



# Usefulness of deep-ocean water pumping for the seasonal monitoring of mesozooplankton



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## HIGHLIGHTS

- Seasonal zooplankton monitoring was achieved by deep-water pumping at 2.5-day intervals.
- Zooplankton abundance, biomass and biovolume were highly correlated with each other.
- Benthopelagic zooplankton was quantitatively collected by deep-water pumping.
- Deep-water pumping may be useful for the collection of rare species and materials.

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## ABSTRACT

The Okhotsk Sea is the southernmost seasonally ice-covered ocean in the Northern Hemisphere. Because of the ice coverage during winter, seasonal monitoring of zooplankton is difficult by ordinary ship-board observation. To overcome this issue, zooplankton monitoring of samples collected by deep-ocean water pumping may be useful. In this study, we evaluated seasonal changes in the zooplankton community based on the samples collected by deep-ocean water pumped from a 350-m depth off of Rausu Harbor in the southern Okhotsk Sea at 2.5-day intervals over two and a half years. Zooplankton abundance and biomass ranged from 20–550 inds. m<sup>-3</sup> and 6–902 mg WM m<sup>-3</sup>, respectively. Both parameters showed similar seasonal changes throughout the study period. Copepods were the most dominant taxa throughout the year and accounted for 90% and 76% of the annual mean zooplankton abundance and biomass, respectively. A total of 20 genera and 33 copepod species were observed. *Metridia okhotensis* and *M. pacifica* accounted for 61% and 12% of the annual mean copepod abundance, respectively. In general, the abundance and biomass of zooplankton collected by pumped-up deep-ocean water were lower than they were in the epipelagic layer (both characteristics of the former corresponded to approximately 60% of the latter). The combined effects of low zooplankton density at the deep layer (350 m) and the low filtering rate of deep-water pumping are considered to be a possible cause of the low zooplankton abundance and biomass in the pumped-up deep-ocean water. For the zooplankton samples from pumped-up deep-ocean water, there was a high abundance of benthopelagic mysids and copepods (*Xanthocalanus* spp.). Because the water inlet of the deep-ocean water pumping is located near the sea bottom, the collected samples may be useful for the evaluation of seasonal changes in the population structure of the less-studied benthopelagic species.

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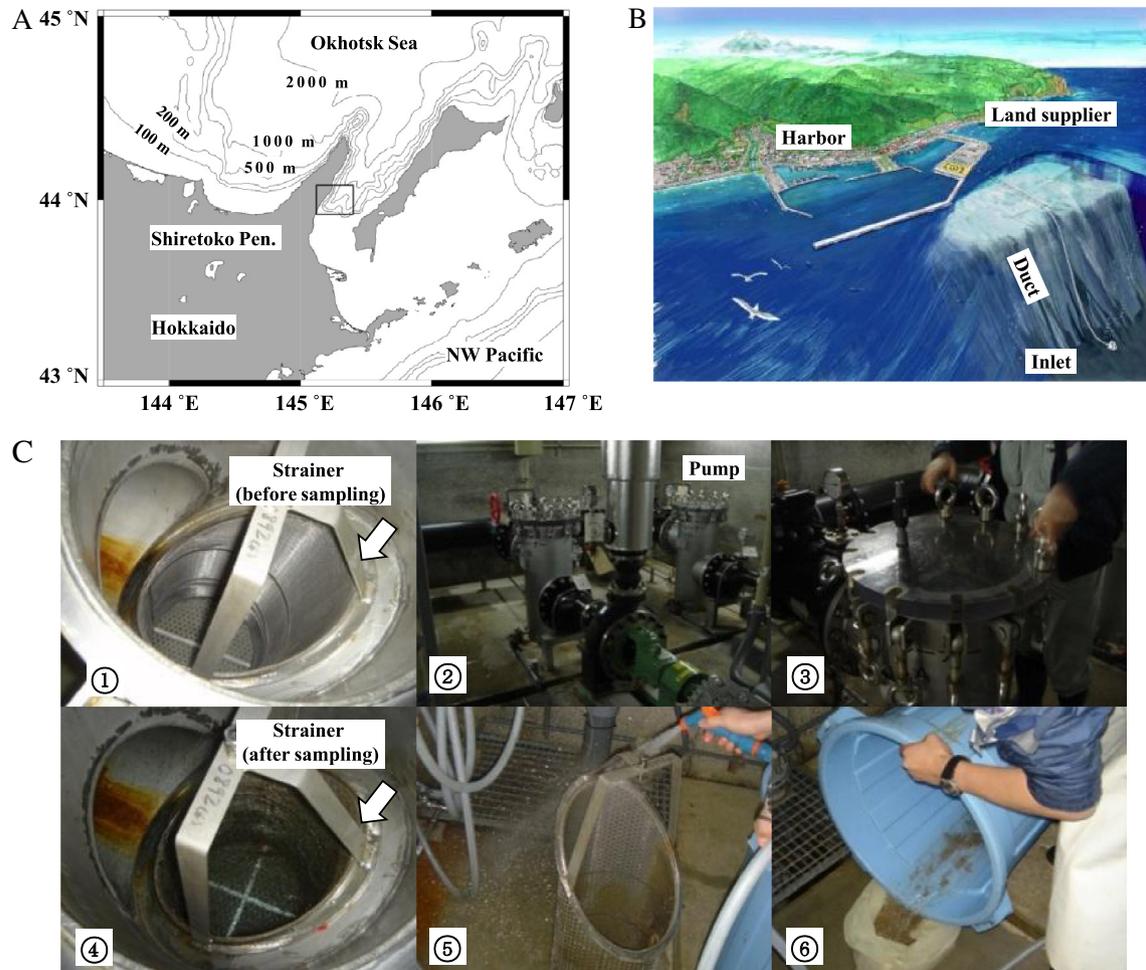
## 1. Introduction

