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Correction Factors for Quantitative Analysis of Anchovy Eggs and Larval Stages from the Southern Waters of Korea

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Abstract – Correction factors based on the catch ratios of egg and larval densities in the southern waters of Korea were estimated for anchovy *Engraulis japonica*. This was undertaken in order to adjust ichthyoplankton data from different sampling methods, gear types and time. Samples were collected during ichthyoplankton surveys in Korean waters from 1983 to 1994. The ratios for egg densities obtained in vertical tows with a NORPAC net (ring Φ, 45 cm) compared to those obtained in oblique tows with a KOB net (ring Φ, 80 cm) were 0.86 (CV = 0.65), 1.22 (CV = 0.36), and 0.93 (CV = 0.42) for early, middle, and later developmental stages, respectively. The ratios for larval densities for vertical and oblique tows varied depending on size. For yolk-sac and small larvae (< 4 mm), the ratios were 3.08 (CV = 0.45) and 1.98 (CV = 1.34), while those of 4-6 mm, 6-8 mm, and 8-10 mm larvae were 0.44 (CV = 1.31), 0.45 (CV = 1.70), and 0.56 (CV = 2.50), respectively. Ratios of day/night densities for larvae of 4-10 mm lengths were lower (0.01-0.06) in offshore catches than values obtained in coastal areas (0.44-0.46) and similar values (0.16-0.04) for vertical and oblique tows. Our results indicated that vertical towing is more efficient for sampling early life stages (from eggs to larvae less than 4 mm long), while oblique towing is more efficient for larvae longer than 4 mm due to depth preferences for each developmental stage (*e.g.*, changes in egg buoyancy and vertical migration of larvae).

Key words – correction factors, ichthyoplankton, oblique, vertical, catch ratio, anchovy

1. Introduction

In Korea, ichthyoplankton surveys have been frequently conducted to study habitats characteristics and population variations of fish stocks (Lim *et al.* 1970; Kim and Lo 2001). Due to the different methodologies applied, raw data cannot be easily used for quantitative analyses of mortality and production in early life stages, as well as comparative studies among regions and years. The change in methodology requires the understanding and intercalibration of measurements produced by different methods if long temporal histories are to be reconstructed (Ohman and Laveniegos 2002). For the effective management of pelagic fish populations that are dependent on recruitment variations affected by early life-stage survival, it may be sufficient to monitor the age structure of the catch as well as the production and survival of embryonic stages (Methot and Lo 1987). In the California current, the abundance of anchovy eggs and larvae was re-estimated using the retention rate of sampling net types, mesh sizes, and the time of day, as correction factors for possible biases (Lo 1983). Several authors have analyzed the sampling efficiency of various methods for zooplankton in the California current (Ohman and Smith 1995; Ohman and Lavaniegos 2002). In Korea, abundance of larval fish sampled in upper and lower layers during the day and at night were compared to investigate a proper sampling method for larval fish off western coastal waters (Cha and Park 1995).

Anchovy (*Engraulis japonica*) is a typical pelagic fisheries resource and is widely distributed in the coastal waters of the Korean peninsula. Here, anchovy juveniles recruit to the fishery within one or two months of spawning. Juvenile production is thus very important to the economy of anchovy fisheries and also to food chains within the marine ecosystem, leading to higher trophic fish levels (Kim and Kim 1986; Huh 1999). Quantitative ichthyoplankton studies enable predictions of the recruitment and population responses to dynamic changes. A correction factor is needed when a net tow does not retain all eggs and larvae are encountered to correct the possible bias. In this paper, we derived correction factors for the quantitative analysis of early life stages for anchovy in Korean waters

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and discussed the necessity for developmental stage-specific correction factors for different sampling methods, gears, and sampling time.

