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Diet Composition and Prey Size of Larval Anchovy, *Engraulis japonicus*, in Toyama Bay, Southern Japan Sea

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Abstract

The types and size of food particles eaten by larvae of the Japanese anchovy, *Engraulis japonicus*, were determined by gut content analysis of larvae (SL: 4.1-8.0mm) captured during daylight hours from Toyama Bay in May 1994. Copepod nauplii were numerically the most important food items. The nauplii eaten consisted of various species. As a whole, cyclopoid (*Oithona* spp.) nauplii were the most important dietary component, with calanoid (mainly *Paracalanus* type) nauplii second. Food composition reflects in part the abundance and species composition of copepods in these waters. The size of prey consumed showed a large range in sizes, from 0.07 to 0.36mm in length and 0.05 to 0.13mm in width. Maximum prey size remarkably increased for larvae from 4.1 to 5.6mm in size (SL), whereas any increase of minimum prey size was indistinct. Within these size restrictions, rare copepod species such as *Calanus sinicus* and *Oithona plumifera* seem to make a major contribution to the larval diet because of the larger body size of these prey, although the more abundant and smaller sized copepods (possibly *Oithona similis*) are more commonly eaten. Food availability for anchovy larvae may depend on reproduction of both warm-and cold-water copepod species in relation to hydrographic structure in the southern Japan Sea.

Key words: copepod nauplii, cyclopoids, *Engraulis japonicus*, gut contents, *Oithona* spp., prey size, Toyama Bay

Introduction

In the Japan Sea, anchovy (*Engraulis japonicus*) is mainly distributed in the waters of the Tsushima Current (FISHERIES AGENCY 1979) and is an important target of many coastal fishing fleets. Anchovy catches have fluctuated by ca. three to six times to the tonnage variation between 1965 and 1988 (HIYAMA 1991). Natural variations in the size of fish populations are generally thought to be determined by mortality during the early life history (planktonic life stage) of fish, as proposed by HJORT (1914, 1926). His hypothesis, as re-stated by MAY (1974), proposed that "the strength of a year-class of fish is determined by the availability of planktonic food shortly after the larval yolk supply has been exhausted"(the so called "critical period"). Apart from predation, the availability of suitable food for larvae is usually considered to be a key factor affecting the mortality of fish larvae and subsequently determining the strength of a year-class.

Data on the feeding habits of larval anchovy are relatively inadequate for the Japan Sea, as compared with those on the Pacific coast (YOKOTA *et al.* 1961; UOTANI *et al.* 1978; UOTANI 1985; UOTANI *et al.* 1988; MITANI 1988 etc.). According to three previous works (AGRICULTURE, FORESTRY

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and FISHERIES RESEARCH COUNCIL 1962; KUWAHARA and SUZUKI 1984; HIRAKAWA and OGAWA 1996), copepod nauplii were the most significant food for anchovy larvae in the Japan Sea as well as in other sea areas. In the warm-water mass (the Tsushima Current), the nauplii eaten consisted of various species, and were suggested to be mainly of temperate and temperate-subtropical copepods (HIRAKAWA and OGAWA 1996). However, there is very little detailed information on the food organisms of the larvae in the southern Japan Sea waters affected by both the warm- and cold-water masses (see NISHIMURA 1969, for a review of the general oceanographic features of the southern Japan Sea) where the dominant prey items found in the larval guts are most likely to be different from those in the warm-water mass alone.

In order to evaluate the effects of the copepod assemblage as a food resource for anchovy larvae, it is necessary to detail the diet of the larvae in relation to the ambient food conditions, which may vary with the hydrographic structure. The present paper describes the characteristic features of the diet composition and prey size of postlarval Japanese anchovy in Toyama Bay, southern Japan Sea. For this purpose, we examined the relationship between the gut-contents of anchovy larvae and the abundance and species composition of the copepod assemblage in the sea. We suggest that the feeding conditions for first feeding anchovy are characterized by the reproduction of both the warm- and cold-water copepod species co-occurring in the southern Japan Sea.

