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Spatial distribution of ciliates, copepod nauplii and eggs, *Engraulis japonicus* post-larvae and microzooplankton herbivorous activity in the Yellow Sea, China

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ABSTRACT: The abundance of anchovy *Engraulis japonicus* larvae, >20 µm ciliates, copepod eggs and nauplii, and microzooplankton herbivorous activity were studied in the Yellow Sea in June 2000. Anchovy juveniles and larvae were found in only 6 of the 19 stations sampled. The ciliate communities were dominated by 2 species: *Laboea strobila* and *Strombidium compressum*. In the surface waters, the abundance of *L. strobila* ranged between 0 and 560 ind. l^{-1}. *S. compressum* only appeared at Stns 15 to 18 (20 to 3300 ind. l^{-1}). *L. strobila* was found mainly in the top 20 m. The abundance of *L. strobila* was less than 50 ind. l^{-1} in waters deeper than 25 m. *S. compressum* showed subsurface abundance peaks at the salinity abnormality. Tintinnids occurred occasionally with abundance lower than 100 ind. l^{-1}. The total ciliate abundance fell in the range of 40 to 3420 ind. l^{-1}. The ciliate biomass in the surface water and the water column ranged between 0.15 and 6.76 µg C l^{-1} and 0.4 and 134.4 mg C m^{-2}, respectively. In the surface waters, the abundance of copepod eggs and nauplii ranged from 0.3 to 3.1 and 1.1 to 15.6 ind. l^{-1}, respectively. The average abundance of copepod eggs and nauplii in 4 depth (0, 5, 10 and 20 m) fell in the range of 0.2 to 2.8 and 1.0 to 29.4 ind. l^{-1}, respectively. As a food item of the *E. japonicus* post-larvae, the abundance of copepod nauplii and eggs appeared to be low. The abundance peaks of ciliate and *E. japonicus* post-larvae coincided. Although not found in the gut of *E. japonicus* post-larvae, aloricate ciliates might be ingested by first-feeding anchovy larvae, preventing initial starvation and prolonging the time to irreversible starvation. On the basis of dilution experiments with positive microzooplankton grazing rates, microzooplankton grazed at rates of 0 to 0.61 d^{-1}. Grazing pressure of microzooplankton on chlorophyll *a* standing stock (P_i) and potential chlorophyll *a* primary production (P_p) were 17 to 46% and 35 to 109% d^{-1}, respectively.

KEY WORDS: Ciliate · *Engraulis japonicus* · Microzooplankton · Dilution incubation

INTRODUCTION

Planktonic ciliates are a morphologically diversified group and are usually divided into loricate (tintinnid) and aloricate (naked) forms. In most marine ecosystems, they dominate the microzooplankton (Beers et al. 1980, Uye et al. 1996), which is the trophic link between the microbial food web and the metazoans (Stoecker & Capuzzo 1990, Gifford 1991). Ciliates are also food items of fish larvae. Tintinnids have been found in the gut of field-collected fish larvae (Jenkins 1987, Govoni & Chester 1990). There is no evidence that fish larvae feed upon naked ciliates in the field because no hard parts of naked ciliates can be identified in the gut of field-

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collected fish larvae. However, naked ciliates were ingested in experimentally reared fish larvae as shown by different techniques (Ohman et al. 1991, Lessard et al. 1996, Nagano et al. 2000). Ohman et al. (1991) detected naked ciliate components in the gut of larval anchovy by immunochemical methods. Using ciliates labeled with 2,4-diamadino-6-phenylindole (DAPI) and fluorescent microspheres, respectively, Lessard et al. (1996) and Nagano et al. (2000) found a trace of ciliates in the guts of fish larvae. In the field, naked ciliates may be more important food for fish larvae than tintinnids because the abundance of naked ciliates is much greater than that of tintinnids (Pierce & Turner 1992).

China-GLOBEC II (Global Ocean Ecosystem Dynamics) has chosen the Eastern China and Yellow Seas to be the study area. Among the 12 projects of China-GLOBEC II were the early recruitment mechanisms of key species (e.g. *Engraulis japonicus*, an anchovy) and process studies on microbial and microplankton production (Tang 2000). From 1997 to 1999, the catch of *E. japonicus* was more than 1 million tons yr^{-1}, which constituted 20 to 25% of the annual fishery catch in the whole country. The anchovy spawn in the Yellow Sea in May and June (Zhu & Iversen 1990). It has long been proposed that the strength of a year-class of fish stock is most probably determined by the mortality at the earliest larval feeding stage (the so-called 'critical period' stated by May 1974), and the mortality is dependent upon the food density when they first begin to feed (Hjort 1914). Copepod nauplii were reported to be numerically the most important food items of *E. japonicus* larvae by examining the larval gut (Hirakawa et al. 1997). In order to investigate the early recruitment mechanisms of this species, a cruise was carried out during June 13 to 28, 2000. The abundance of copepod nauplii and ciliates, which can be potential food items of anchovy larvae, were studied in this cruise. Microzoopankton herbivorous activities were also evaluated using the dilution incubation technique.

MATERIALS AND METHODS

Stations. Measurements were made on board RV 'Beidou' during June 13 to 28, 2000. The study stations are shown in Fig. 1. Nineteen grid stations (Stns 1 to 19) with depths between 22 (Stn 4) and 79 m (Stn 8) were investigated from June 13 to 18. At each of the grid stations, an SBE19 Seacat Profiler (Model SBE19-03) was cast at first and the profiles of temperature and salinity were displayed using the Seasoft® (version 4.233) software (Sea-Bird Electronics) immediately onboard the ship. Water samples for the chemical and biological studies were collected with 2.5 l Niskin bottles on a Rossette conductivity, temperature and depth profiler (CTD). Sample depths (generally 4 to 6 depths) were chosen depending on the location of the thermocline. Nutrients were analyzed by colorimetry method (Zhang et al. 1997).