2. Materials and Methods

Thirty-seven ichthyoplankton surveys were conducted in the southern waters of Korea from 1983 to 1994 (Fig. 1). From 1983 to 1989, ichthyoplankton samples were collected with a KOB 80 net (80-cm diameter ring net with a 0.333-mm mesh size) obliquely for 10 minutes, from a depth of 30 m to the surface at speeds of 1-2 knots for the vessel in charge of the survey. From 1987 to 1994, samples were collected with a NORPAC net (45-cm diameter ring with the same mesh size) towed vertically from the bottom to the surface at speeds of 0.5-1.0 m $\cdot$ sec^{-1}. A flowmeter is mounted in the mouth of each net to provide data on the volume of water filtered during each tow. The procedures of sampling and flowmeter calibration in the vessel followed the methods of Smith and Richardson (1977).

Anchovy eggs and larvae were removed from ichthyoplankton samples, identified, staged/measured, and counted. Values were standardized to numbers per 1 m^2 surface area in each sample (Smith and Richardson 1977). Eggs were classified into one of three developmental stages on the basis of segmentation and blastodisc formation (stage I), blastoderm development (stage II), and separation of the tail from the yolk mass (stage III) (Kim and Lo 2001). Larvae were classified into the following groups: the yolk-sac stage, < 4 mm, 4-6, 6-8, and 8-10 mm in total length.

To calculate correction factors based on the catch ratios associated with net types and towing methods, we compared egg and larval densities from 103 paired samples taken with both net types in 1988-1989. The mean density and standard deviation of eggs and yolk-sac larvae were tested with 1000 bootstrap iterations. CV (Coefficient of Variation) is the ratio of standard error to the mean.

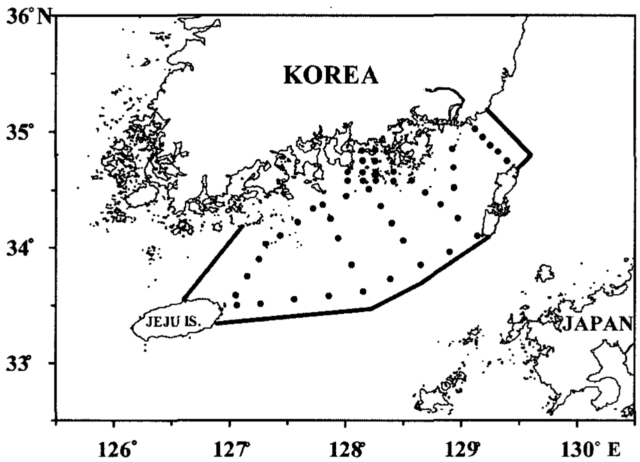

Fig. 1. Map showing sampling area and typical stations for anchovy eggs and larvae in the southern waters of Korea during 1983-1994.

To understand the day and night differences in the catches of eggs and larvae, we compared the densities of eggs and larvae every hour to those obtained at night (21:00-04:00) by computing the ratio of densities based on samples taken by a KOB net in 1983-1989 and a NORPAC net in 1987-1994. Catch ratios in all samples for each larval group (4-6, 6-8, 8-10 mm), divided by the density during the night (21:00-04:00), were estimated to indicate intense daytime biases in the samples by the KOB net in 1983-1989 and the NORPAC net in 1987-1994.

The correction factors for the net avoidance in each larval group of different length classifications (4-6, 6-8, 8-10 mm) and different sampling methods (vertical and oblique tows) were estimated by ratios in the morning, day, evening and night in 4 intervals (*i.e.*, 21:00-04:00, 04:00-09:00, 09:00-16:00, and 16:00-21:00) divided by the density obtained during the night (21:00-04:00). We compared the net avoidance for coastal (above 50 m bottom layer) and offshore areas (below 50 m bottom layer) by computing the ratio of the mean density in the morning, day, evening and night in 4 intervals (*i.e.*, 21:00-04:00, 04:00-09:00, 09:00-16:00, and 16:00-21:00) divided by the density during the night (21:00-04:00).