Materials and Methods

1 Samplings of postlarvae and plankton

The survey was performed in the southern Japan Sea from Toyama Bay to Wakasa Bay (18-31 May, 1994), by the training ship "Mizunagi" of the Kyoto Prefectural Kaiyo Senior High School as part of a routine study (GOTO *et al.* 1996) on the recruitment variability and reproductive biology of pelagic fish populations. This study deals with samples obtained in Toyama Bay which forms the main spawning ground of anchovy from spring to summer (FISHERIES AGENCY 1979). Anchovy larvae used for diet analysis were collected on the 21st of May, 1994 at five stations in the Bay (Fig. 1). Collections were made using fish-larval nets (80cm mouth diameter, 0.50mm mesh aperture), which were towed obliquely from 75m depth to the surface.

A zooplankton sample was taken at Stn. 18 which appears to be located on the main route of the warm Tsushima Current which flows into the Bay (IMAMURA *et al.* 1985). The sample was collected by vertical tow of a NORPAC net (45cm mouth diameter, 0.06mm mesh aperture) from 50m depth to the surface. The net was equipped with a Rigosha flow-meter on the mouth ring to register the volume of water passing through the net.

After collection, all the samples were preserved in 5-10% buffered formalin-seawater solution.

2 Gut content analysis

For the anchovy larvae collected during the night at Stns. 13 and 14, there were few individuals with gut contents, as for larval sardine examined from the same samples (HIRAKAWA and GOTO 1996). In other larval anchovy species (*E. mordax* and *E. ringens*), gut contents analyses have indicated that their feeding behaviour is confined to daylight hours: more than 90% of the total number of the larvae with food were caught during the day (BERNER 1959; de MENDIOLA 1974; ARTHUR 1976).

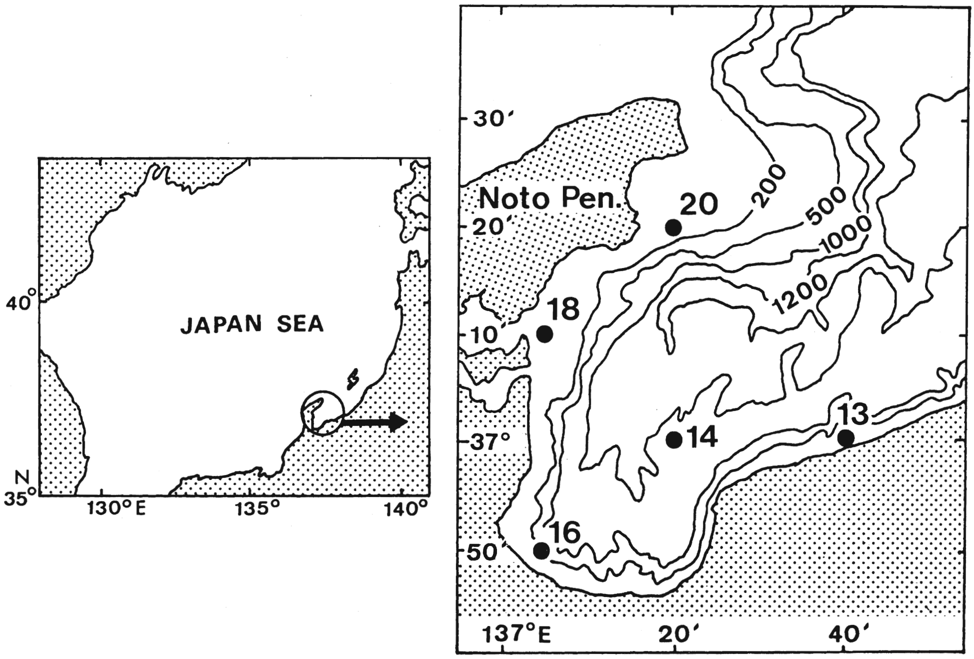

Fig. 1 . Maps showing the location of Toyama Bay (left panel), and the position of the present sampling stations in Toyama Bay (right panel). Bathymetric contours (200, 500, 1000 and 1200m) in Toyama Bay are also given.