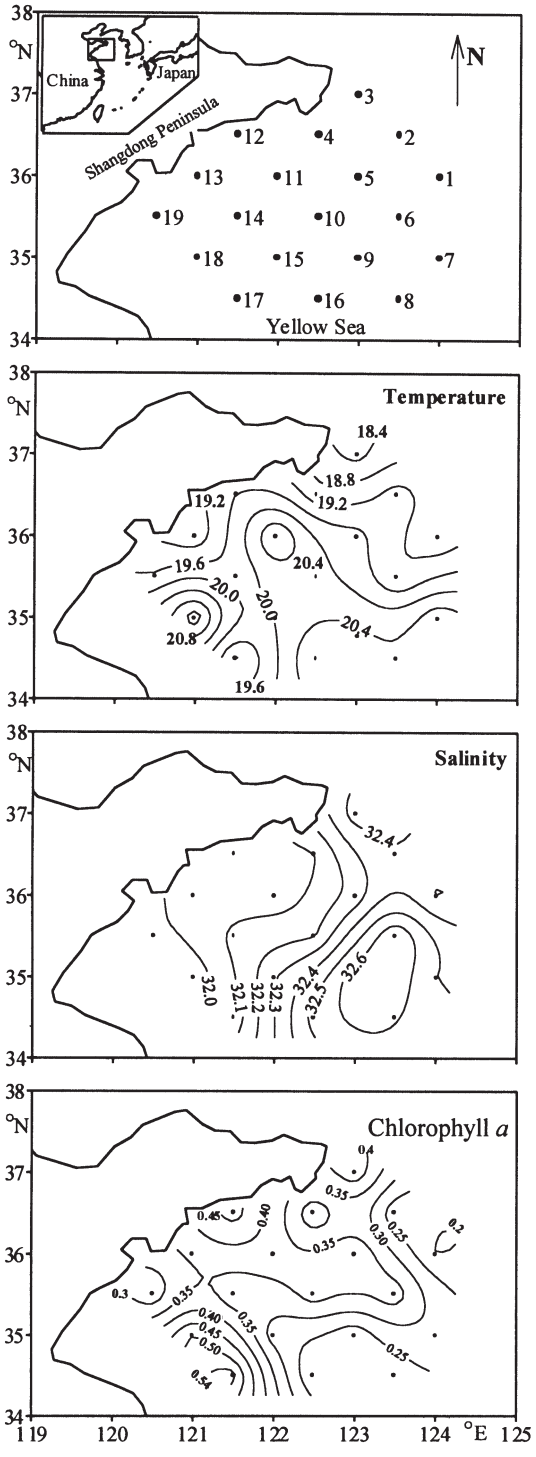

Fig. 1. Map of the study area and the distributions of surface temperature (°C), salinity and chlorophyll *a* concentration (μg l^{-1})

One liter of surface water was sampled to measure the chlorophyll *a* concentration. Chlorophyll *a* concentration was determined as follows. Water samples were filtered through GF/F glass fiber filters. The filters were extracted with 90% acetone at –20°C in darkness for 24 h. The concentrations were determined using a Turner Designs (Model II) fluorometer that was calibrated with pure chlorophyll *a* from Sigma (Strickland & Parsons 1972).

Anchovy eggs and larvae. At each station, abundances of anchovy larvae and eggs were investigated by horizontally towing a net (0.8 m mouth diameter with 500 µm mesh, 2.7 m in length) on the sea surface at a speed of 3.2 knots. The unit of anchovy larvae abundance, ind. net^{-1}, means the catch in a 10 min towing (towing distance was ca 988 m). The total body lengths were measured, and gut contents of anchovy larvae and juveniles were examined using a stereomicroscope.

Ciliates. A 1 l sample of water from each depth was poured into a 1 l plastic bottle and fixed in 1% acid Lugol's iodine solution (Longhurst et al. 1990, Harris et al. 2000). The bottles were stored cool and in darkness until analyzed in the laboratory within 2 mo. In the laboratory, the fixed water samples were settled for at least 24 h in the plastic bottles. The upper water was siphoned out and 100 ml was left. Then 25 ml (50 or 100 ml if necessary) of the samples was settled in sedimentation chambers and counted using a Zeiss microscope at 150× magnification. In each sample, ciliates (including aloricate ciliates and tintinnids) with a minimum preserved dimension of >20 µm were counted.

For taxonomic studies, Lugol's solution-fixed samples were replaced by Bouin's fluid for protargol staining (Lynn 1992). The protargol impregnation technique (Wilbert 1975) was partially used to reveal the infraciliature of ciliates, regarded as the key feature for identification. The classification of ciliates was based on common literature (Kofoid & Campbell 1929, Maeda & Carey 1985, Small & Lynn 1985, Lynn et al. 1991, 1988, Montagnes & Lynn 1991, Carey 1992). The dimensions of the ciliates were measured and the cell volume of each species was estimated using appropriate geometric shapes (cone, ball, cylinder, cuneiform and their combinations). The carbon:volume ratio used to calculate biomass was 0.19 pg C $µm^{-3}$ (Putt & Stoecker 1989). The plasma of tintinnids was assumed to occupy 30% of the lorica volume (Gilron & Lynn 1989). Water column biomass was calculated as the integral of the biomass from bottom to surface.