3. Results

We computed the mean densities of anchovy egg and larval developmental stages obtained by NORPAC (*i.e.*, vertical tow) and KOB (*i.e.*, oblique tow) in 1988-1989. Differences in catch were found with respect to gear types/sampling methods (Fig. 2). We found that the mean density of stage I eggs from vertical net tows was 84% of that obtained by oblique tows. Stage II eggs, however, obtained by oblique tows, was 79% of that obtained by vertical tows. Little difference in density in stage III eggs were found between these two nets. During the yolk-sac and larval stages (smaller than 4 mm in total length), densities collected by vertical tows were, respectively, 3-fold and 1.8-fold the densities collected by oblique tows. Larger larvae for 4-6 mm, 6-8 mm, and 8-10 mm were 0.42, 0.42, and 0.50 times less in vertical tows than oblique tows.

For the three egg stages, 1000 bootstrap iterations estimated that the ratios of egg densities sampled by the NORPAC net relative to the KOB 80 net were 0.86 (CV = 0.65), 1.22 (CV = 0.36), and 0.93 (CV = 0.42) (Table 1). For yolk-sac larvae, this ratio was 3.08 (CV = 0.45). The ratio for larvae smaller than 4 mm in total length was 1.98 (CV = 1.34); for larvae 4-6 mm long, 0.44 (CV = 1.31); for larvae 6-8 mm long, 0.45 (CV = 1.70); and for larvae 8-10 mm long, 0.56 (CV = 2.50). Each stage of eggs and length-group of

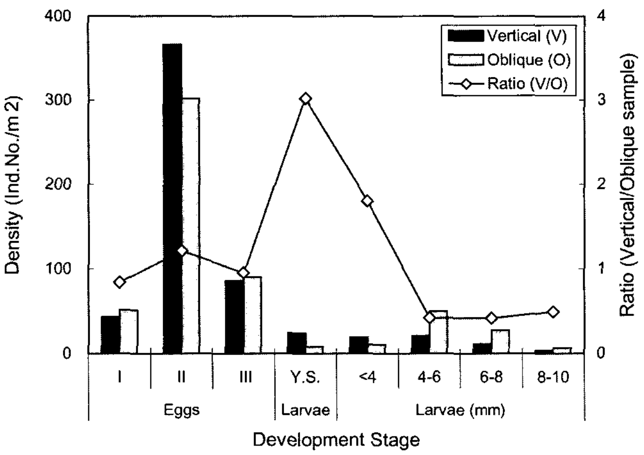

Fig. 2. Anchovy egg and larval mean densities and ratios for each stage sampled vertically with a NORPAC net (45-cm ring) and obliquely with a KOB net (80-cm ring) in the southern waters of Korea during 1988-1989. Eggs were assigned to one of three developmental stages: segmentation and blastodisc formation (I), development of blast (II), and separation of the tail from the yolk mass (III). Larvae were assigned to the yolk-sac larval stage (Y.S.), larvae less than 4 mm in total length, and larvae 4-10 mm by 2 mm intervals.

larvae showed significant differences in catch due to gears of NORPAC and KOB 80 net, and sampling methods of vertical and oblique towing ($P<0.05$).

We computed the ratios of egg and larval density every hour to the mean density at night (22:00-04:00) for samples of vertical tows of the NORPAC net from 1988 to 1994 and oblique tows of the KOB net from 1983 to 1989 (Fig. 3). For both these two nets, results were similar for eggs and larval stages smaller than 4 mm in total length. Fewer larvae (4-10 mm total length) were collected during the day relative to catches at night.

Catch ratios, the overall mean value in 24 hours divided by the density at night (22:00-04:00), showed that larvae longer than 4 mm appeared to be able to avoid vertical NORPAC tows with a higher degree than the oblique KOB 80 tows (Fig. 4). These values indicating for net avoidance behaviors by anchovy larvae of all lengths (4-10 mm) ranged from 0.20-0.27 for the vertical towing and 0.24-0.36 for the oblique towing. Catch ratio for the net avoidance ranged from 0.27-0.36 for larvae 4-6 mm in total length and decreased with larval growth, reaching 0.20-0.24 in larvae of 8-10 mm for both towing methods.