From these findings, we concluded that diet composition of the anchovy larvae is represented by that obtained during daylight hours (local time: 05:36 AM-00:12 PM), as judged in gut analysis of sardine larvae (HIRAKAWA and GOTO 1996).

Based on the size (SL: standard length) frequency distribution of the anchovy larvae (GOTO *et al.* unpubl.) in Toyama Bay, we sorted the undamaded specimens in the size category (SL: 4.1 - 8.0mm) from the daytime samples (Stns. 16, 18 and 20) and dipped them into methyl blue-lactic acid solution for staining. They were subsequently measured under a dissecting microscope (SL to the nearest 0.10mm). After this, the guts of 155 larvae were dissected to examine the food organisms inside. The guts of the anchovy larvae were removed and opened with needles. Plankton food organisms in the gut contents were counted, and copepod nauplii were classified into the three fundamental groups [orders: Calanoida, Cyclopoida (-Poecilostomatoida) and Harpacticoida] as characterized by DIETRICH (1915) and BJÖRNBERG (1972). The diagnostic features of copepod nauplii used for the identification to family and genus levels, whenever possible, were the number and arrangement of spines and/or setae on the caudal armature, as described by OGILVIE (1953), FABER (1966), BJÖRNBERG (1972) and KOGA (1984).

Results

1 Gut contents of the early postlarval anchovy

Table 1 shows ingested food items identified from the larvae (mean SL: 5.98-6.48mm) collected

Table 1. Percentage composition of the gut contents of postlarval anchovy (*Engraulis japonicus*) at the three stations.

Food items		Station No. 16	18	20
COPEPODA	Eggs	9.8	7.4	12.5
	Nauplii	80.4	92.1	75.0
	Copepodids	9.8	0.5	
DINOPHYCEAE (Peridineaceae)				12.5
Total No. of food items		51	216	8
No. of larvae examined		26	104	25
SL (mm) of larvae examined (mean ± 1 SD)		6.48±0.91	6.30±0.99	5.98±0.95
Feeding incidence (%)		61.5	48.1	32.0

at the three stations. The larvae fed mostly on copepods of different developmental stages, particularly nauplii. As a whole, copepod nauplii were the most important food item, constituting 75.0-92.1% (mean: 82.5%) of the food items at the three stations; copepod eggs were the second most important (7.4-12.5% of the total) for the larvae. Dinoflagellates (Dinophyceae) were also found, but only occasionally. Feeding incidence (percentage of larvae containing at least one food particle at a particular station) ranged from 32.0% (Stn. 20) to 61.5% (Stn. 16), with a mean of 47.2% which was higher than values reported by YOKOTA *et al.* (1961) and KUWAHARA and SUZUKI (1984) for *E. japonicus*. It is clear that low feeding incidences reported for many larval fishes are not an inviolable rule among early larval clupeoids, as proposed by MAY (1974).

In Table 2, the results of all the observations at the three stations have been combined to give an overall character of the diet in the four larval length groups. Copepod nauplii became increasingly important as anchovy larvae increased in length (SL) and composed almost all (94.7-96.0%) of the ingested food organisms for larval sizes of 6.1-8.0mm. In contrast, the proportion of copepod eggs decreased with increasing size of larvae; from 33.3% for 4.1-5.0mm SL larvae to 1.3% for 7.1-8.0mm SL larvae. Copepodids and dinoflagellates (Dinophyceae) were rarely ingested by larvae of 5.1-8.0 and 7.1-8.0mm SL, respectively.

Table 2. Percentage composition of the gut contents of postlarval anchovy (*Engraulis japonicus*) in the four length groups (SL: 4.1-5.0, 5.1-6.0, 6.1-7.0 and 7.1-8.0mm).

