



Diet source of *Euphausia pacifica* revealed using carbon- and nitrogen-stable isotopes in the Yellow Sea Cold Water Mass in summer

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Abstract

Euphausia pacifica (*E. pacifica*) is an important food source for fish and other organisms in marine ecosystems. We analyzed the carbon- and nitrogen-stable isotope ratios in potential diet sources to determine the feeding characteristics of *E. pacifica* in the Yellow Sea Cold Water Mass (YSCWM), a habitat for overwintering *E. pacifica* was collected in the spring and summer from the central and coastal parts of the Yellow Sea from 2012 to 2014, and potential prey were collected in summer 2014. Although the $\delta^{13}\text{C}$ values of *E. pacifica* differed significantly in spring each year, they narrowed to a specific range in summer. The $\delta^{15}\text{N}$ values were always 2‰ higher in summer than in spring, suggesting that the diet of *E. pacifica* inhabiting the YSCWM was limited. In coastal stations, the contribution of any major food source calculated using an isotope mixing model was not more than 20%, suggesting that *E. pacifica* is omnivorous. However, at two stations in the YSCWM, 2.0–5.0-mm and > 5-mm plankton contributed 42 and 22% of their diet on average, respectively. These results indicate that *E. pacifica* inhabiting waters affected by cold water feeds on relatively large zooplankton such as the copepods *Calanus sinicus*. Therefore, the YSCWM likely plays a role in the trophic shift of *E. pacifica* during the summer.

Keywords *Euphausia pacifica* · Stable isotope · Diet source · Overwintering · Yellow Sea Cold Water Mass

1 Introduction

Euphausiids are the dominant taxa among zooplankters that inhabit coastal and offshore regions (Marshall 1979). They play an important role in the transport of organic matter and energy from the surface layer, where primary production takes place, to a deeper layer, and from zooplankton

of the upper trophic level to nekton and even seabirds (Mauchline and Fisher 1969). They constitute 8–15% of the planktonic organism biomass that inhabits the epipelagic and mesopelagic waters of the Northwest Pacific (Aizawa 1974). In addition, they have species-specific distribution, water temperature preference and tolerance, day and night vertical movement patterns, and feeding habits (Lalli and Parsons 1997; Sogawa et al. 2013). These specificities have been studied in various topographical areas. For example, *Euphausia pacifica* (*E. pacifica*), which is found in most areas in the North Pacific (Feinberg et al. 2013), has different vertical distributions and physiological and life patterns in different regions from the subtropical to subarctic (Gómez-Gutiérrez et al. 2006; Iguchi and Ikeda 1995). Since euphausiids inhabiting different regions have specific correlations with ambient ocean conditions, identifying their ecological characteristics makes it possible to more clearly elucidate the relationship between the specific water mass and community structure (Sogawa et al. 2013). They are important secondary consumers in coastal ecosystems, major feeders of primary producer phytoplankton, and major

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food sources for many commercial fish species (Chae et al. 2008; Sugisaki and Kurita 2004).

The Yellow Sea is a semi-enclosed marginal sea of the Western North Pacific, located on the continental shelf (Li et al. 2016) between China and Korea. It has an average depth of 44 m, and its central region extends in a north–south direction parallel to land with a depth of ≥ 70 m (Wei et al. 2016). Owing to the particular topography, different water temperatures, and hydrodynamic factors, the seasonal thermocline in the Yellow Sea covers a cold water mass low in the water column, resulting in the formation of a unique cold water mass known as the Yellow Sea Cold Water Mass (YSCWM; Ho et al. 1959; Yu et al. 2006, Fig. 1). This water column is very firmly seated in the lower layer during summer, when the surface water temperature rises sharply (Zhang et al. 2008). The YSCWM begins to form in spring concurrent with thermocline development; it is most strongly developed from July to August before gradually weakening as the thermocline dissipates. In November, the thermocline and the YSCWM simultaneously disappear (Xu et al. 2016). The YSCWM has a considerable influence on the growth and propagation of phytoplankton that inhabit the Yellow Sea (Ho et al. 1959; Wang 2001; Yu et al. 2006; Zhang et al. 1996).

The spatial distribution of *E. pacifica* in the Yellow Sea appears to vary seasonally, which is closely related to the developmental stage, seawater temperature, and concentration of chlorophyll *a* (chl-*a*; Sun et al. 2011; Yoon et al.

2000, 2006). In particular, adults inhabit water with temperatures below 10 °C in summer. This characteristic is similar to that of *Calanus sinicus* (Wang et al. 2003). The biomass and production of zooplankton larger than 1 mm when the YSCWM appeared were higher inside than outside the cold water mass, whereas zooplankton smaller than 1 mm showed opposite results (Huo et al. 2012). In previous studies, relatively large zooplankton, such as *E. pacifica*, were found in the YSCWM in summer because they avoid high temperature in the surface layer. The feeding characteristics of the zooplankton in the cold water mass has, however, not yet been studied.