Copepod eggs and nauplii. A large-volume (60 l) water sampler was used to collect water from different depths (0, 5, 10, 20 m and so on) at every grid station. The water samples were poured into a net (38 µm mesh size). Copepod eggs and nauplii in the cod end were preserved with a 5% buffered formalin-seawater solution. Abundances of eggs and nauplii were counted under dissecting microscope later in the laboratory.

Dilution incubations. After the grid investigation, anchovy eggs, of which 99% were dead, were found only at Stn 10. The peak of anchovy larvae abundance was found at Stn 17. These 2 positions were taken as anchor stations. After the investigation at anchor Stn 10, the larvae peak, which appeared at Stn 17 during the grid investigation, moved to the position of Stn 18. As a result, Stns 10 and 18 were the anchor stations.

A total of 6 dilution incubation experiments were carried out (Table 1) at the 2 anchor stations. The experimental protocols by Landry & Hassett (1982) and Burkill et al. (1990) were followed. The experimental items including 25 l polycarbonate carboy, 1.5 l

Table 1. Results of the dilution incubation experiments in June 2000. Chl *a*: chlorophyll *a*; *g*: microzooplankton grazing rate; *k*: potential phytoplankton growth rate; P_i: microzooplankton grazing pressure on chl *a* standing stock; P_p: primary production. The unit of nutrient concentration is µmol l^{-1}

Station/ depth (m)	Time (hour/date)	NH_4^+	NO_2^-	NO_3^-	PO_4^{3-}	SiO_3^{3-}	Chl *a* (µg l^{-1})	*k* (d^{-1})	*g* (d^{-1})	r^2	P_i (%)	P_p (%)
Stn 10/ 10	12:00/19 to 12:00/20	1	0.05	0.38	0.04	0.80	0.22	0.52 to 0.7	0	–	–	–
Stn10/ 10	09:00/21 to 09:00/22	0.54	0.01	0.53	0.01	0.38	0.25	0.74 (0.080)	0.41 (0.116)	0.67	34	64
Stn10/ 0	10:30/21 to 10:30/22	0.71	0.01	0.84	0.00	0.47	0.18	0.14 to 0.52	0	–	–	–
Stn18/ 0	08:00/23 to 08:00/24	0.96	0.06	0.83	0.05	1.09	0.78	0.64 (0.042)	0.61 (0.029)	0.97	46	97
Stn18/ 10	11:00/23 to 11:00/24	0.74	0.08	0.24	0.06	4.33	0.92	0.20 (0.056)	0.22 (0.082)	0.56	20	109
Stn18/ 0	09:00/25 to 09:00/26	0.50	0.03	0.57	0.08	0.28	0.55	0.69 (0.045)	0.19 (0.066)	0.58	17	35

polycarbonate bottles, glass filter bottles, etc. were soaked with 10% HCl and rinsed with filtered seawater (FSW) before use. At each of the above stations, the 0 and 10 m waters were chosen as experimental depths. To begin incubation, 24 l seawater from the goal depth was collected using Niskin bottles and transferred to a polycarbonate carboy. Part of this water was filtered through GF/F filters. The FSW was assumed to be free of predator and prey. The FSW was added to make concentrations of 100, 75, 50 and 25% (dilution factor) of ambient seawater in polycarbonate bottles. A mesh (38 µm) was used to eliminate the macrozooplankton. From each concentration, 500 ml water was sampled for the determination of initial chlorophyll *a* concentration. The rest of the water was poured into two 1.5 l polycarbonate bottles. Caution was taken to avoid air bubbles in the bottles. We did not add nutrients to the dilution bottles.

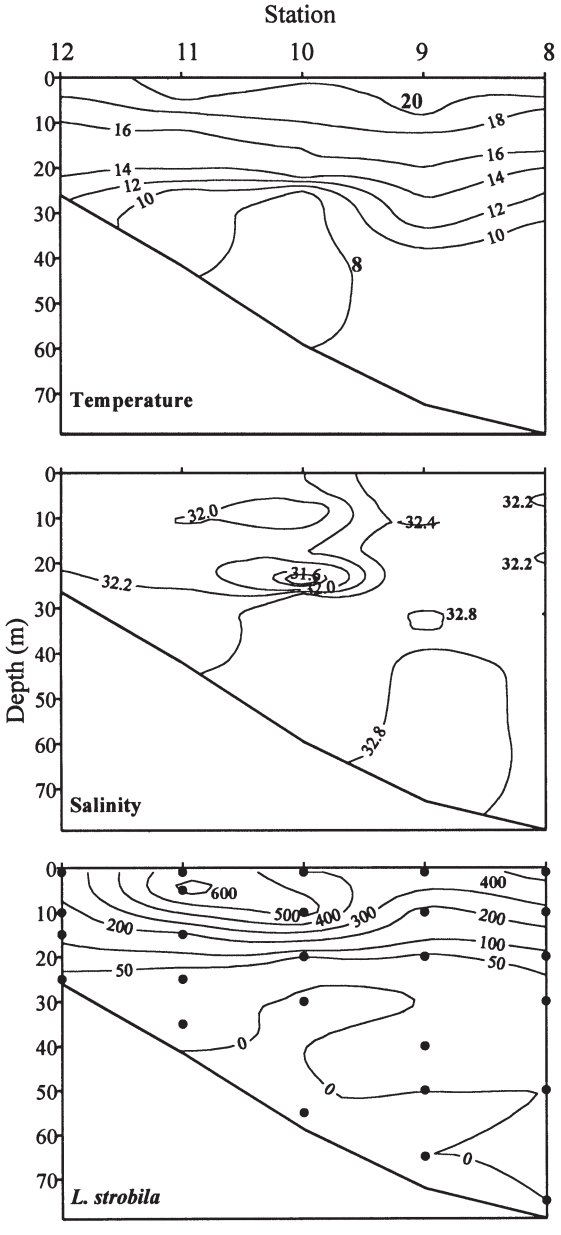

Fig. 2. Vertical distributions of temperature (°C), salinity and *Laboea strobila* abundance (ind. l^{-1}) along the transect through Stns 8 to 12