The Okhotsk Sea is the southernmost seasonally ice-covered ocean in the Northern Hemisphere (Parkinson and Grantz, 1983). In the Okhotsk Sea, phytoplankton bloom is observed during the

spring, and zooplankton (mainly composed by copepods) ingests phytoplankton and grows during that time (Pinchuk and Paul, 2000). Zooplankton is preyed upon by pelagic and demersal fish, marine birds and mammals; thus, they play an important role in energy transfer between primary producers and higher trophic levels (Dulepova, 1998; Volkov, 2000; Tamura and Fujise, 2002). Monitoring zooplankton with fine temporal resolution is difficult by ordinary ship-board observation during periods when temporal ice covers the ocean. As a result, all of these studies were conducted during ice-free seasons.

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**Fig. 1.** Location of the Rausu harbor (box), Shiretoko Peninsula, eastern Hokkaido (A), schema of the Rausu deep-ocean water sampling site (B), and the procedure for sampling in the land supplier (C). The time course of sampling in the land supplier is shown with circled numbers (1–6).

The town of Rausu, located on the southern coast of the Okhotsk Sea, collects deep-ocean water by pumping it up from a depth of 350 m every day for the purpose of fishery product refrigeration, food processing and other industrial needs (Takahashi and Yamashita, 2005; Takahashi et al., 2014). The particles contained in the deep-ocean water (mainly zooplankton) are removed through straining (mesh size: 420  $\mu\text{m}$ ) at the Rausu deep-ocean water sampling site. Because year-around zooplankton samples may be temporally covered by fine time resolution, analysis of these zooplankton samples collected at the deep-ocean water sampling site may provide valuable information on the zooplankton community in the Okhotsk Sea.

In this study, we collected zooplankton samples at the Rausu deep-ocean water sampling site with fine temporal resolution (2.5-day interval in average) over two and a half years during June 2007–December 2009. Based on the preserved zooplankton samples, seasonal changes in zooplankton abundance, biovolume and taxonomic composition were analyzed. The results were compared with those of the pelagic samples collected by net-towing, and the characteristics of the zooplankton community collected by the deep-ocean water pumping are discussed.

## 2. Materials and methods

### 2.1. Zooplankton sampling

Seasonal zooplankton samples were collected at the Rausu deep-ocean water-sampling site at the Rausu harbor (44° 00'N,

145° 15'E) in the southern Okhotsk Sea (Fig. 1(A)). This facility can pump up deep-ocean water from an inlet pipe (268 mm diameter) at a depth of approximately 350 m, 2.8 km off from the shore with a maximum speed of 200  $\text{m}^3 \text{h}^{-1}$  (Fig. 1(B)). The deep-ocean water was filtered through a strainer (mesh size: 420  $\mu\text{m}$ ), and the remaining zooplankton was collected (Fig. 1(C)). After 3–135 (mean: 20 h) hours of deep-ocean water pumping at a speed of 106  $\text{m}^3 \text{h}^{-1}$ , the remaining zooplankton in the strainer was collected and preserved with 5%–10% (v/v) formalin. A total of 352 zooplankton samples were collected during 20 June 2007–18 December 2009 (mean sampling interval: 2.5 days = 897 day/352 samples). The temperature and volume of the filtered water ( $\text{m}^3$ ) were recorded at the same time.

