Thesis for the Degree of Master of Science

Age and growth of the white-spotted conger eel, *Conger myriaster* (Brevoort) in the Southern sea of Korea

by

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(남해안에 서식하는 붕장어의 연령과 성장)

Advisor: Prof. Chul-Woong Oh

by

Jin Ho Bae

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Abstract

The present study is to provide a better understanding of the age and growth of the white-spotted conger eel, *Conger myriaster* in the Southern sea of Korea. This study has been investigated about age, growth and reproductive traits of the white-spotted conger eel. Samples were collected by trap, shrimp trawl and bottom trawl from April 2009 to November 2011 from the Southern areas and the coastal water in Geoje Island.

Age validation is observed by UV light and the relative growth is estimated based on the total length, weight and otolith readings. This is a detailed report clarifying the accuracy of the age determination by using a UV light observation. In addition, the age structure is estimated by defining the relationship between otoliths measurements and age. The age of 212 otoliths ranged from 1 to 13 years. The age composition of congers in coastal water of Geoje Island is turned out to be dominant from 1 to 3 years old while that

in offshore of southern sea ranged from 3 to 13 years old. Conger eel showed a wide variation of total length in the same age composition. The parameters of von Bertalanffy growth function were estimated from three-methods by non-linear regression: back-calculation method, mean total length at age and otolith weight at age. The results of adjusted coefficient of determination nearly matched with estimated values for the three methods (r^2>0.97). The best fitted result was estimated as following VBGF parameters; L_∞=127.95 cm, K=0.102, t_0=-0.922.

Maximum oocyte diameter ranged from 50 to 430 ㎛. Reproductive traits of ovaries showed positive relationship between GSI and MOD in the regression of power function. It is suggested that oogenesis begins to develop from 4 years old at 45cm TL.

1. Introduction

The white-spotted conger eel, *Conger myriaster* (Brevoort, 1856) is distributed from the East China Sea to the waters around Korea and Japan to the edge of the continental shelf (Kubota, 1961; Yamada, 1986; Park, 2001; Tokimura, 2001). This species has leptocephalus larval stage, which lasts for about six months (Lee and Byun, 1996). Anguilliform fish larvae are transported from offshore to inshore coastal waters. It is a large-scaled reproductive migration (Correia et al., 2004; Kimura et al., 2004; Katayama and Kurogi, 2008). Spawning area of *C. myriaster* has remained as mystery for a long time. However, the latest reference has found spawning sites by using genetic identification of collected preleptocephali in the Pacific subtropical gyres. The migration route of Conger eel showed to be similar to that of Japanese eel, *Anguilla japonica*, as both of them are transported with same currents (Tsukamoto, 2006; Miller et al., 2011).

Transported conger eel larvae have metamorphic stage, which are finished in a river estuary, near shore or coastal waters. After metamorphosis juveniles started strictly marine benthic life on the rocky and sandy bottoms (Lee and Byun, 1996; Wang, 2000; Kimura et al., 2004; Yagi et al., 2010). This species is known as carnivores of vigorous appetite that exploited the available prey such as fishes, shrimp, crabs and cephalopods (Huh and Kwak, 1998; Choi et al., 2008).

The white-spotted conger eel is one of the most important commercial demersal fish in the Korea, which also has an ecological importance in the community of coastal waters. These commercial fish species is very important for fisheries market and industry. Therefore, it should be intensive managed for sustainable supply. In order to manage of *C. myriaster* stock, it is necessary to carry out a research of population dynamics. As a first step, analysis of otolith is one of the most popular methods to determine age and growth for the fisheries management. The most commonly used ageing technique is to count the annuli in the hard part of fishes, such as sagittal otolith, scale, vertebrae, etc. The otolith is used extensively in population studies to determine age, growth rate, mortality and length at age relationship. However, it is useful only when age validation can be accurate. Conventional methods of age determination are based on the estimation of growth increments in sagittal otolith and annuli counted. In general, the sagittal otolith from fish is directly removed to determine the age and growth pattern, count of the annual growth increments. It is difficult for the directed observation to count the annuli of the older fish of numerous anguilliform fishes in the tropic and temperate regions (Kubota, 1961; Hood et al., 1988; Sullivan et al., 2003; Gorie et al., 2004). Because, multiple banding of the opaque band was formed when appear with alternating translucent and opaque zones from the nucleus. In recent years, development of new techniques and methods (such as the EDTA etching method, three dimension image analysis, Burnt and UV light observation and the morphological measurement of otolith) have been used for more accurate age determination. In addition, these are contributed to cost, effort and time saving. Various methods have been proposed, but size and shape of otoliths vary by species. Therefore, the otolith processing method should be developed on a species-specific basis. UV light observation was initially used to determine the age of Conger eel in Japan (Katayama et al., 2002).