The bottles were incubated in 2 plastic boxes (semi-transparent with the openings upward) at 10 m depth suspended from the ship or in a temperature-control container (not transparent) under natural light on board the ship. In the latter case, the surface water was pumped and flowed through the container to maintain the *in situ* temperature. Further samples for chlorophyll *a* concentration were taken after 24 h incubation. Chlorophyll *a* concentrations were determined as described above.

According to Landry & Hassett (1982), microzooplankton grazing rate (g, d^{-1}) and the potential phytoplankton growth rate (k, d^{-1}) can be expressed as follows:

$$1/t \ln(P_t/P_0) = k - c \times g$$

where P_t is the chlorophyll *a* concentration at time t; P_0 is the initial chlorophyll *a* concentration; and c is the dilution factor. Values of k and g were determined from linear regression of the apparent chlorophyll *a* growth rates against the dilution factors. Microzooplankton grazing pressure on chlorophyll *a* standing stock (P_i) and primary production (P_p) were calculated according to Verity et al. (1993):

$$P_i = 1 - e^{-gt} \times 100\%$$

$$P_p = [e^{kt} - e^{(k-g)t}]/(e^{kt} - 1) \times 100\%$$

RESULTS

Hydrographic conditions and chlorophyll *a*

The surface temperature was between 18.3 and 21.5°C. Water temperature in the northern part of the study area was lower than that in the south. But surface temperature at Stn 17 was comparatively low in the southwestern part of the study area (Fig. 1). The vertical profile of temperature showed strong stratification. All of the stations showed obvious thermoclines at different depths. For example, temperature drops from the surface to about 30 m along the transect through Stns 8 to 12. Waters below 30 m were lower than 9°C (Fig. 2). In the surface waters, salinity (31.95 to 32.70‰) was lower in the near shore area (Fig. 1). Parallel to the surface salinity contour in Fig. 1, there

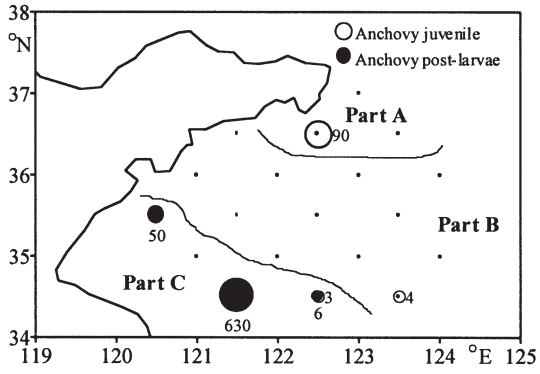

Fig. 3. Chart showing the division of the study area. The numbers in the figure indicate the abundance (ind. net^{-1}) of the anchovy juveniles and post-larvae. Numbers to the right of the circles: abundance of juveniles. Numbers below the circle: abundance of post-larvae

was a zone with low salinity at 26 m depth (Fig. 2). Chlorophyll *a* concentrations in the surface waters ranged between 0.20 and 0.56 µg l^{-1} (Fig. 1). The maximum concentration was at Stn 17.

Anchovy eggs and larvae, gut content of larvae

Anchovy eggs, of which 99% were dead, were found only at Stn 10. The distribution of anchovy larvae is shown in Fig. 3. At Stn 4, 90 ind. net^{-1} anchovy juveniles (20 to 25 mm in total body length) were found. There were only 4 and 3 ind. net^{-1} of juveniles at Stns 8 and 16, respectively. There were 630 post-larvae (5 to 10 mm in total body length, 2.5 to 8.9 mm in notochord length) and 12 pre-larvae net^{-1} at Stn 17. Fifty post-larvae net^{-1} were found at Stn 19. The abundance of anchovy larvae at other stations was even lower.

Food incidence (percentage of larvae containing at least one food particle for a particular sample) was 16.2%. Gut contents of the post-larvae were dinoflagellates, copepods of different development stages (eggs, nauplii), etc. Numerically, copepod eggs and nauplii accounted for 42 and 30.4%, respectively, of the food particles found in the guts of anchovy larvae.