### 2.2. Sample analysis

In the laboratory, the settling volumes of all of the samples were measured with a precision of 10 ml. Within the 352 samples, microscopic observation was made for the 62 samples selected to cover the approximately two-week interval throughout the sampling period. For each sample, subsamples (1%–3% of the total volume) were taken with the aid of a wide-bore pipette, and identification and enumeration were performed at the taxonomic level. Due to our sampling methods, damaging some of the specimens during collection was inevitable. However, the resulting damage had little effect on the identification of crustacean copepods. For each taxon, the biomass (wet mass: WM) was measured by

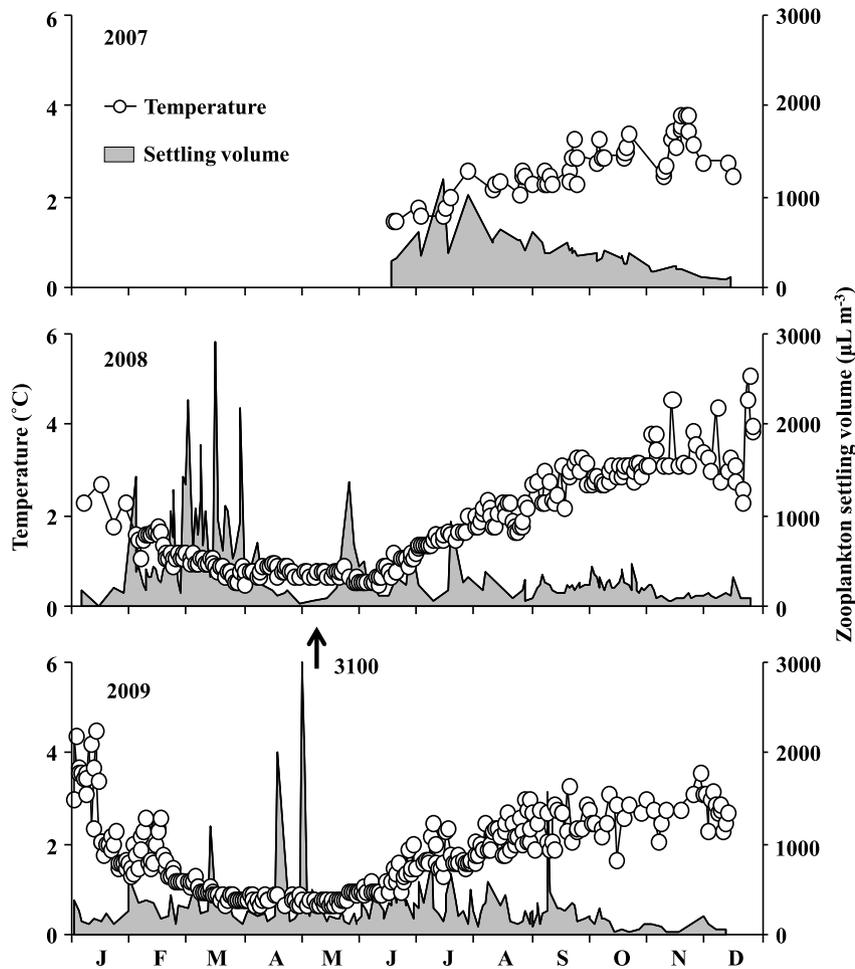


Fig. 2. Seasonal changes in temperature and zooplankton settling volume in the Rausu deep-ocean water during June 2007–December 2009.

an electronic microbalance with a precision of 10 mg using pre-weighed mesh. After the measurement of WM, species identification and enumeration were performed on the most numerous taxa, i.e., calanoid copepods. Species identification of the calanoid copepods was performed according to Brodsky (1967) and Ohtsuka et al. (1997).