Stable isotope analysis is useful for studying the nutritional pathways in a food web that understands organic matter transfer and energy flow from basal organisms to top predators (Fry 2006; Layman et al. 2012). One of the greatest advantages of stable isotope analysis in food web studies is that information on stable isotope ratios in organisms can provide time-integrated information rather than snapshots obtained from gut content analysis. The carbon-stable isotope ratio is suitable for identifying the primary carbon source underlying the food web (Peterson 1999), and the nitrogen-stable isotope ratio is a good tracer to determine the trophic position of organisms (Cabana and Rasmussen 1996); it is most commonly used to understand the energy flow of food webs (Grey 2006). Furthermore, the isotopic mixing model, which utilizes a combination of stable isotope ratios for feeder and potential food sources, can be used to calculate the relative contribution of each food source for the consumer (Parnell et al. 2010).

In the present study, we investigate the feeding characteristics of *E. pacifica* at the YSCWM development stage by using carbon- and nitrogen-stable isotopes. The ecological importance of the YSCWM as the over-summering site of *E. pacifica* is discussed on the basis of the results.

2 Materials and methods

2.1 Collection of zooplankton including *E. pacifica*

Plankton sampling was conducted on the research vessel *Eardo* (Korea Institute of Ocean Science and Technology). The collection of *E. pacifica* was performed at four stations in the Yellow Sea in spring (April) prior to YSCWM formation and summer (August) from 2012 to 2014, when the YSCWM was strongly developed (Fig. 1). After confirming the distribution depth of *E. pacifica* by using a scientific echo sounder (DT-X, BioSonics Inc., USA), a conical net (1 m in diameter with mesh size of 330 μm) was set to the corresponding water depth with a blinker, set for more than 5 min and was then hauled vertically. In 2014, the collection of spring samples was not possible owing to a ship accident

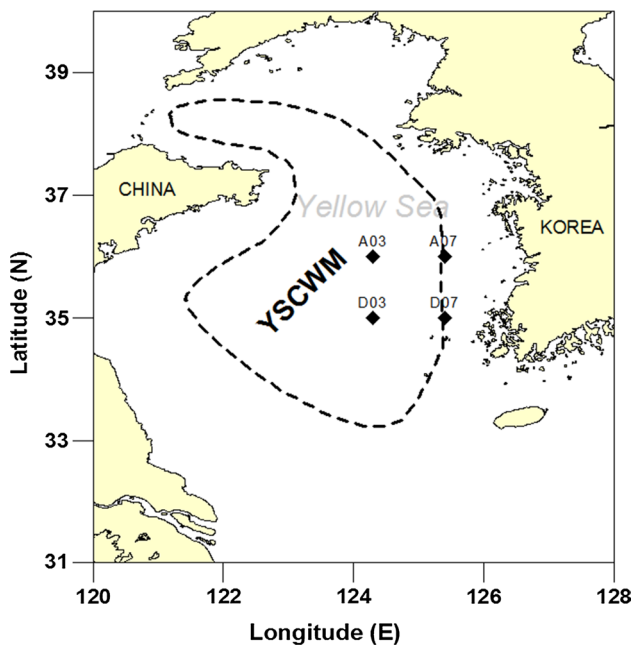


Fig. 1 Sampling stations in the Yellow Sea. The dashed line represents the general range of the Yellow Sea Cold Water Mass in summer (redrawn from Wei et al. 2016)

near the survey station, and sampling was conducted only in summer. Zooplankton, a potential food source of *E. pacifica*, were collected by vertical tow from the bottom to the surface with a zooplankton net (60 cm in diameter with mesh size of 200 μm) at the same stations for *E. pacifica* sampling in August 2014. The zooplankton were classified by size using pore sieves of 5, 2, 1, 0.5, and 0.2 mm. The length of *E. pacifica* was measured using a ruler in mm after collection. All samples were stored at $-20\text{ }^{\circ}\text{C}$ in a freezer in a pre-combusted glass jar until analysis.

2.2 Water environment and collection of size-fractionated plankton (particulate organic matter)

The physico-chemical properties of water such as temperature, salinity, fluorescence, and dissolved oxygen in summer (August) from four stations in the Yellow Sea were measured by using a conductivity, temperature, and depth (CTD) device (SBE 911plus, SeaBrid, USA). The subsurface chlorophyll maximum (SCM) layer was determined by using a fluorescence sensor attached to a CTD on a Rosette sampler. Water samples (40 L) were collected from the SCM layer and the layer 5 m above the bottom. The collected seawater was filtered by a 200- μm sieve to remove large-sized particles and was then passed through a pre-combusted Whatman glass fiber filter (GF/D) with a pore size of 2.7 μm and diameter of 25 mm to collect nanoplankton (2–20 μm) and microplankton (20–200 μm). The filter was also used to collect particles 2.7–200 μm in size. Seawater passing through the GF/D was filtered by using a Whatman GF/F with a pore size of 0.7 μm to collect particles 0.7–2.7 μm , which is similar to the size of picoplankton (0.2–2.0 μm). The filter samples were divided for carbon- and nitrogen-stable isotope analyses, and each sample was replicated three times. After filtration, the filter was placed in a Petri dish, blocked with a foil, and stored in an on-board freezer at $-20\text{ }^{\circ}\text{C}$.