Currently, this method is the most useful way of the age validation of Conger eels. Previous studies indicated that there are some relationship between the otolith weight and the estimated age. Fish age could be better estimated from otolith measurements than fish length, as in a number of temperate species (Boehlert, 1985; Pawson, 1990; Fletcher 1991; Wilson et al., 1991; Cardinale et al., 2000; Francis et al., 2005; Lou et al., 2005). Several studies has showed that the relationship between the otolith weight and age are best suited for age determination (Worthington et al., 1995; Newman et al., 2000; Francis and Campana, 2004; Lou et al., 2005). This can be applied for determining the age structures predicted from otolith weights to estimate key population parameters relevant to fisheries assessment and management in the white-spotted conger eel.

Traditionally growth studies have been used the von Bertalanffy growth model to determine average growth curves for populations. Unfortunately, age and growth of the white-spotted conger eel was examined in a few studies (Katayama et al., 2002; Gorie et al., 2004). However, age determination of other conger eels has been studied in many regions. The von Bertalanffy growth functions have been estimated for the European conger eel caught in the east coast of Ireland, Irish coastal waters and Atlantic Iberian waters (Fannon et al., 1990; Sullivan et al., 2003; Correia et al., 2009). These studies showed that the maximum age of the European conger eel was around 12 to 20 years old. The previous reports suggested a possibility that lifespan of same genus could be similar to age for the other species.

Our hypothesis from age determination have approached that the lifespan is more than decade for *C. myriaster*.

Reproductive characters and strategies of Conger eels have a wide range of migration, and the maturation is occurred only one time of during a lifespan (Kurogi et al., 2002; Bell et al., 2003; Utoh et al., 2003; Kimura et al., 2004; Katayama and Shimizu, 2006; Miller et al., 2011). It is the common feature of several eels (Tsukamoto, 1992 and 2006; McCleave and Miller, 1994; Correia et al., 2002). Utoh *et al*. (2003) investigated the process and characteristics of oogenesis in the white-spotted conger eel. They examined between the oocyte diameter and gonadosomatic index (GSI) that was related to oocyte development.

In this study, the oogenesis of white-spotted conger eel was estimated to relate oocyte diameter and GSI value. It was referenced by Horie *et al*. (2002) and Utoh *et al*. (2003).

We examined the otolith record of age and growth of the white-spotted conger eel, to provide a better understanding of the otolith reading and more accuracy of the age validation using by UV light. The age structure of population of conger eel in the study area was estimated from relationship between the otolith measurements and age validation. We also investigated the relationship between oocyte diameter and GSI to reveal the initiation time of the oogenesis. This paper presents the first detailed information on the population dynamics of the white-spotted conger eel, *C. myriaster* in Southern Sea of Korea.

2. Materials and Methods

2-1. Sample collection

A total of 635 conger eel were sampled from the southern of Korean waters (Fig. 1). Fresh samples were collected from fishermen during the period between April 2009 and November 2011. One hundred thirty five specimens were taken by trap and shrimp beam trawl in coastal water of the Geoje Island (10 – 15m depth). Another 500 specimens were captured by bottom trawl at depths around 100m offshore the Southern Sea of Korea.

Total length (cm), whole body weight (g), gutted weight (g), gonad weight (g) and liver weight (g) were measured for all specimens. Sagittal otoliths were removed from the cranial cavity, cleaned from adherent tissues and stored dry in labeled box. All undamaged sagittal otoliths were measured for length to the nearest 0.01mm under the stereoscopic microscope (Carl zeiss Discovery v.8). Otoliths were weighed to the nearest 0.0001g using the electronic balance (Sartorius CPA224S). The weights of otoliths were statistically tested to detect the significant difference between left and right otolith of individual Conger eel, *C. myriaster*. Furthermore, morphological characters such as length, width and radius of otolith were measured under the microscope equipped with the image analyzer.