### 3. Results

The temperature of the deep-ocean water from Rausu ranged from 0.5 to 5.1 °C, with a maximum temperature reached around November to early January and the minimum during March to May (Fig. 2). The zooplankton settling volume ranged from 100 to 3100  $\mu\text{L m}^{-3}$ , was high in March 2008 and late April 2009, and was low during November to January, the high temperature season (Fig. 2).

The zooplankton abundance ranged from 20 to 550 inds.  $\text{m}^{-3}$  and demonstrated similar seasonal changes to those of the settling volume (Fig. 3). Copepods were the most dominant taxa throughout the year and included 90% of the annual mean total zooplankton abundance. Following copepods, chaetognaths were the second most dominant taxa and reached nearly half of the total abundance during March–April 2009.

Zooplankton biomass ranged from 6 to 902 mg WM  $\text{m}^{-3}$  and had seasonal changes in parallel with abundance (Fig. 4). Similar to abundance, copepods were the most dominant taxa throughout the year (annual mean: 76%), and chaetognaths were the secondary dominant taxa (10%). It should be noted that the less abundant taxa (mysids) had a certain composition (9%) in biomass.

Throughout the study period, a total of 33 calanoid copepod species belonging to 11 families and 20 genera were observed (Table 1). Within the copepods, *Metridia okhotensis* was the most dominant species (61% of annual mean copepod abundance), followed by the congener *M. pacifica* (12%). The abundant season varied between two *Metridia* species; thus, *M. okhotensis* was dominant from December to July, while *M. pacifica* was dominant during August to November (Fig. 5). Other species, *Bradydium pacificus*, *Neocalanus flemingeri* and *Xanthocalanus kurilensis*, accounted for approximately 4% of the total copepod abundance (Table 1). From the perspective of seasonal change, *Xanthocalanus* dominated during November–December 2007. It also should be noted that in both April of 2008 and 2009, the proportion of *Metridia* species suddenly decreased, and consequently, those of the other species increased.

### 4. Discussion

#### 4.1. Zooplankton monitoring by pumped-up deep-ocean water

Oceanic zooplankton is commonly collected by ship-board net towing. From ship-board observations, the sampling intervals tend to be long (nearly one month) and the evaluation of detailed temporal changes is difficult in most cases. To overcome these problems, analyses on zooplankton samples that are collected by a mooring sediment trap with fine-time resolution throughout the year may be useful. From zooplankton analysis on sediment trap samples, horizontal advection of oceanic zooplankton in Arctic fjords (Willis et al., 2006), maturation and reproduction of *Calanus*