2.3 Stable carbon and nitrogen isotope analysis

All samples collected on board were transferred to the laboratory in an icebox containing dry ice and were stored in a deep freezer at $-80\text{ }^{\circ}\text{C}$ until pre-treatment for stable isotope analysis. Frozen samples were dried in a freeze dryer, and zooplankton samples were pooled by size, powdered, and homogenized using a mortar and pestle. *E. pacifica* samples from 2012 and 2013 were pooled and used for stable isotope analysis. In 2014, samples were individually analyzed. Homogenized biological samples were subsampled for carbon- and nitrogen-stable isotope analyses. Subsamples for carbon were used to remove inorganic carbon and lipids to prevent the interference of carbon other than the assimilated carbon in the whole

body. Biota subsamples for carbon were soaked in 1 N HCl, shaken overnight for remove inorganic carbon, washed with ultrapure water three times to remove HCl, and dried. After the addition of chloroform/methanol (2:1, vol:vol), removed lipids were extracted by ultrasonication. The supernatant was discarded three times to completely remove the lipids and the residue was then dried. The filter paper samples were placed in a desiccator with 35% HCl and were vacuumed for one day to remove the inorganic carbon. They were then neutralized by adding NaOH and were kept in a desiccator in vacuum. The subsamples for nitrogen were used without extra pretreatment after the lyophilization of biota and filter samples. Each prepared sample was put in a tin capsule and analyzed for carbon- and nitrogen-stable isotopes by using a stable isotope mass spectrometer (Isoprime100, Isoprime Ltd., UK) connected to an element analyzer (EA3000, Eurovector, Italy) in a tin capsule. The analyzed values are expressed in terms of delta notation and permil (‰), and the stable isotope ratio defines the difference between the reference material and analytical sample, according to the following equation:

$$\delta^{13}\text{C} \text{ (or } \delta^{15}\text{N})\text{‰} = \left[\left(\frac{R_{\text{sample}}}{R_{\text{standard}}} - 1 \right) \times 1000 \right]$$

$$R = {}^{13}\text{C}/{}^{12}\text{C} \text{ (or } {}^{15}\text{N}/{}^{14}\text{N}).$$

The reference material used Vienna Pee Dee Belemnite for carbon isotope analysis and atmospheric nitrogen for nitrogen isotope analysis. The substances CH-6 and N-1, certified by the International Atomic Energy Agency, were analyzed once every 12 samples for calibrating the sample values. Their standard deviation was within 0.2‰ for carbon and 0.3‰ for nitrogen.

2.4 Statistical analysis

The quantitative contribution of the expected food source of *E. pacifica* was calculated by using the Bayesian mixing model SIAR v. 4.2 (Stable Isotope Analysis in R; Parnell et al. 2010). The potential food sources entered into SIAR were size-fractionated plankton and zooplankton from different water layers at the same stations. Some size-fractionated zooplankton samples were analyzed by pooling because of their small amount. In such cases, the standard deviation was not able to be determined, and SIAR was calculated by inputting the standard deviation as zero. The overall stable isotope fractionation factors of the diet tissue generally average 0.4‰ for carbon and 3.4‰ for nitrogen (Post 2002). In the present study, the contribution of food source was calculated by applying $0.4 \pm 0.17\text{‰}$ for carbon and $2.3 \pm 0.28\text{‰}$ for nitrogen, as reported in an aquatic food web (McCutchan et al. 2003). The statistical

significance of the isotopic values for each sample was assessed using the Student's *t* test and one-way analysis of variance (ANOVA) in SPSS V12.0 software.

3 Results

3.1 Hydrographic physico-chemical profile data

In summer (August), the average surface water temperature at stations A03, A07, and D03 was 24 ± 0.9 °C and the water depth was 87, 72, and 92 m, respectively. However, that at D07, which had the shallowest water depth of 39 m, showed a relatively lower surface temperature (Fig. 2). The average temperature in the bottom layer of 4 stations was 12.4 ± 4.8 °C. In particular, stations A03 and D03, at depths > 80 m, showed a well-developed cold water mass

where the temperature was 10 °C lower than that at the surface. The average salinity was 31.9 ± 0.2 at the surface layer and 32.8 ± 0.2 at the bottom layer. Stratification occurred as a result of the density difference between the surface and bottom layers, which is consistent with the depth of the thermocline. The maximum fluorescence occurred at an average depth of 26.3 ± 9.9 m, which is consistent with the bottom of the thermocline. The dissolved oxygen showed a similar vertical distribution pattern to that of fluorescence except for that measured at D07.