Both vertical and oblique tows showed low density ratios for the larvae 4-10 mm during the day (Table 2). Catch ratios for larvae (4-10 mm total length) obtained in daytime versus those caught at night were in the range of 0.16-0.04 for vertical tows and 0.12-0.04 for oblique tows. These values increased in the range of 0.07-0.66 in the morning, and decreased in the range of 0.03-0.12 in the evening. In daytime, larvae of this length were caught at higher density ratios in coastal areas (0.44-0.46) as opposed to offshore (0.01-0.06) (Table 3).

Table 1. Comparison of anchovy egg and larval densities determined by vertical towing with a NORPAC net (ring diameter, 45 cm) and oblique towing with a KOB 80 net (ring diameter, 80 cm) in Korean waters during May-July of 1988 to 1989. Re-sampling for the mean and variation was carried out by 1000 bootstrap iterations for the estimation of variance (Var), standard deviation (SD), covariance (Covar) and coefficient variation (CV).

Parameter/Stage	Eggs			Y.L.[4]	Larvae (mm)			
	a[1]	b[2]	c[3]		< 4	4-6	6-8	8-10
Vertical (V)								
Mean	43.80	368.22	84.69	24.78	21.26	21.76	11.84	3.86
SD	19.92	90.40	21.89	8.32	27.86	25.44	18.57	9.25
Var	396.65	8172.62	479.39	69.22	775.98	647.19	344.72	85.61
Oblique(O)								
Mean	51.13	301.06	90.62	8.04	10.73	50.01	26.10	6.83
SD	22.92	79.27	29.95	2.51	3.15	29.83	17.24	4.85
Var	525.51	6283.19	897.24	6.30	9.93	889.66	297.08	23.50
Covar (V,O)	-9.50	-61.03	11.46	0.33	0.69	4.63	2.26	0.03
Ver/Obl (V/O)								
ratio	0.86	1.22	0.93	3.08	1.98	0.44	0.45	0.56
var	0.31	0.20	0.15	1.97	7.06	0.32	0.59	1.99
SD	0.55	0.44	0.39	1.40	2.66	0.57	0.77	1.41
CV	0.65	0.36	0.42	0.45	1.34	1.31	1.70	2.50

[1]Segmentation and blastodisc formation
[2]Development of blast
[3]Separation of the tail from the yolk mass
[4]Y.L.: Yolk-sac larvae