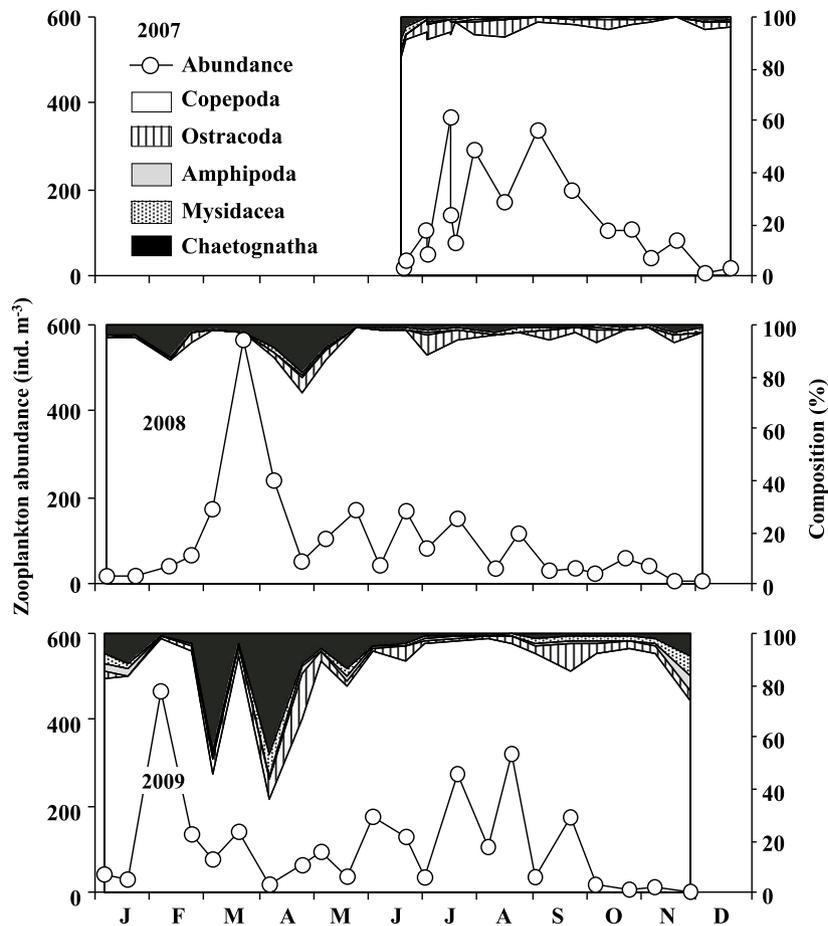


Fig. 3. Seasonal changes in zooplankton abundance and their taxonomic composition in the Rausu deep-ocean water during June 2007–November 2009.

*hyperboreus* under sea ice in the Amundsen Gulf (Ota et al., 2008), responses of the zooplankton community on early summer phytoplankton bloom in the western North Pacific (Ohashi et al., 2011) and year-round population structures of dominant copepods in the Arctic Ocean (Matsuno et al., 2014) were evaluated. Also in the Okhotsk Sea, Shimizu (2009) applied a sediment trap to monitor seasonal changes in zooplankton. While there are advantages because the sediment trap collects passive ocean flux, less quantitative collection of zooplankton and somewhat of a bias on abundance are inevitable for zooplankton analysis based on the sediment trap samples (cf. Buesseler et al., 2007). Compared to the sediment trap, the pumped-up collection of deep-ocean water (Takahashi and Yamashita, 2005; Takahashi et al., 2014) may be a more accurate quantification (note that the sediment trap collects samples passively), with high-frequency for zooplankton sampling.

Within the Rausu deep-ocean water, zooplankton settling volume, abundance and biomass showed highly significant correlation with each other ( $p < 0.001$ , Fig. 6). Spike-like fluctuations in settling volume (Fig. 2) were also the result of abundance and biomass. Zooplankton abundance in deep-ocean water ranged from 20 to 550 inds.  $m^{-3}$  (Fig. 3). These values are lower than those at the epipelagic layer in the southern Okhotsk Sea during April–July (100–800 inds.  $cm^{-3}$ ; Asami et al., 2009). Zooplankton biomass in this study ranged from 6–902 mg WM  $m^{-3}$  and is lower than the mean epipelagic biomass around Kuril Islands during July–August (1000 mg WM  $m^{-3}$ ; Shuntov et al., 1993). A possible reason for low zooplankton abundance and biomass in the deep-ocean water, low zooplankton density at deep layers (350 m) and the slow filtering rate also should be considered. The mean

filtering rate of deep-ocean water was  $106 \text{ m}^3 \text{ h}^{-1}$  throughout the sampling period. This filtration is expressed as  $1.8 \text{ m}^3 \text{ min}^{-1}$  ( $=106/60$ ). To make quantitative comparisons with standard net zooplankton sampling, filtration of NORPAC net (Motoda, 1957) from a 150-m depth with speed of  $1 \text{ m s}^{-1}$  is compared. If the filtering efficiency of the net is assumed to be 95%, the filtering volume of a 45-cm diameter NORPAC net is calculated as  $22.7 \text{ m}^3$  ( $=\pi \times [0.45/2]^2 \times 150 \times 0.95$ ). The towing from 150 m at a speed of  $1 \text{ m s}^{-1}$  requires 2.5 min ( $=150/60$ ). Thus, the filtering rate of a standard NORPAC net is calculated as  $9.1 \text{ m}^3 \text{ min}^{-1}$  ( $=22.7/2.5$ ). Therefore, compared with the standard NORPAC net, the filtering rate of pumped-up deep-ocean water ( $1.8 \text{ m}^3 \text{ min}^{-1}$ ) was at approximately 20% ( $=100 \times 1.8/9.1$ ). Zooplankton avoidance under this slow filtering rate is also considered to be a possible cause of the low zooplankton abundance and biomass of pumped-up deep-ocean water.

At the Rausu deep-ocean water sampling site, in addition to the zooplankton in this study, various deep-sea fish, shrimp and benthos were also collected. Based on the specimen collected by the Rausu deep-ocean water, one new liparid fish (*Careproctus rausuensis*) was described (Machi et al., 2012). For the other deep-ocean water sampling site, fluorescent protein was isolated from the copepod *Chiridius poppei* collected by the deep-ocean water sampling site (inlet at 321 m depth) in Toyama Bay, southern Japan Sea (Masuda et al., 2006). Later, the fluorescent protein was patented and developed into a new fluorescent *Torenia* flower for commercial base by combining flower genomes (Sasaki et al., 2014). These facts suggest that the deep-ocean water is useful for collecting rare marine organisms and new materials.