3.2 Seasonal change in stable isotope ratios of *E. pacifica*

The nitrogen isotopes of *E. pacifica* were always approximately 2‰ higher in August than that in April in both 2012 and 2013 (Fig. 3). The carbon isotope value in April differed

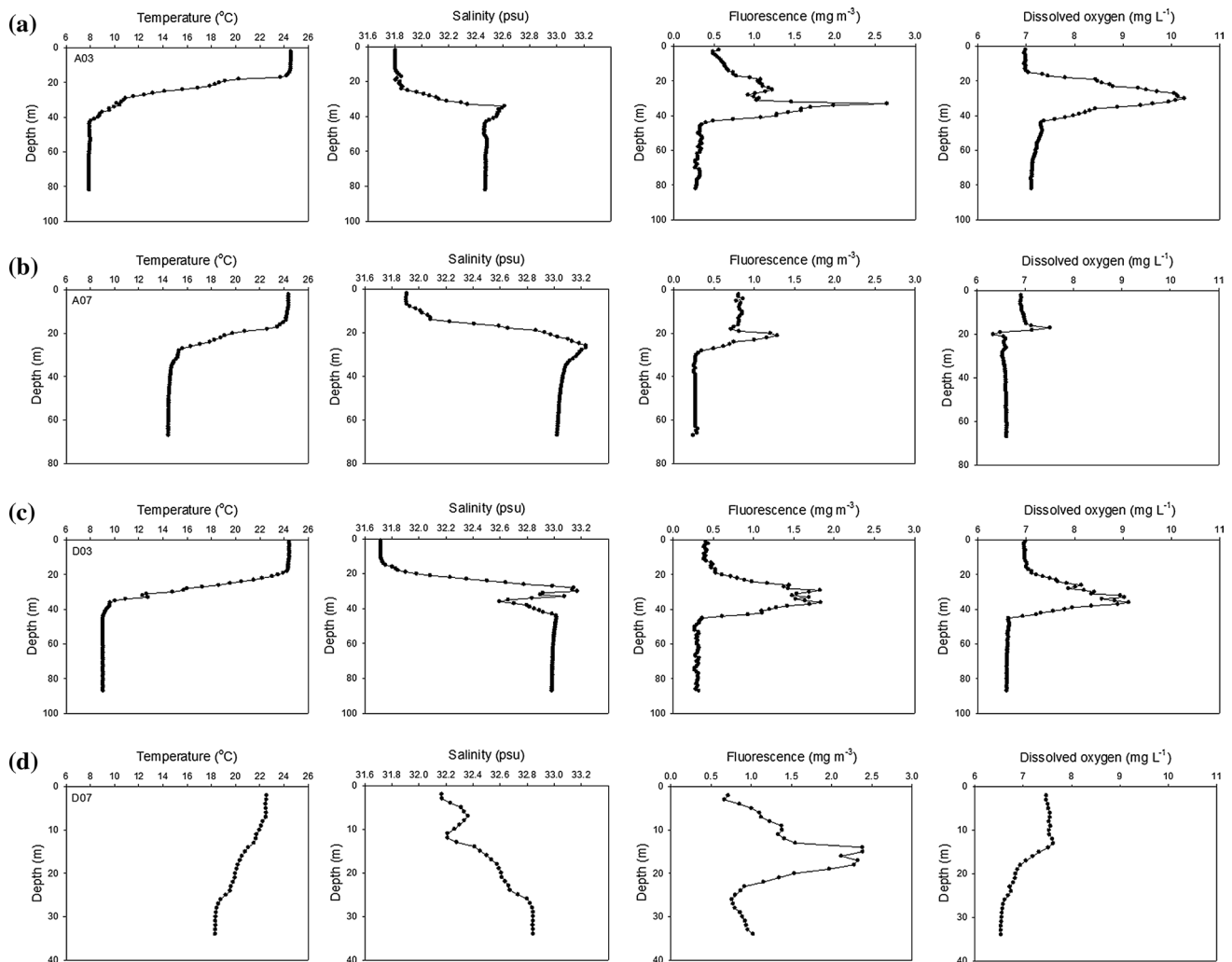


Fig. 2 Vertical distribution of temperature, salinity, and fluorescence concentration at sampling stations **a** A03, **b** A07, **c** D03, and **d** D07 in August 2014

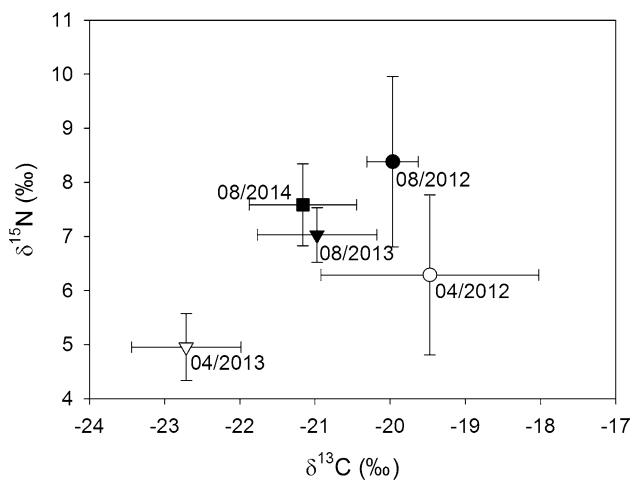


Fig. 3 Biplot of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values (mean \pm SD) of *E. pacifica* from 2012 to 2014 in the Yellow Sea (white: spring; black: summer; circle: 2012; inverted triangle: 2013; square: 2014)

significantly between years ($P < 0.05$). However, no significant difference was found in August, and the mean value ranged from -21 to -20‰ . The nitrogen isotope values in August in all three years were not significantly different each other ($P > 0.05$).