Ciliate composition and distribution

A total of 17 species were identified, as listed in Table 2. There were 2 dominant ciliates: *Laboea strobila* Lohmann, 1908 and *Strombidium compressum* Kahl, 1932. In the surface waters, the abundance of *L. strobila* ranged between 0 (Stns 3, 4 and 17) and 560 ind. l^{-1} (Stn 11). Stns 7, 8, 10 and 11 had abundances more than 400 ind. l^{-1} (Fig. 4). *S. compressum*

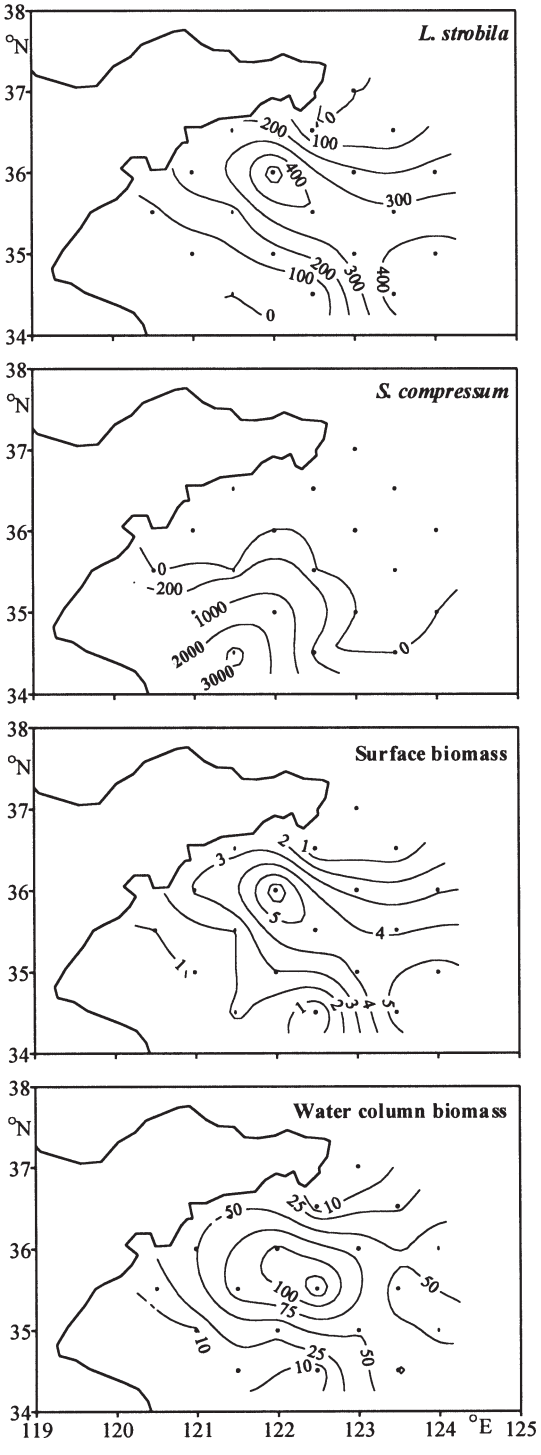

Fig. 4. Spatial distributions of the abundance (ind. l^{-1}) of *Laboea strobila*, abundance (ind. l^{-1}) of *Strombidium compressum*, ciliate surface biomass (µg C l^{-1}) and ciliate water column biomass (mg C m^{-2})

only appeared in Stns 15 to 18 (1632, 20, 3300 and 480 ind. l^{-1}, respectively).

Vertically, *Laboea strobila* occurred mainly above 20 m. The maximum abundance was 640 ind. l^{-1} at the

Table 2. List of ciliate taxa found in the Yellow Sea

Taxon	Size (length × maximum width) (μm)
Oligotrichida	
Oligotrichina	
Laboea strobila	100–120 × 45–50
Strombidium compressum	25–30 × 20–25
Strombidium reticulatum	30–50 × 20–35
Strombidium sp.	18–20 × 15–20
Strombidinopsis sphaira	30 × 25
Tintinnina	
Tintinnopsis beroidea	50–60 × 25–30
Codonelopsis lusitanica	85 × 40
Stenosemella pacifica	40 × 35
Stenosemella steini	80–90 × 70–80
Parafavella ventricola	310–420 × 65–80
Gymnostomatea	
Mesodinium pupula	25–30 × 18–20
Mesodinium pulex	18–25 × 15–18
Mesodinium rubrum	25–30 × 20–25
Didinium gargantua	85–90 × 55–60
Cyclotrichium sphaericum	75–90 × 70–80
Cyclotrichium sp.	20 × 10
Gen. sp. (unknown)	25 × 15
Total number of species	17

depth of 5 m in Stn 11. In waters deeper than 25 m, the abundance of *L. strobila* was less than 50 ind. l^{-1} (Fig. 2). The abundance (138 ind. l^{-1}) of *L. strobila* was found to be lower in the surface waters in Stn 14, while it was high in the subsurface waters (432 and 312 ind. l^{-1} at 5 and 15 m depths, respectively). In the case of *Strombidium compressum* (Fig. 5), there were peaks in the surface and subsurface waters with the exception of Stn 16, where there was no surface abundance peak.

The abundance of other aloricate ciliates was lower than 80 ind. l^{-1}. Tintinnids occurred occasionally. The maximum tintinnid abundance was 100 ind. l^{-1} in the surface waters of Stns 3 and 4. At other stations and depths, tintinnid abundance appeared to be lower than 20 ind. l^{-1}. The total ciliate abundance fell in the range of 40 to 3420 ind. l^{-1}.

Ciliate biomass in surface water and the water column ranged from 0.15 to 6.76 μg C l^{-1} and 0.4 to 134.4 mg C m^{-2}, respectively (Fig. 4). Although the abundance of *Strombidium compressum* was much higher than that of *Laboea strobila*, the biomass of *L. strobila* was dominant both at the surface and in the column.