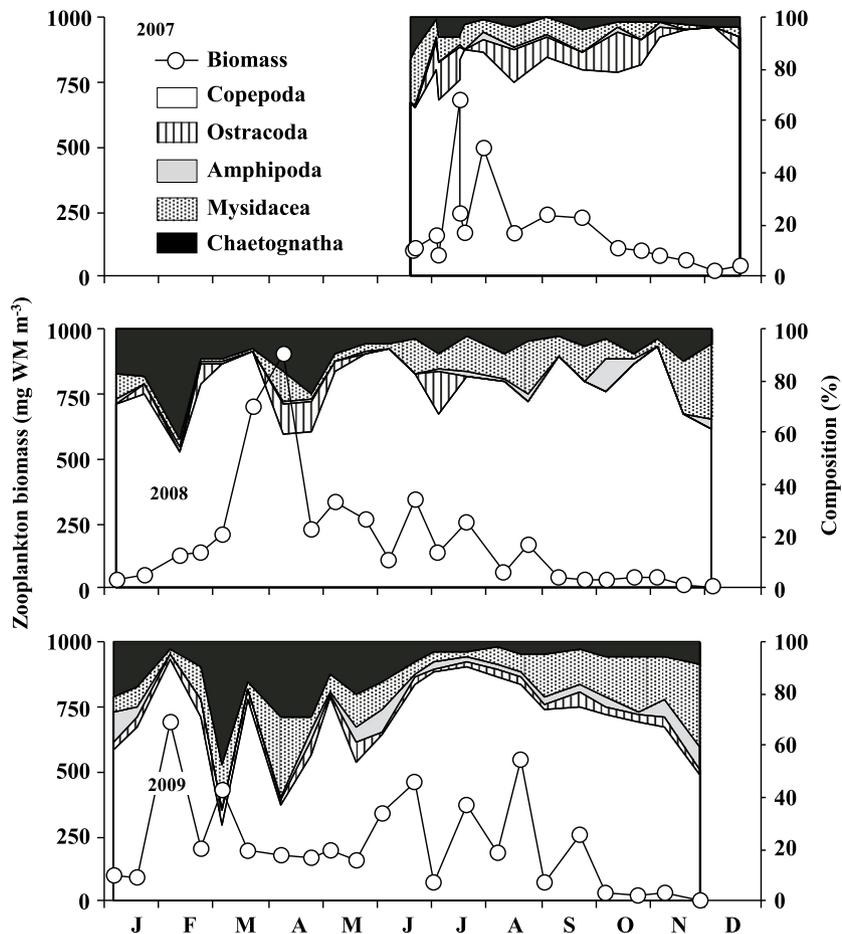


Fig. 4. Seasonal changes in zooplankton biomass (wet mass: WM) and their taxonomic composition in the Rausu deep-ocean water during June 2007–November 2009.

#### 4.2. Characteristics of zooplankton community

In the Rausu deep-ocean water, most of the zooplankton species corresponded to the oceanic species in the Okhotsk Sea. However, within the dominant species of this study, mysids and copepod *Bradyidius* and *Xanthocalanus* spp. are very rare oceanic fauna (Pinchuk and Paul, 2000). Mysids, *Bradyidius* spp. and *Xanthocalanus* spp. are known as benthopelagic species (Grice and Hülsemann, 1970; Grice, 1972). The anomalous high abundance of these benthopelagic species in this study (Fig. 4, Table 1) may be a reflection of the water inlet of deep-ocean water located near the sea bottom. Thus, because of the inlet located near the sea bottom, zooplankton collected by deep-ocean water may have characteristics of benthopelagic fauna compared with the ordinary pelagic net plankton samples.