3.3 Stable isotope signatures of size-fractionated plankton

In A03 and D03 affected by cold water mass, the picoplankton of the SCM layer showed the lightest carbon-stable isotope values and picoplankton of the bottom layer showed the heaviest nitrogen isotope values (Fig. 4). The nano + microplankton showed significantly different carbon and nitrogen isotopic values ($P < 0.05$), respectively, in the SCM and bottom layer. However, the difference was lower than that of the picoplankton, and the nitrogen values in the bottom layer were slightly heavier than those in the SCM layer. In coastal waters A07 and D07, the picoplankton of the bottom layer showed the lowest carbon and highest nitrogen-stable isotope values among the size-fractionated plankton, and the nano + microplankton of the SCM layer showed the highest carbon and lowest nitrogen isotopic values. Overall, the size-fractionated plankton in the area affected by the cold water mass showed lighter carbon isotopic values than those in the coastal area.

3.4 Stable isotope signatures of large-sized zooplankton and *E. pacifica*

The carbon-stable isotopic values of zooplankton and *E. pacifica* were approximately 2‰ lighter in the cold water mass (A03 and D03) than in coastal waters (A07 and D07);

a similar result was also shown in the large-sized plankton (Fig. 4). In the coastal waters, the carbon-stable isotopic values of *E. pacifica* in A07 and D07 were not significantly different ($P > 0.05$), although the nitrogen values were differed significantly ($P < 0.05$). In addition, the nitrogen-stable isotopic values of zooplankton and *E. pacifica* were similar in A07, although the range was relatively broad in A03. The carbon and nitrogen-stable isotopic values of *E. pacifica* in the cold water mass showed no significant difference ($P > 0.05$). On the other hand, *E. pacifica* in the cold water mass showed the lowest nitrogen-stable isotope values among the collected zooplankton. The body length of *E. pacifica* was 14.0 ± 3.4 mm ($n = 38$), and there was no significant difference ($P > 0.05$) between the stations.

3.5 Diet sources of *E. pacifica*

Among the 7 components of size-fractionated plankton collected as potential food sources of *E. pacifica*, the zooplankton of 2.0–5.0 mm had a mean contribution of 42%, and that of 0.5–1.0 mm accounted for 25% of the contribution at station A03, which had cold water mass below the thermocline in summer (Fig. 5). At station D03, zooplankton > 5.0 mm had the highest mean contribution of 22%, followed by zooplankton of 2.0–5.0 mm, at 17%. At coastal station A07, the nano + microplankton of the SCM layer had the highest contribution, at 15%. At station D07, zooplankton of 0.2–0.5 mm had the highest contribution, at 20%. The major food sources of *E. pacifica* in the Yellow Sea at stations A03 and D03, the cold water mass areas, were relatively large zooplankton of 2.0–5.0 mm and > 5.0 mm, respectively, whereas at the coastal area, they fed on all sizes of zooplankton and nano + microplankton. Picoplankton played a minor role as a diet source for *E. pacifica* at all stations.

4 Discussion

In this study, the major food source of *E. pacifica* in the YSCWM in summer was large-sized zooplankton of 2.0–5.0 mm, which was revealed by SIAR using stable isotope values. However, *E. pacifica* was omnivorous rather than carnivorous in the coastal waters, where the influence of cold water is low. According to a study by Kang et al. (unpublished data), the most abundant zooplankton species in same area in 2012 was copepod *Paracalanus parvus*. In addition, copepods *Calanus sinicus*, *Oithona atlantica*, *O. similis*, *Ditrichocorycaeus affinis* (*Corycaeus affinis*), *Acartia hongii*, *A. omorii*, *A. hudsonica*, and Sagittidae *Aidanosagitta crassa* (*Sagitta crassa*) were dominant species. The sizes of the dominant species were *P. parvus*, 0.6–1.3 mm; *C. sinicus*, 2.07–3.6 mm; *O. atlantica*, 1.11–1.43 mm; *O. similis*, 0.6–1.2 mm; *D. affinis*,

