 10⁻¹ and 10⁻² dilution revels

were put for 12 hours at a temperature of 20 °C under an illumination of 50 μ mol photons $m^{-2} \ s^{-1}$. The centric diatom resting stage cells and benthic diatoms were observed and counted by using an inverted epifluorescence microscope under blue light exicitation. Sediments suspensions 10^{-1} or 10^{-2} dilution were appropriately used to count and more than 100 cells were checked for each samples.

The procedure of the extinction dilution method (MPN method) followed that of Imai et al. (1984) [7].

Results and discussion

By the direct count method, the centric diatom resting stage cells in bottom sediments were distributed from 2.5 x 10^3 to 7.0 x 10^4 cells g⁻¹ wet sediments in Suo–Nada, and from 2.5 x 10^4 to 1.3×10^5 cells g⁻¹ wet sediments in Saiki Bay (Fig. 1). The benthic diatoms ranged from 8.0 x 10^3 to 5.2 x 10^4 cells g⁻¹ wet sediments in Suo–Nada and from 6.0 x 10^3 to 1.4×10^5 cells g⁻¹ wet sediments in Saiki Bay (Fig. 1). Densities tended to be higher in Saiki Bay than Suo–Nada.

Table 1 shows the relationship between water depth and the proportion of benthic diatoms. Benthic diatoms were generally more abundant at shallow water stations than deep water sites. The proportion of benthic diatoms based on MPN method is higher than that obtained with direct count method. By MPN method, the centric diatom resting stage cells in bottom sediments were distributed from 7.8 x 10^2 to 2.6 x 10^4 MPN g⁻¹ wet sediments in Suo–Nada and from 2.9 x 10^3 to 3.3 x 10^4 MPN g⁻¹ wet sediments in Saiki Bay. The vegetative cells of benthic diatoms ranged from 0 to 1.9×10^5 MPN g⁻¹ wet sediments in Suo–Nada and from 4.2 x 10^3 to 1.8×10^5 MPN g⁻¹ wet sediments in Saiki Bay. Relatively higher contributions were observed for *Skeletonema* spp., *Chaetoceros* spp., and *Nitzschia* spp. (Fig. 2).

High densities of resting stage cells of centric diatoms were detected in Suo-Nada and Saiki Bay, suggesting that resting stage cells in the bottom



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sediments well function as a seed population of planktonic diatoms. Different distributional features were noticed for centric diatom resting stage cells and



Fig. 1. Spatial distribution of cell densities and taxa composition of centric diatom resting stage cells and pennate diatoms in the bottom sediments collected from Suo-Nada and Saiki Bay.

Table 1. Comparison of water depth, density of total diatoms and the proportion of pennate diatoms for each station.

Area and Station	Depth - (m)	direct count method		MPN method	
		Total number	Pennate	Total number	Pennate
		of diatoms	diatoms (%)	of diatoms	diatoms (%)
Suo–Nada					
11	13	$7.7 \text{ x } 10^4$	68.0	2.1 x 10 ⁵	87.7
15	18.5	$1.6 \ge 10^4$	51.6	$7.8 \ge 10^2$	0.0
16	10.4	$1.1x \ 10^4$	76.2	$6.5 \ge 10^3$	68.8
19	12.1	6.4 x 10 ⁴	53.5	$3.5 \ge 10^4$	91.5
Nagasu Port	3.6	$9.9 \ge 10^4$	30.3	$2.5 \text{ x } 10^4$	63.3
Collei Deve					
Saiki Bay		4		4	
1	24.3	3.7 x 10 ⁴	16.4	1.1 x 10 ⁴	48.7
2	36.1	6.0 x 10 ⁴	48.3	$7.2 \text{ x } 10^4$	29.4
3	22.9	$9.0 \ge 10^4$	50.6	$1.1 \ge 10^4$	38.0
4	9.6	1.2 x 10 ⁵	52.8	$2.5 \text{ x } 10^4$	28.7
5	9.7	$1.7 \text{ x } 10^5$	80.5	1.9 x 10 ⁵	98.1
6	14.3	$6.8 \ge 10^4$	63.2	$1.7 \text{ x } 10^5$	96.3
7	18.9	$9.8 \ge 10^4$	46.4	$3.9 \text{ x } 10^4$	15.4
8	13.7	2.5 x 10 ⁵	47.9	$3.6 \ge 10^4$	19.2
9	9.7	$6.8 \ge 10^4$	59.6	$1.2 \ge 10^5$	75.6
10	23.2	1.1 x 10 ⁵	41.9	$5.3 \text{ x } 10^4$	33.5
11	14.8	1.9 x 10 ⁵	75.0	1.7 x 10 ⁵	98.3

benthic diatoms (mainly pennates). The resting stage cells of centric diatoms were found with high densities near the estuary of river mouth, where nutrient rich river water is flowing in.



Fig. 2. Cell densities (MPN g^1 wet sediments: \bullet) and taxa composition of the centric diatom resting stage cells and pennate diatoms in the bottom sediments of Suo-Nada (a) and Saiki Bay (b).

Benthic diatoms were generally abundant at shallow water stations because the light reached the bottom sediments as compared with deep sea bottom.

Benthic diatoms tend to be detected with higher proportions by the MPN method. Diatoms in the sediments are incubated for about 1 week for MPN method. Benthic diatoms cells that were not initially detected by direct count method were activated during incubation. It is necessary to investigate whether benthic diatoms in the fields are physiologically active vegetative cells or some kind of resting cells.

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