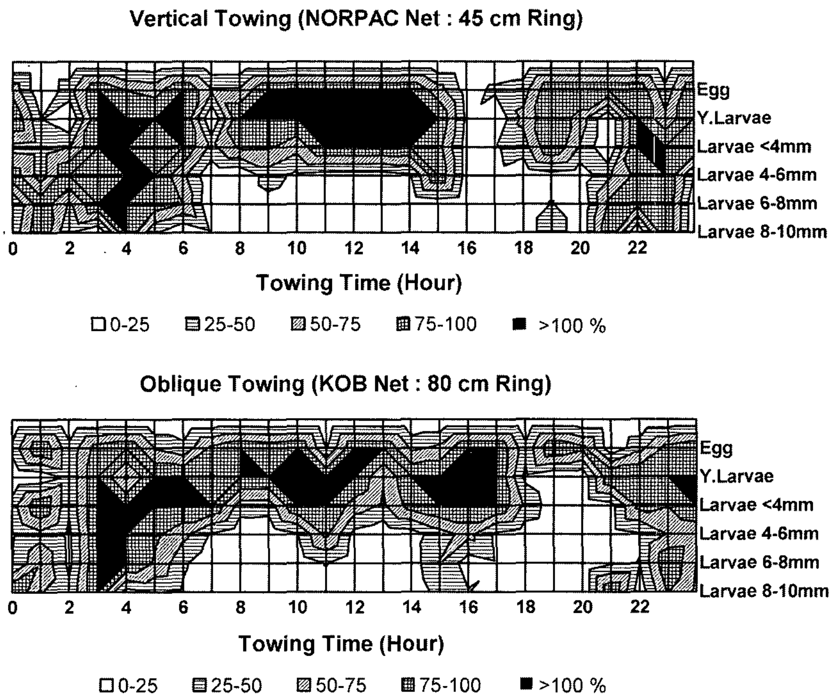

Fig. 3. Time-based variation of the anchovy eggs and larval relative densities in comparison with the mean density during nighttime (21:00-04:00) for each stage determined by vertical towing with a NORPAC net during 1988-1994 (upper) and by oblique towing with a KOB net during 1983-1989 (lower).

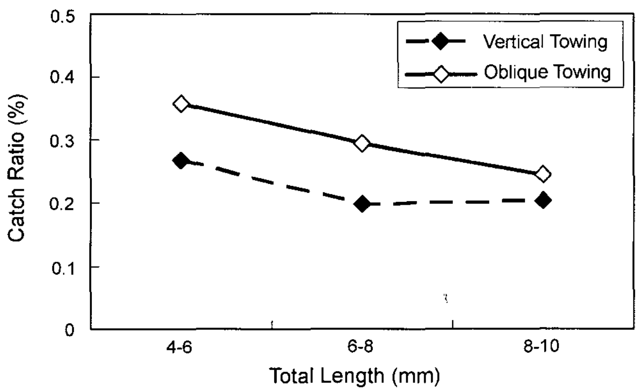

Fig. 4. Avoidance rates from all anchovy larval densities for each length group (4-6, 6-8, 8-10 mm) divided by the density obtained during the night (21:00-04:00), by vertical towing with a NORPAC net during 1988-1994 and by oblique towing with a KOB net during 1983-1989 in Korea waters.

4. Discussion

For eggs, the correction factors based on the catch ratios for towing methods were 7-14% lower for vertical tows relative to oblique tows during cell-division and tail-free stages, but significantly higher during the blastodisc stage (P<005). There were also significant differences in catch for all class sizes for yolk-sac and larvae according to net and sampling types (P<005). These results showed that the vertical distributions of eggs and larvae in the upper 100 m of the water column were different in terms of their developmental stages, because eggs are spawned at 30-60 m depth and descend to the deeper area during the development of eggs and yolk-sac larvae (Kim and Choi 1988). We sampled obliquely with a KOB 80 net for 10 minutes from a depth of 30 m to the surface and vertically with a NORPAC net from the bottom to the surface. The perceived distribution of each stage of eggs and larvae was significantly different between vertical and oblique tows owing to differences in developmental stage-based distribution depth, sampling layer depth, and size of the net ring. For larvae > 4 mm, the lower densities by vertical tows may mean they avoid the vertical tows more than that of oblique tows. Our results showed that the depth preferences for each developmental stage (*e.g.*, changes in egg buoyancy and vertical migration of larvae) significantly affect apparent sample density according to gear type and sampling time.

When comparing macrozooplankton sampling methods over the course of the CalCOFI time series, Ohman and Smith (1995) found that biomass density was greater in shallower samples and that bongo nets were more effective collectors of juveniles and adults, while 1.0-ring nets were better collectors of young larval stages. Different species diversities are collected by these two net types (Ohman

Table 2. Comparison of larval densities for each length group with respect to the towing type and the towing time obliquely with a KOB net (80-cm ring) during 1983-1988 and vertically with a NORPAC net (45-cm ring) during 1988-1994 in the southern waters of Korea. Avoidance rates of anchovy larvae for each length group were estimated for the catch ratios of the mean density during the morning, day, evening, and night divided by the mean density during the night.