Both zooplankton abundance and biomass had peaks around winter to spring (Figs. 3, 4). These peaks were caused by the seasonal changes in copepods, especially for the most dominant *M. okhotensis* (Fig. 5). It is well known that *M. okhotensis* is the predominant species both in zooplankton abundance and biomass in the southern Okhotsk Sea (Asami et al., 2009; Yamaguchi, 2009; Shimada et al., 2012). The zooplankton community in the Okhotsk Sea is reported to be divided into 4 groups (Pinchuk and Paul, 2000), and the zooplankton community in this study is similar to the “Central Okhotsk Sea Group” classification of Pinchuk and Paul (2000). The “Central Okhotsk Sea Group” includes numerous small copepod *Pseudocalanus minutus* (Pinchuk and Paul, 2000). However, because of the coarse mesh size of this study (420  $\mu\text{m}$ ), the small-sized *P. minutus* (1–2 mm in adults) may not have been quantitatively collected in this study.

For a number of calanoid copepod species, occurrences of 18 genera and 26 species were reported in the western Okhotsk Sea during the summer (Itoh et al., 2014). This number of copepod species is similar to this study (20 genera and 33 species). The slightly high species number of this study compared with Itoh et al. (2014) may be caused by differences in the sampling methods. Thus, this study included deep-sea and benthopelagic species collected by deep sampling of the inlet (350 m) compared with net sampling from the epipelagic layer (0–150 m) of Itoh et al. (2014).

In conclusion, because the water inlet is located near sea bottom, zooplankton collected by pumped-up deep-ocean water does not always correspond to those of the pelagic samples collected by the plankton net. Despite this shortcoming, zooplankton sampling by deep-ocean water sampling at this site makes it possible to collect deep-sea plankton with fine time resolution throughout the year. As a site for the collection of rare deep-sea species and materials, this deep-ocean water sampling site may be useful for future studies.

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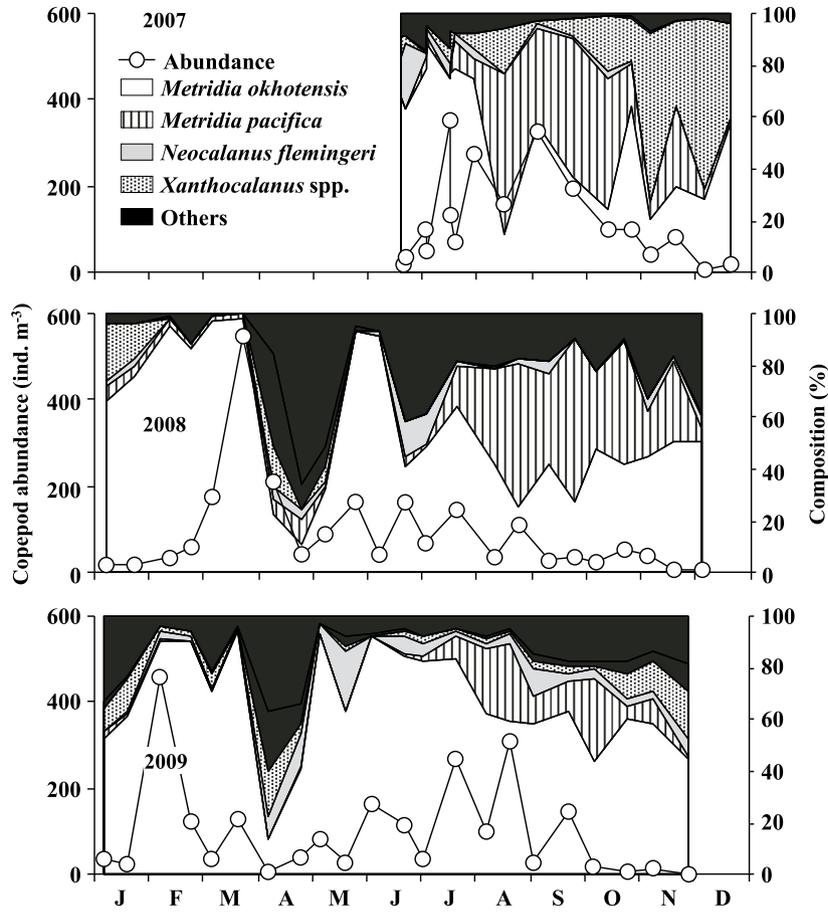


Fig. 5. Seasonal changes in copepod abundance and their species composition in the Rausu deep-ocean water during June 2007–November 2009.