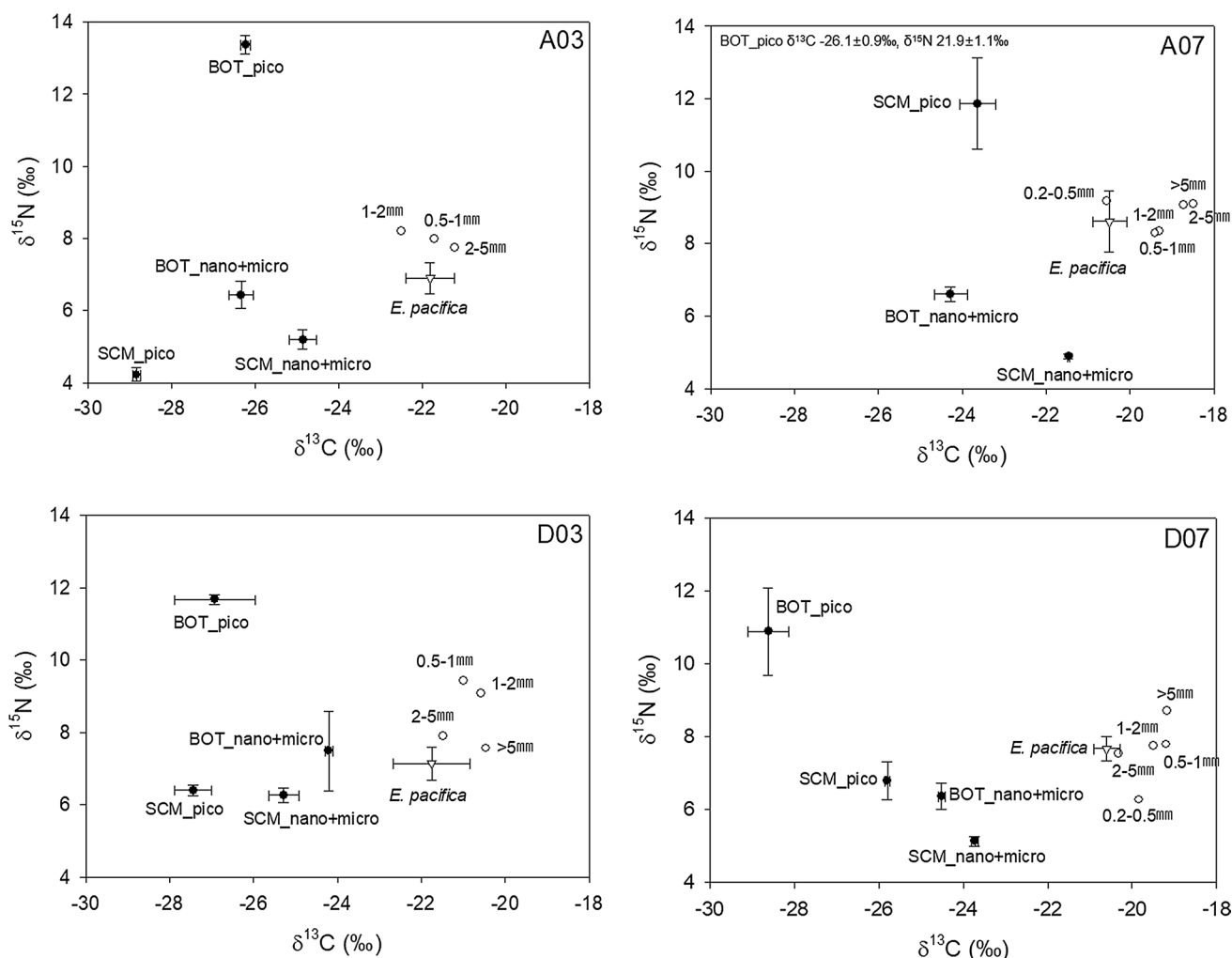


Fig. 4 Biplots of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values (mean \pm SD) of size-fractionated plankton and *E. pacifica* at sampling stations in August 2014 (filled diamond: particulate organic matter; open circle: zooplankton;

inverted triangle: *E. pacifica*. BOT bottom layer, SCM surface chlorophyll maximum layer)

0.62–0.87 mm; *Acartia* sp., 0.659–2.1 mm; and Sagittidae, 10–90 mm (WoRMS Editorial Board 2017). *P. parvus* showed high density at mainly 0–20-m depths (Kang et al. unpublished data). On the contrary, *C. sinicus* was predominant in terms of biomass and were distributed in a relatively wide range of water depth (Kang et al. unpublished data). Their approximately 2–3-mm size classifies them as 2–5-mm zooplankton, the stable isotopes of which were analyzed in this study. Therefore, the major food source of *E. pacifica* in cold water mass is likely *C. sinicus*.

The diet of Euphausiid can be divided into three types: phytoplankton, zooplankton, and organic detritus (Mauchline and Fisher 1969). *Euphausia* species including *E. pacifica* are considered to be omnivorous filter feeders (Ohman 1984; Suh and Choi 1998). They appeared to be a selective omnivore in feeding experiments conducted with diatoms and copepods as dietary sources, and small

components such as bacteria in the food web could not be fed on directly or effectively (Ohman 1984). These species feed on marine snow formed directly by gelatinous zooplankton, such as coagulated small particles of phytoplankton and granular excrement or mucilage prey structures (Alldredge and Silver 1988; Dilling et al. 1998). The grazing rates of *E. pacifica* on marine snow do not depend on food quality (Dilling et al. 1998). In general, stable carbon isotopes in living organisms can reflect the origin of their food source in food web studies. The picoplankton sample, which had the smallest particle size in this study, showed a difference of more than 4‰ in the $\delta^{13}\text{C}$ value from *E. pacifica* in the SCM and bottom layers. *E. pacifica* did not appear to directly feed on picoplankton, and it was presumed that its role as a food source was minimal.