SL (mm)	COPEPODA Eggs	Nauplii	Copepodids	DINOPHYCEAE (Peridineaceae)	Total number of food items
4.1-5.0	33.3	66.7			24
5.1-6.0	12.2	83.8	4.0		74
6.1-7.0	3.0	96.0	1.0		99
7.1-8.0	1.3	94.7	2.7	1.4	76

Table 3 shows the species composition of copepod nauplii found in the guts of the larvae at the three stations. Most of the nauplii were in poor condition making species identification difficult, although some were identified to the genus level. Nauplii consisted of four orders (Calanoida, Cyclopoida, Poecilostomatoida and Harpacticoida) including various species. Cyclopoid nauplii, dominated by *Oithona* spp. , were the most dominant dietary component of larvae at each station, accounting for 58.6-91.0% (mean: 77.6%) of the total number of food items in the study area. Calanoid nauplii were the second most important food item; specifically *Calanus* spp. at Stn. 20 (16.7%), and *Paracalanus* type at Stns. 16 (31.7%) and 18 (5.0%). Poecilostomatoid (*Oncaea* spp.) and harpacticoid (*Microsetella* spp. and *Euterpina acutifrons*) nauplii were found at only one station. Of these copepods, *Microsetella* spp. were the third most important contributor (4.9%) to the total number of food items at Stn. 16. Diet composition was more variable at both Stns. 16 and 18 than that seen at Stn. 20. This difference may have been due to the small sample size from Stn. 20.

Table 4 shows the species composition of copepod nauplii in the guts of larvae in the four length groups, using the data combined for all the observations at the three stations. Cyclopoid nauplii (mostly *Oithona* spp.) were the most significant dietary component in each length group of the larvae, constituting 79.2-88.7% (mean: 84.1%) of the total number of food items. Of calanoid nauplii, *Paracalanus* type was the second most important dietary item of the larvae from 5.1 to 8.0mm SL and *Calanus* spp. the thrid most important for larvae from 6.1 to 8.0mm SL. Poecilostomatoid (*Oncaea* spp.) and harpacticoid (*Microsetella* spp. and *Euterpina acutifrons*) nauplii were an added minor food source (ca. 1.0% of the total) for the larvae from 6.1-7.0mm SL.

Table 3. Composition of copepod nauplii in the guts of postlarval anchovy (*Engraulis japonicus*) at the three stations. Numbers denote their relative abundance (%) to the total (*N*) .

Copepod nauplii		Station No. 16 (*N*=41)	18 (*N*=199)	20 (*N*= 6)
CALANOIDA				
	Calanus spp.	2.4	1.0	16.7
	Paracalanus type[a]	31.7	5.0	
	unidentified		0.5	
CYCLOPOIDA				
	Oithona spp.	58.6	90.5	83.3
	unidentified		0.5	
POECILOSTOMATOIDA				
	Oncaea spp.		0.5	
HARPACTICOIDA				
	Microsetella spp.	4.9		
	Euterpina acutifrons	2.4		
Unidentified			2.0	

[a] including both Paracalanidae and Pseudocalanidae.

Table 4. Composition of copepod nauplii in the guts of postlarval anchovy (*Engraulis japonicus*) in the four length groups (SL: 4.1-5.0, 5.1-6.0, 6.1-7.0 and 7.1-8.0mm). Numbers denote their relative abundance (%) to the total (*N*).

Copepod nauplii		Larval size (SL, mm)			
		4.1-5.0 (*N*=16)	5.1-6.0 (*N*=62)	6.1-7.0 (*N*=95)	7.1-8.0 (*N*=72)
CALANOIDA					
	Calanus spp.			2.1	2.8
	Paracalanus type[a]		9.7	7.4	13.9
	unidentified	6.3			
CYCLOPOIDA					
	Oithona spp.	81.2	88.7	87.4	79.2
	unidentified		1.6		
POECILOSTOMATOIDA					
	Oncaea spp.			1.0	
HARPACTICOIDA					
	Microsetella spp.			1.0	1.4
	Euterpina acutifrons			1.1	
Unidentified		12.5			2.7

[a] including both Paracalanidae and Pseudocalanidae.