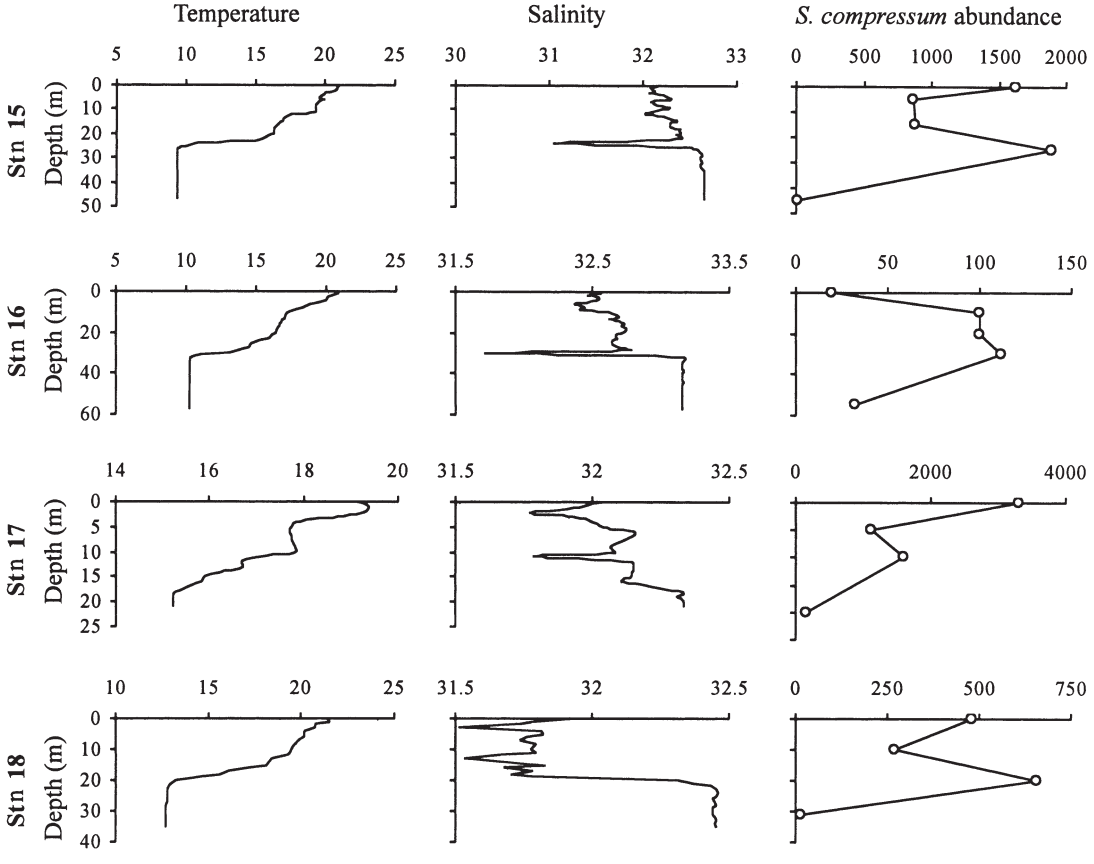

Fig. 5. Vertical profiles of temperature (°C), salinity and *Strombidium compressum* abundance (ind. l^{-1}) at Stns 15, 16, 17 and 18

Copepod egg and nauplii distribution

The distributions of copepod eggs and nauplii are shown in Fig. 6. Eggs and nauplii in surface water were considered to be the food immediately available to the fish larvae, while those at depth were the potential food with vertical migration. In the surface waters, the abundance of copepod eggs ranged between 0.3 and 3.1 ind. l^{-1} with more than 2.0 ind. l^{-1} at Stns 1, 4, 15 and 17. The average abundance among the upper 4 depths fell in the range of 0.2 to 2.8 ind. l^{-1}; Stns 4, 5 and 17 had values more than 2.5 ind. l^{-1}.

The copepod nauplii abundance in the surface waters fell in the range of 1.0 to 29.4 ind. l^{-1} with more than 10 ind. l^{-1} at Stns 10, 15 and 18. The average abundance among the upper 4 depths was 1.1 to 15.6 ind. l^{-1}; Stns 6, 15 and 18 had abundances more than 10 ind. l^{-1}.

Microzooplankton grazing pressure

The results of the 6 dilution incubations are shown in Fig. 7 and Table 1. Phytoplankton grew at rates 0.2 to 0.74 d^{-1}. Microzooplankton grazed at rates 0 to 0.61 d^{-1}. Two of the experiments showed no microzooplankton grazing. For the experiments with positive microzooplankton grazing, P_i and P_p were 17 to 46% and 35 to 109% d^{-1}, respectively.

DISCUSSION

Ciliates

The final concentration of acid Lugol's solution for the fixation of the ciliates was only 1% in this study. This concentration is at the lower end of those (0.6 to 20%) in the references (Harris et al. 2000). High concentrations (10% in Throndsen 1978, 10 to 20% in Stoecker et al. 1994) of acid Lugol's solution caused the lowest losses in the ciliate counts. Therefore, the ciliate concentrations in this study may have been underestimated.