In a study on the grazing and metabolism of *E. pacifica* collected in the Yellow Sea, a culture feeding experiment

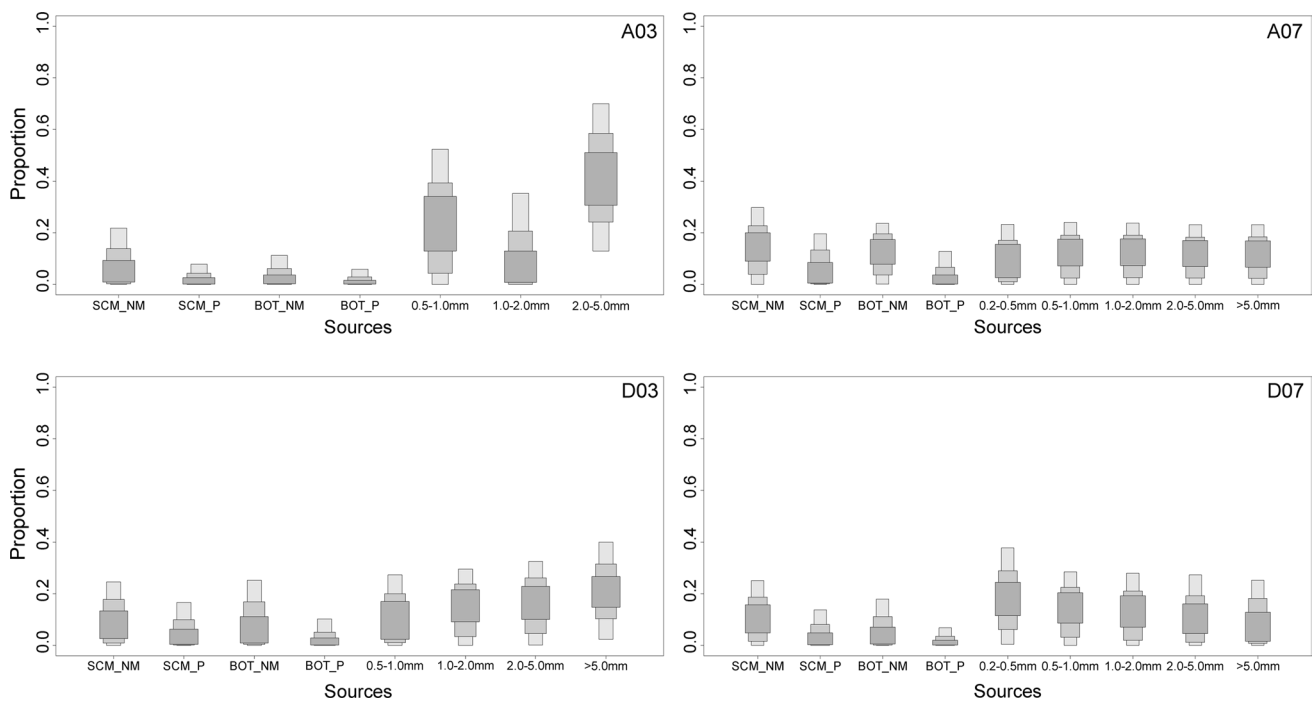


Fig. 5 SIAR boxplots showing the proportional contribution of potential *E. pacifica* prey at sampling stations in August 2014. The gray scale (from light to dark) indicates 95, 75, and 50% confidence intervals

using a Coulter counter showed that *E. pacifica* fed mostly on microzooplankton containing ciliates in August and September (Tao et al. 2015). In contrast, the oxygen consumption rate peaked in April and was four times higher than that in September, which is related to high breeding and feeding rates. Moreover, the O-to-N ratio of *E. pacifica* was highest in April, coinciding with spawning and the highest prey abundance, resulting in active metabolism (Sun et al. 2011; Tao et al. 2015). During September and December, the O-to-N ratio of *E. pacifica* was 90% lower than April with low phytoplankton concentrations (Tao et al. 2015). It generally indicates a stressed condition, such as starvation (Mayzaud and Conover 1988) and consequence of reduced respiration rates (Tao et al. 2015). However, most energy sources from September to December were proteins derived from a carnivorous diet containing microzooplankton. In the YSCWM in summer and autumn, the energy consumption in *E. pacifica* remained low. The O-to-N ratio of *E. pacifica* collected in April, September, and December showed a negative correlation with surface water temperature, and there was no significant correlation with chl-*a* concentration (Tao et al. 2015). Therefore, water temperature is an important factor in *E. pacifica* metabolism. In the present study, we calculated the contribution of predicted food sources to *E. pacifica* by using carbon- and nitrogen-stable isotopes. Zooplankton of the 2.0–5.0-mm size class at station A03, where the cold bottom layer was the most developed, contributed an average of 42.3% to its diet. Therefore, *E. pacifica* mainly fed

on large-sized zooplankton. In previous studies, the potential food sources of *E. pacifica* ranged from small organic matter to microplankton (Tao et al. 2015). However, in the present study, we considered larger plankton as a possible food source. On the basis of dual stable isotope analysis, we determined that the contributions of mesoplankton and microplankton are considerable.