Moreover, *Microsetella* spp. were present but rare in the guts of larvae from 7.1-8.0mm SL. Thus, diet composition of first feeding anchovy larvae diversified with increased larval length.

2 Relation between prey size and larval length

All prey size (length and width of ingested copepod eggs and nauplii) data were plotted against the length (SL) of the larval anchovy in Fig. 2. There is a tendency for a marked increase in the size range of prey eaten by larvae between 4.1 and 5.6mm SL; 0.07-036mm in length and 0.06-0.13mm in width. Although maximum prey size did not increase isometrically with the larval length from 5.6 to 8.0mm, the size range of prey selected by the larvae increases as they increase in size (SL: > 8.0mm), as is documented for many other marine larval fish including the three engraulid species, *Engraulis japonicus*, *E. mordax* and *E. ringens* (cf. HUNTER 1981). Any increase of minimum prey size was indistinct.

Each size range of the ingested eggs and eight naupliar taxa of copepods is indicated on the right of Fig. 2, for determining constituents of diet of larvae within a certain size range. In prey length (upper panel in Fig. 2), copepod eggs were limited to a size range of 0.07 to 0.11mm. Taxa including some of the small nauplii (*Oithona* spp. , *Paracalanus* type, *Oncaea* spp. -*Euterpina acutifrons*, *Microsetella* spp.) showed a size range of 0.10 to 0.25mm. Larger nauplii, consisting of mainly *Oithona* spp. and *Calanus* spp. , showed a size range of 0.26 to 0.36mm. As larvae increased in size, there was an apparent shift of preferred food from small to large nauplii. This shift in diet composition was also recognizable by increased prey width (lower panel in Fig. 2).

The large-sized naupliar group is an important food resource for increasing the maximum size of prey items ingested by larval anchovy from 4.7 to 8.0mm SL. While this prey size group

(> 0.26mm in length) comprised only 3.4% of the total prey items, the small-sized naupliar group with a length range of 0.11 to 0.25mm accounted for 87.8% of the total (Fig. 3). In addition, prey items with a width range from 0.05 to 0.11mm accounted for 96.2% of the total and peaked at 0.07mm width (Fig. 3). Thus, the fish larvae infrequently took the largest prey, but in general fed