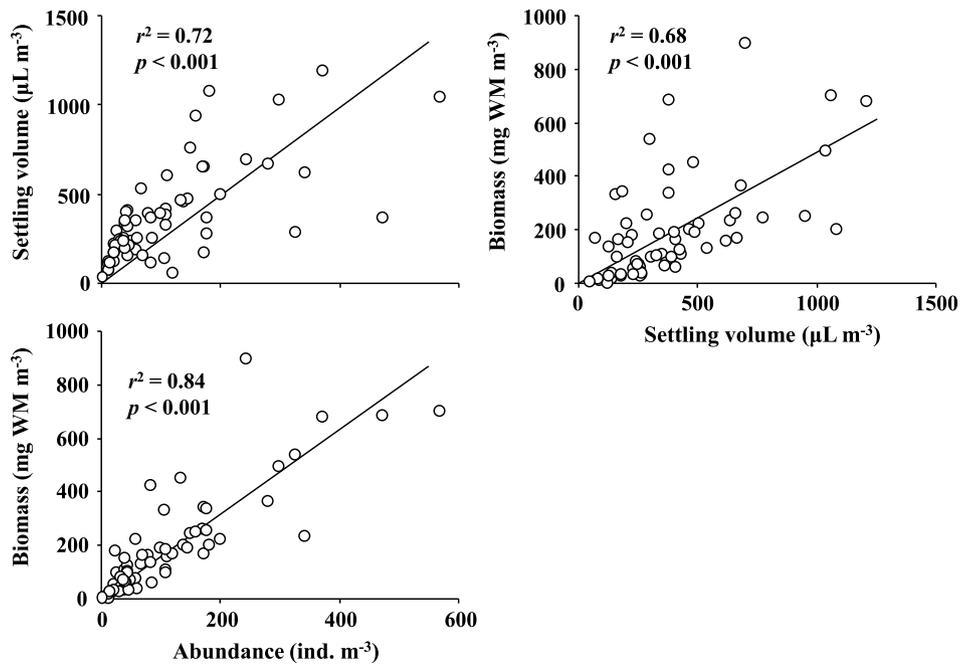


Fig. 6. Relationships between zooplankton abundance, settling volume and biomass collected from the Rausu deep-ocean water during June 2007–November 2009.

**Table 1**

List of calanoid copepod species identified from the Rausu deep-ocean water during June 2007–November 2009. Note that species identification was only made on large-body sized Calanoida. Based on the grand mean of abundance throughout the sampling period, species composition was calculated.

Family	Species	Composition (%)
Aetideidae	<i>Aetideus pacificus</i>	<0.1
	<i>Bradyidius pacificus</i>	4.0
	<i>Chiridius</i> spp.	<0.1
	<i>Gaetanus brevispinus</i>	<0.1
	<i>Gaetanus minutus</i>	0.2
	<i>Gaetanus simplex</i>	0.4
	<i>Gaetanus tenuispinus</i>	<0.1
	<i>Pseudochirella polyspina</i>	<0.1
	<i>Pseudochirella spinifera</i>	<0.1
	<i>Candacia bipinnata</i>	<0.1
Candaciidae	<i>Candacia columbiae</i>	0.3
	<i>Calanus glacialis</i>	1.3
Calanidae	<i>Calanus pacificus</i>	0.2
	<i>Neocalanus cristatus</i>	0.9
	<i>Neocalanus flemingeri</i>	4.8
	<i>Neocalanus plumchrus</i>	1.0
	<i>Centropages abdominalis</i>	<0.1
Centropagidae	<i>Microcalanus pygmaeus</i>	1.0
	<i>Pseudocalanus minutus</i>	0.6
Clausocalanidae	<i>Eucalanus bungii</i>	1.3
	<i>Euchaeta elongata</i>	3.1
Euchaetidae	<i>Pareuchaeta rubra</i>	<0.1
	<i>Heterorhabdus tanneri</i>	0.5
Heterorhabdidae	<i>Heterostylites major</i>	<0.1
	<i>Metridia pacifica</i>	11.6
	<i>Metridia okhotensis</i>	61.4
	<i>Metridia</i> sp.	0.2
Metridinidae	<i>Pleuromamma scutullata</i>	0.1
	<i>Xanthocalanus kurilensis</i>	4.0
	<i>Xanthocalanus</i> sp.	0.7
Phaennidae	<i>Racovitzanus antarcticus</i>	<0.1
	<i>Scolecithricella minor</i>	0.5
Scolecitrichidae	<i>Scolecithricella ovata</i>	<0.1

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