Length Group	Vertical Towing						Oblique Towing					
	Density(No./m²)		Catch Ratio				Density(No./m²)		Catch Ratio			
(mm)	D[3]	N[1]	N[1]	M[2]	D[3]	E[4]	D[3]	N[1]	N[1]	M[2]	D[3]	E[4]
4-6	5.64	35.54	1	0.50	0.16	0.09	6.49	53.01	1	0.45	0.12	0.12
6-8	1.28	19.89	1	0.48	0.06	0.06	2.22	33.65	1	0.28	0.07	0.04
8-10	0.24	5.96	1	0.66	0.04	0.12	0.79	20.81	1	0.07	0.04	0.03
No. of Sample	316	50	50	36	316	61	227	110	110	84	227	100

[1]Night (21:00-04:00), [2]Morning (04:00-09:00), [3]Day (09:00-16:00), [4]Evening (16:00-21:00)

Table 3. Comparison of larval densities for each length group with respect to the towing time in the coastal (<50 m depth layer) and offshore areas (>50 m depth layer) obliquely with a KOB net (80-cm ring) during 1983-1988 in the Korean waters. The avoidance rates of anchovy larvae for each length group were estimated for the catch ratios of the mean density during the morning, day, evening, and night, divided by the mean density obtained during the night.

Length Group	Coastal Area						Offshore Area					
	Density(No./m²)		Catch Ratio				Density(No./m²)		Catch Ratio			
(mm)	D[3]	N[1]	N[1]	M[2]	D[3]	E[4]	D[3]	N[1]	N[1]	M[2]	D[3]	E[4]
4-6	11.00	25.18	1	0.73	0.44	0.60	3.40	53.86	1	0.60	0.06	0.06
6-8	4.29	9.00	1	0.81	0.48	0.36	0.79	36.05	1	0.33	0.02	0.02
8-10	1.71	3.73	1	0.55	0.46	0.60	0.22	23.23	1	0.02	0.01	0.01
No. of Sample	164	45	45	57	164	40	63	64	64	27	63	60

[1]Night (21:00-04:00), [2]Morning (04:00-09:00), [3]Day (09:00-16:00), [4]Evening (16:00-21:00)

and Laveniegos 2002). We found lower catch ratio for larvae 4-10 mm in length in the coastal area than offshore with oblique tows. Larvae of 4-10 mm were more likely to be caught in coastal areas than offshore in the middle of the day (Table 3). Our observations indicated that vertical towing is more efficient for sampling early life stages (from eggs to larvae less than 4 mm long), while oblique towing is more efficient for larvae longer than 4 mm. Those are consistent with the survey practice in California water: Vertical tows for anchovy and sardine eggs and yolk-sac larvae and oblique tows for larvae < 10 mm (Lo 1983; Lo *et al.* 1989).

Ichthyoplankton sampling results are subject to biases related to extrusion and avoidance specific for each sampling gear, towing method, and towing time (Lo, 1983; Lo *et al.* 1989). For example, large anchovy larvae can avoid bongo nets of less than 1.0-m in diameter (Hewitt 1980). Samples at upper layers during the daytime causes an underestimation of the number of species and the abundance (Cha and Park 1995).

In this study, avoidance was detected by larvae longer than 4 mm (Fig. 4). The ratio from all samples of each length group (4-6, 6-8, 8-10 mm) to the density at nighttime ranged from 0.36-0.20, a lower value than the ratio obtained in the morning, day, evening and nighttime (Fig. 4, Tables 2 and 3). This indicates that avoidance has some effects on the towing time such as concentration of daytime surveys in Korean waters, 1983-1994. In the western coastal waters of Korea, abundance through the whole water column during the daytime is not significantly smaller than those measured after nightfall (Cha and Park 1995). When we compare the ratios of day/night densities for larvae in the offshore and coastal waters, the values obtained offshore were lower (0.01-0.06) than those of coastal areas (0.44-0.46). Therefore, careful planning for sampling is necessary in future Korean surveys to eliminate biases resulting from towing time and towing depth.

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