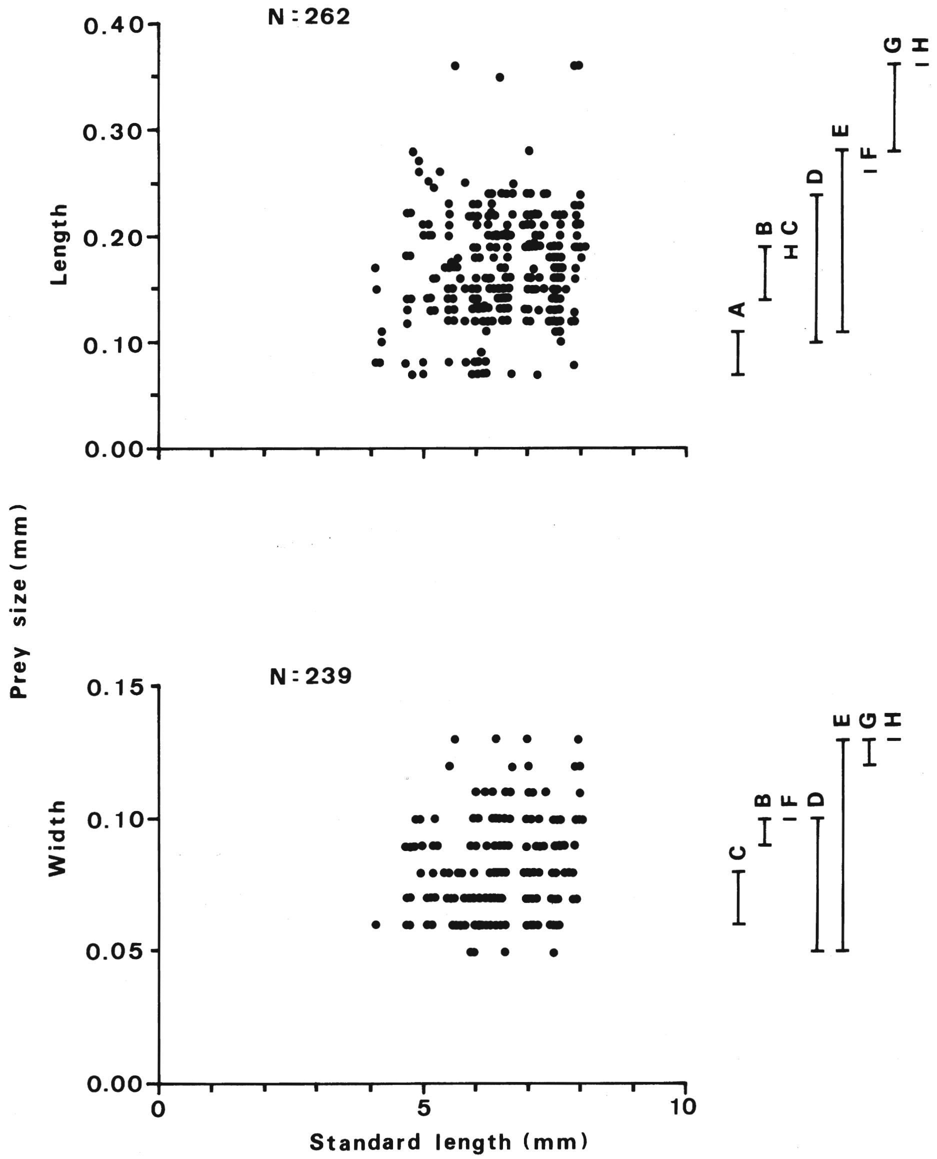

Fig. 2 . Relation between prey size (upper panel: naupliar length including egg diameter of copepods, lower panel: naupliar width) and larval standard length of *Engraulis japonicus*. Vertical bars on the right denote the size ranges of copepod eggs (A) and of the eight naupliar taxa (B: *Microsetella* spp. -*Euterpina acutifrons*, C: *Oncaea* spp. , D: *Paracalanus* type, E: *Oithona* spp. , F: Calanoida, G: *Calanus* spp. , H: Cyclopoida) found in the guts of the larvae.

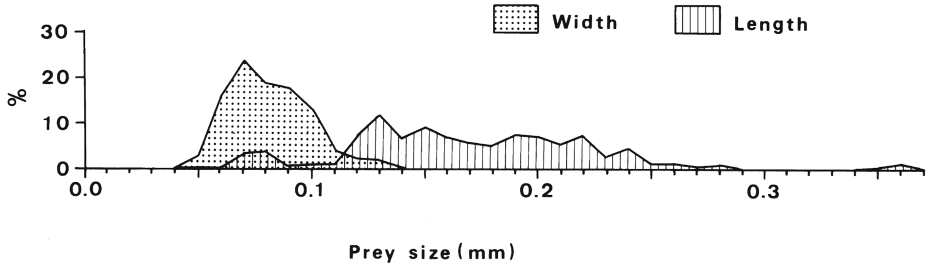

Fig. 3 . Percentage size (length and width) distribution of copepod eggs and nauplii found in the guts of postlarval anchovy (*Engraulis japonicus*). The data are the same as in Fig. 2.

mainly on food items in the prey size range considerably less than the maximum prey size which could have been ingested. This implies that the small-sized copepods (naupliar size: 0.11 to 0.25mm in length, 0.05 to 0.11mm in width) are more available to the larvae than the large-sized copepods.