The optimum temperature of *E. pacifica* in the marginal sea of the western North Pacific was 11.4 °C, and they cannot survive more than one day at temperatures above 20 °C (Iguchi and Ikeda 1995). Higher summer temperatures may alter the balance of parameters that have inter-dependence between molting rates and chl-*a*, and between molting increment and temperature in metabolic responses. It may inhibit the growth of *E. pacifica* (Pinchuk and Hopcroft 2007). From summer to autumn, the surface water temperature in the Yellow Sea is above 20 °C. *E. pacifica* inhabiting the YSCWM exhibited decline in grazing, metabolism, and reproduction rates (Tao et al. 2015). The YSCWM and the water layer below the thermocline may provide conditions necessary for *E. pacifica* growth. In the present study, an acoustic scattering layer of *E. pacifica* identified by an echo sounder existed at depths close to the seabed in the cold waters but at 10–20 m above the seabed in coastal waters. It is believed that the activity of *E. pacifica* is significantly reduced in the bottom cold water mass, where the thermocline is strongly developed and the water temperature is less than 10 °C. The stable isotope values of *E. pacifica* for the

3-year period showed that the average carbon isotope values in summer were less variable from -21 to -20‰ compared with spring. The relatively small range in carbon isotope of *E. pacifica* might be attributed to the limited habitat available in the cold water mass in summer, which provides very limited diet sources such as zooplankton. However, *E. pacifica* in spring appears to have access to a variety of food sources that promote active feeding and reproductive activities, which are supported by the comparatively wide range in carbon isotope values in spring. The habitat of *E. pacifica* is restricted to the cold water mass in summer, which provides very limited diet sources such as zooplankton. In addition, the heavier nitrogen isotope signatures in summer compared with those in spring suggest that *E. pacifica* feeds more on carnivorous diet sources. Therefore, considering the current dual-isotope results, *E. pacifica* may feed mainly on zooplankton larger than 2.0 mm such as copepods *C. sinicus* even though they might be physiologically less active in the YSCWM in summer.

The stomach content analysis revealed that the feeding behavior of *E. pacifica* at the northeastern coast of Japan varies according to the surrounding food conditions (Nakagawa et al. 2001; Taki et al. 2002). In addition, the prey of this species differs seasonally from herbivorous feeding of diatoms in the spring to more carnivorous feeding of mainly copepods and detritus in the summer, fall, and winter (Endo 1981; Taki et al. 2002). The analysis of stomach contents and stable isotopes for adult *Euphausia vallentini* in the Southern Ocean revealed differing results in the two methods (Gurney 2000). The stomach contents showed high phytoplankton and low metazoan compositions, whereas stable isotope analysis showed a high omnivorous trend. Nakagawa et al. (2001, 2002) highlighted the importance of feeding on heterotrophic prey such as copepods in terms of the carbon weight calculation of heterotrophic prey and pigment content of autotrophic prey among *E. pacifica* prey. Sogawa et al. (2017) reported that no seasonal variation was noted in the nitrogen-stable isotopic values of *E. pacifica* with a standard deviation of 0.2‰ in the northeastern coast of Japan. Therefore, even though phytoplankton with frustles such as diatoms remain intact in the stomach contents, the effect of the phytoplankton spring bloom of Oyashio water on the trophic level fluctuation of *E. pacifica* would be small. In this study, *E. pacifica* in the Yellow Sea, with a seasonally varying water environment, showed heavier nitrogen isotopic values than those measured in spring, and the carbon-stable isotopic values were narrowed to a particular range. Therefore, it is suggested that food source of *E. pacifica* was limited in the YSCWM where *E. pacifica* escaped from warm surface water.

On the contrary, a study conducted in the southern part of the Yellow Sea in 2009 revealed that the nitrogen-stable isotope ratio of nitrate in seawater was $6.7 \pm 0.8\text{‰}$ ($n = 61$)

in February and $6.9 \pm 2.6\text{‰}$ ($n = 23$) in July (Umezawa et al. 2014). Although a slight change was noted in the mean value between the two periods, it increased significantly at some stations. In this study, an increase in the nitrogen isotopic values of *E. pacifica* may be attributed to changes in primary producers and nitrogen sources. Therefore, it is necessary to understand the seasonal changes in isotopic values between primary producers and krill for more accurate interpretation.

5 Conclusion

We studied the feeding characteristics of *E. pacifica* in the YSCWM by using carbon- and nitrogen-stable isotopes. The stable isotope signature of *E. pacifica* inhabiting the cold water mass was confined to a specific range, which means that the food sources of *E. pacifica* are limited. Previous studies have reported that the physiological activity of *E. pacifica* in the cold water mass is reduced in summer. However, stable isotope analysis in the present study revealed that their grazing activity on relatively large zooplankton is still present in summer. Therefore, the YSCWM, which is strongly developed in summer, is a very important habitat for the survival of *E. pacifica* and provides shelter for avoiding the high-temperature seawater surface as well as a zooplankton diet source.

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