The ciliate communities can be divided into 3 parts, as shown in Fig. 3. Part A was a cool area occupied by tintinnids. Part B was an area with high temperature where *Laboea strobila* peaked. *Strombidium compressum* was common in Part C. The characteristic vertical profile of *L. strobila* abundance, if not light related as it is a mixotrophic species (Stoecker et al. 1987), might be attributed to an inclination for warm water. Unlike *L. strobila*, *S. compressum* seemed to prefer the thermocline (Fig. 2). Many authors have discussed the occurrence of ciliates and hydrographic conditions

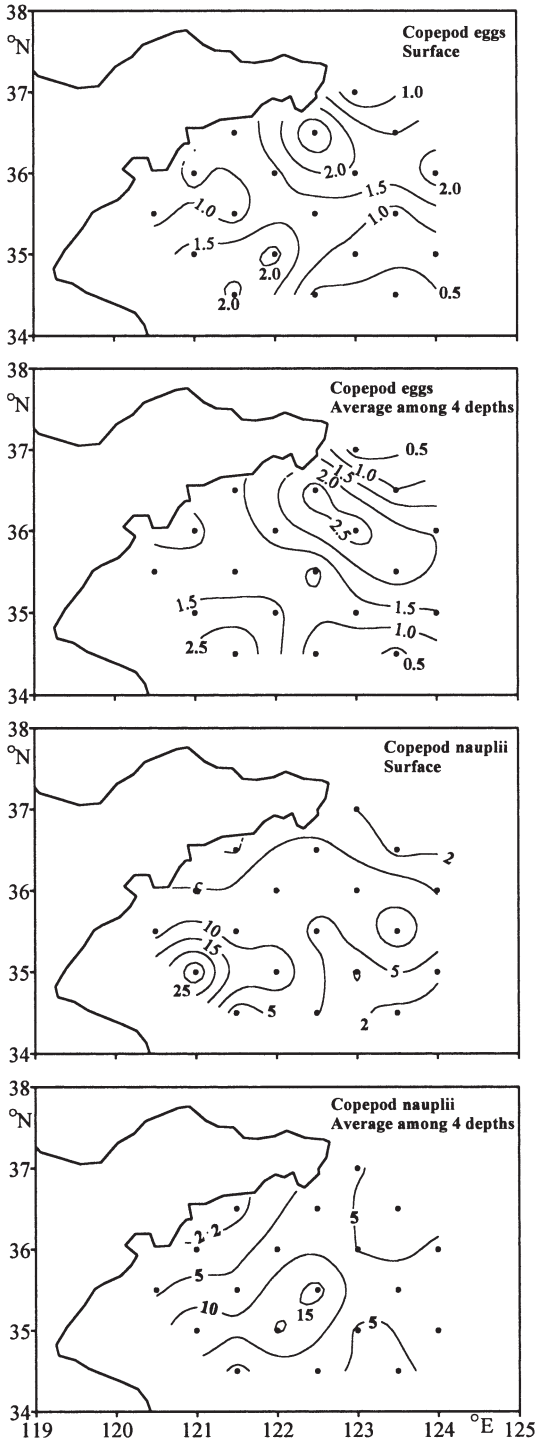

Fig. 6. Abundance (ind. l^{-1}) of copepod eggs and nauplii in the surface waters and the average among 4 depths (surface, 5, 10 and 20 m)

(Sanders 1987, Kamiyama & Tsujino 1996, Cordeiro et al. 1997). Sanders (1987) suggested that microzooplankton abundance and distribution are determined in part by physical factors including temperature,

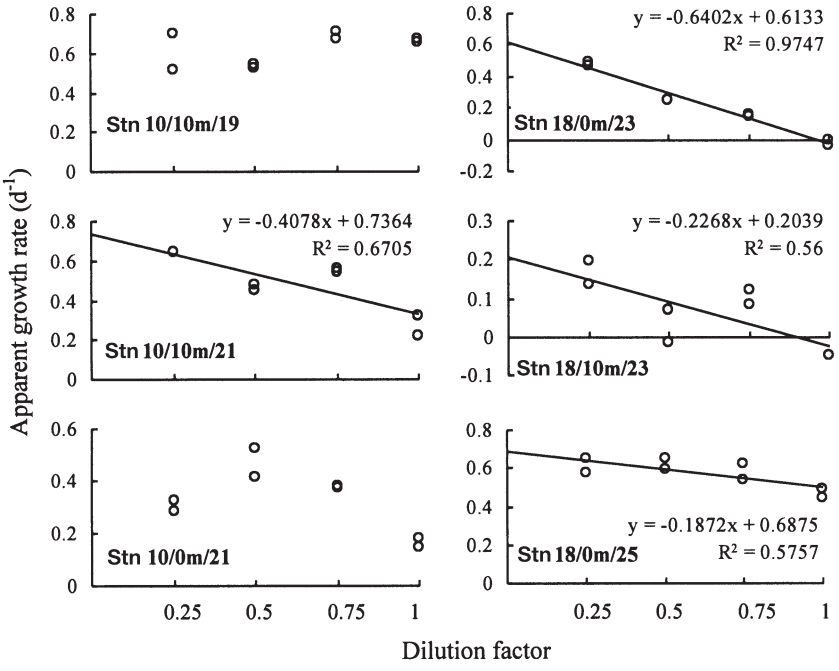

Fig. 7. Regressions between the dilution factors and the apparent growth rates of the dilution experiments

salinity and currents. Kamiyama & Tsujino (1996) noted that the occurrence of most tintinnids was strongly related to temperature, water column stability and abundance of nanophytoplankton, but the particular response to these environmental factors was species specific. In the study of Cordeiro et al. (1997), the distribution patterns of tintinnid species showed a certain relationship with the circulation of waters in the North Sea. In this study, water circulation influenced the distribution of ciliates. It seems that the different ciliate species prefer different environmental factors (especially temperature).

Relationship between copepod nauplii and anchovy larvae

Feeding habits of larval anchovy have been extensively studied by examining gut contents (Park & Cha 1995, Hirakawa & Ogawa 1996, Hirakawa et al. 1997 and references therein). As in this study, copepod eggs and nauplii were the dominant food items while no aloricate ciliate was found in the guts of anchovy larvae in the past studies.