3 Species composition of the copepod assemblage in Toyama Bay

Table 5 shows the species composition in the two developmental stages (nauplii and copepodids-adults) of copepods collected from Stn. 18. In naupliar stages, cyclopoids (mostly *Oithona* spp.) were most dominant, accounting for 80.5% of the total number of nauplii. Calanoids were composed of three taxa of which *Paracalanus* type was the next most important nauplii (11.4%), following *Oithona* spp. The harpacticoid *Microsetella norvegica* was the third most important component

Table 5. Species composition in the two developmental stages (nauplii and copepodids-adults) of copepods collected from Stn. 18 in Toyama Bay on 21 May, 1994. Numbers denote their relative abundance (%) to the total (N: ind/m^3).

Orders	Developmental stages			
	Nauplii (N=578)		Copepodids-Adults (N=143)	
CALANOIDA	*Calanus* spp.	1.7	*Calanus sinicus*	1.1
	Paracalanus type [a]	11.4	*Paracalanus parvus*	18.4
	Labidocera spp.	0.6	*Clausocalanus pergens*	3.8
			Acartia sp.	0.6
CYCLOPOIDA	*Oithona* spp.	80.5	*Oithona similis*	32.3
	unidentified	0.3	*Oithona plumifera*	1.2
			Oithona nana	0.1
POECILOSTOMATOIDA	*Oncaea* spp.	1.5	*Oncaea media*	24.8
			Oncaea venusta	0.2
			Corycaeus affinis	1.1
			Saphirella sp.	0.4
HARPACTICOIDA	*Microsetella* spp.	4.0	*Microsetella norvegica*	15.9
			Microsetella rosea	0.1

[a] including both Paracalanidae and Pseudocalanidae.

of the copepod nauplii. In the copepodid-adult stages, *Oithona similis*, *Oncaea media*, *Paracalanus parvus* and *Microsetella norvegica* which represented each order, occurred more abundantly. Of these copepods, *O. similis* was the most dominant (32.3% of the total), followed by *O. media* (24.8% of the total). *P. parvus* and *M. norvegica* were almost equal in their relative abundance (18.4% and 15.9% respectively). From these findings, it is apparent that cyclopoids, dominated by *O. similis* are the most important constituent in the copepod assemblage throughout the developmental stages.

Discussion

From the gut content analysis of larval anchovy (SL: $<$ 8.0mm), the important food source (copepod nauplii) for the larvae is shown to be dependent on the nauplii production of various copepod species listed in Table 3. Of these copepod nauplii, the relative abundances (Table 3) of nauplii of *Calanus* spp. , *Paracalanus* type and *Oithona* spp. found in the guts of larvae collected from St. 18 closely correlate with those observed in the plankton net samples (Table 5). Therefore, the diet composition is in part a reflection of the abundance and species composition of the copepod assemblage in the field.

On the other hand, the nauplii of *Oncaea* spp. and *Microsetella* spp. were scarce (Table 3) in the diet composition of the larvae in spite of the higher relative abundance of copepodids and adults of *O. media* (24.8%), and *M. norvegica* (15.9%) in the field. It is possible that the nauplii of these species were not well sampled by the 0.06mm mesh net used in this study because of their short body length; 0.06-0.07mm for *O. media* (MALT 1982) and 0.08mm for *M. norvegica* (HIRAKAWA 1974). However, nauplii of *Oncaea* spp. and *Microsetella* spp. were also less abundant in the plankton samples retained on the 0.02mm mesh net by filtering 10 l seawater collected with a 12 l Niskin bottle (HIRAKAWA unpubl.). These inconsistent results may be due to an inactive period of the reproductive cycle of these species rather than the influence of loss of nauplii through the net.

Oithona spp. are clearly important as food items for first feeding anchovy larvae in Toyama Bay, because of their dominance in both the gut contents and field zooplankton samples. In particular, *O. similis* was (1) the most dominant species of all the copepods sampled (Table 5), with (2) their naupliar I-VI stages ranging from 0.11 to 0.23mm in body length (GIBBONS and OGILVIE 1933) which was (3) within the size range (0.05-0.36mm) of ingestible prey for anchovy larvae (Fig. 3). Moreover, *O. similis* (copepodids and adults) occurred more abundantly from April to July in Toyama Bay, based on data obtained throughout the year (HIRAKAWA unpubl.). Thus, it is possible that this species is most useful as a food source for larval anchovy in copepod assemblages from spring to summer, as well as for larval sardine (*Sardinops melanostictus*) obtained from the same collections (HIRAKAWA and GOTO 1996). *S. sagax* in the California Current may also utilize *O. similis* as its main food source (ARTHUR 1977).

Analyses to species level were difficult for the large-sized oithonid nauplii ($>$ 0.24mm in body length) in the present study (Fig. 2). Of the three oithonid species listed in Table 5, the largest species, *O. plumifera*, appears to produce large-sized nauplii (I to VI stages) from 0.15 to ca. 0.30mm in body length (BJÖRNBERG 1972 ; HIRAKAWA unpubl.). Moreover, nauplii of *Calanus* spp. are the major constituent of the food organims larger than 0.30mm in body length (Fig. 2). From the zooplankton samples, *C. sinicus* was the only representative of the calanoid genus *Calanus* (Table 5). This species shows a body length range of 0.19 to 0.52mm for the naupliar stages Ⅰ to Ⅵ (KOGA 1984; UYE 1988) and therefore their nauplii are relatively large, as compared with those of *O. plumifera*. Although *C. sinicus* and *O. plumifera* occurred rarely in the field, both species play a potentially significant role for larval growth (4.7 to 8.0mm in SL), considering the higher nutritive value of larger prey from the relation between width of copepods and their weight (HUNTER 1981).

Oithona similis is widely distributed in the Atlantic, Indian and Pacific Oceans, and is adapted to somewhat low temperature conditions (NISHIDA *et al.* 1977). In addition, this species occurred more abundantly in the areas of cold water upwelling or mixing with warm water in the waters of south Sakhalin (including the northern Japan Sea), the southern Kurile Islands (BRODSKY 1959), and the central part of the Japan Sea where the influence of the warm Tsushima Current was minimal (NISHIDA and MARUMO 1982). From these previous records, *O. similis* is regarded as a cold-water species exhibiting ecological requirements different from those of *C. sinicus* and *O. plumifera* which are warm-water species grouped as the temperate and temperate-subtropical species, respectively (HIRAKAWA and OGAWA 1996).

In conclusion, it is suggested that the food availability to anchovy larvae depends on the reproductive activities of both the warm-water (large-sized) species and cold-water (small-sized) species which are closely associated with the water masses in the southern Japan Sea coastal waters.

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富山湾におけるカタクチイワシ仔魚の餌料組成とそのサイズ

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カタクチイワシ(*Engraulis japonicus*)の初期餌料特性を明らかにするため，1994年5月富山湾から採集された後期仔魚(体長：4.1～8.0mm)の消化管内容物を分析した．カイアシ類ノープリウス幼生が個体数で最も重要な餌料生物であり，複数種から成る．ノープリウス幼生のうち，餌料としてCyclopoida(*Oithona* spp.)が最重要であり，次にCalanoida(主に*Paracalanus* type)が続く．最大餌料サイズ(体長・体幅)は摂餌開始直後の仔魚の成長に伴い増加したが，最小餌料サイズはほぼ一定であった。これら結果と野外におけるカイアシ類群集の種組成との関連から，日本海南部沿岸水域におけるカタクチイワシ仔魚の餌料生物として，暖水性カイアシ類と冷水性カイアシ類(恐らく*Oithona similis*)の両者が重要な役割を担っていることを指摘した．