We found that the patch of anchovy larvae did not coincide with that of copepod nauplii and eggs. Anchovy post-larvae were abundant at Stn 17, where the copepod nauplii abundance was low while copepod egg abundance was high. If copepod eggs and nauplii were considered together as the food particles of anchovy larvae, the peak of anchovy larvae abundance was not coincident with that of copepod eggs and nauplii. Similar results were also found in other studies. In Hiroshima Bay, Uye & Yamaoka (1990) found that the spawning ground of the anchovy *Engraulis japonica* was the open bay area while the high densities of copepod nauplii were in the innermost part of the bay. Results of Matsushita et al. (1988) showed that patches of larvae rarely coincided with those of food organisms (copepod nauplii).

Additionally, the abundances of copepod nauplii and eggs were much lower than the threshold food density for survival estimated by rearing experiments. On the basis of laboratory experiment, O'Connell & Raymond (1970) reported that northern anchovy larvae required a minimum concentration of 1000 copepod nauplii l^{-1}. In the study of Laurence (1974), 500 to 3000 copepod nauplii were required for the haddock *Melanogrammus aeglefinus* to survive. Bay anchovy *Anchoa mitchilli* Valenciennes larvae required 1500 to 2000 copepod nauplii l^{-1} to achieve significant survival (Saksena & Houde 1972). The abundance of copepod nauplii in the sea is usually lower than this level (Houde 1978).

Relationship between ciliates and anchovy larvae

Because the abundance of copepod nauplii is usually below threshold concentrations, several authors have suggested that larvae exploit other prey. Lasker et al. (1970) reported that the dinoflagellate *Gymnodinium splendens* might have an important role for survival of first-feeding larvae of the northern anchovy *Engraulis mordax*. Uye & Yamaoka (1990) hypothesized that *E. japonicus* larvae might depend on small scale nauplii patchness, which was not detected in their study (62 to 309 m interval) or on phytoplankton as a supplementary food under scarce nauplii supply. The first-feeding cod larvae seem to have high trophic flexibility by incorporation of algae (as small as 10 µm) and ciliates in their diet. At low copepod density, cod larvae may achieve maintenance ration if they feed on both algae and ciliates (Van der Meeren & Nass 1993). The study of Nagano et al. (2000) showed that both tintinnid and naked ciliates play important roles as alternative food

sources to copepod nauplii by enhancing the survival of fish larvae. Therefore, the ciliates in Stn 17 may have been prey for first-feeding anchovy larvae, preventing initial starvation and prolonging the time to irreversible starvation of fish larvae.

The ciliates in this study were appropriate prey of anchovy larvae in terms of size. The mouth size of the first-feeding anchovy larvae was in the range of 230 to 280 µm (Shirota 1970). According to Hunter & Kimbrell (1980), if the mean diameter of prey eaten by anchovy larvae were about 40% of their mouth width, the anchovy larvae could eat food particles smaller than 80 µm. Laboratory experiments have shown that fish larvae of some species ingest quite small particles such as phytoplankton <10 µm diameter (Van der Meeren 1991). The dimensions of the ciliate species listed in Table 2 fell in the above range.

The dilution incubations

In the design of the dilution incubation method, nutrients should be added to the dilution bottles to prevent them from becoming depleted in the less dilute treatment (Landry & Hassett 1982), especially when nutrient concentrations are low as in this study. The estimated k will be overestimated when no nutrient is added to the dilution series. However, no trace of overestimation of phytoplankton growth rate showed up in Fig. 7. On the other hand, results of dilution incubations with nutrient addition cannot be considered to be *in situ* growth rate of phytoplankton. McManus & Ederington-Cantrell (1992) reported 2 parallel dilution series with and without nutrient addition. The phytoplankton growth rate of the series with nutrient addition was 4-fold higher than that of the series without nutrient addition, while the 2 grazing rates were similar.

Dilution experiments have been carried out in many sites around the world (Dolan et al. 2000). The results of this study (k: 0.14 to 0.74; g: 0 to 0.61) were within the wide ranges of the previous data (k: 0 to 2.14; g: 0 to 2.11; Zhang & Wang 2000 and references therein), but at the lower end. There are 2 possible explanations for the low phytoplankton growth rate and microzooplankton grazing rate. Firstly, the studied area was obviously oligotrophic. Low nutrient (Table 1) concentration limited the phytoplankton growth. Secondly, there may be toxic effects of Niskin sampling bottles. The Niskin water bottles used in this study had internal rubber closure spring and O-rings, which are toxic to a number of protists (Price et al. 1986, Harris et al. 2000).

In this study, 3 parallel experiments were carried out at every station. The growth rates in the same depths of each station were roughly equal (10 m at Stn 10 and 0 m at Stn 18). However, microzooplankton grazing rates showed large variations (0.41 and 0 at Stn 10, 0.61 and 0.19 at Stn 18). The differences suggested that the results of the dilution experiments could only be considered as an instantaneous indication.

Conclusion

We examined the spatial distribution of ciliates, copepod nauplii and eggs, *Engraulis japonicus* post-larvae and microzooplankton herbivorous activity. As a food item of the *E. japonicus* post-larvae, the abundance of copepod nauplii and eggs was low. The peaks of ciliate and *E. japonicus* post-larvae abundance coincided. Although not found in the gut of *E. japonicus* post-larvae, aloricate ciliates might help the first-feeding anchovy larvae to prevent initial starvation and prolong the time to irreversible starvation. The dilution experiments showed that both the phytoplankton growth rate and microzooplankton grazing rate were comparatively low.

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