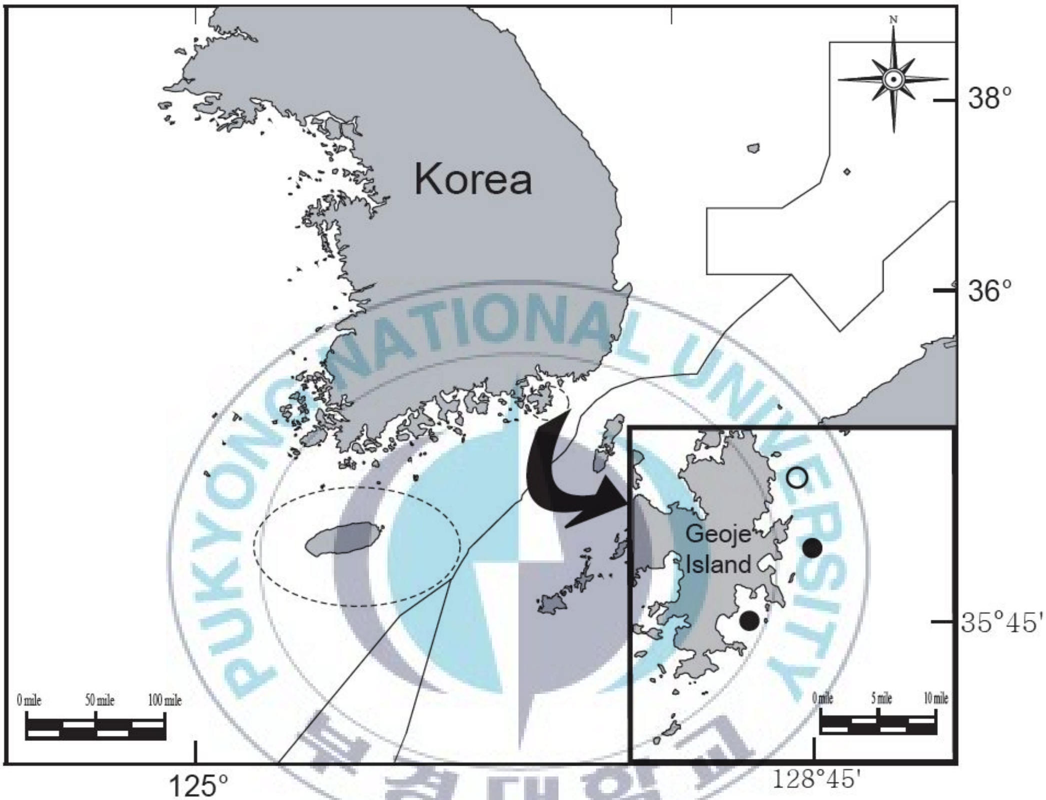


Fig. 1. Sampling sites of *Conger myriaster* in the Southern Sea of Korea (A dotted line circle indicates the location of sampling by the bottom trawl, open circle indicates sampling by the shrimp beam trawl and solid circle indicates sampling by the trap).

2-2. Age determination and growth curves

The sagittal otoliths of white-spotted conger can be used to determine age. Katayama *et al.* (2002) examined by observing burnt sagittal otolith under UV light. In our study, we have adapted this technique and more developmental observation to our specimens. The right otoliths of 635 specimens were separated for age determination. Sagittal otoliths were removed from each fish, cleaned, dried, and stored in tube until further observation. The prepared otoliths were embedded in the epoxy resin and serially sectioned along the longest axis across the core at about 0.3mm intervals with a diamond saw. The polishing was tried using #2000-grit grinding paper, after the samples were examined under the stereo microscope that magnification from 10 to 60×. The UV light at strong wavelength can be used for several industries (such as drying, curing, etc.). Using characters of sagittal otolith burned by drying oven and UV light makes observation easier between opaque and translucent zone. The illumination wave length was used on the 380 ~ 420㎚ (Lichtzen Inno-cure 100N). Growth annuli observed from each otolith have been counted on three times, with an interval of about four months between counts by researcher. The sagittal otoliths with multiple banding patterns were rejected.

Marginal increment ratio (MIR) was used to validate the annual growth pattern increment deposition that is calculated by the equation:

$$\mathrm{MIR} = \frac{R - r_n}{r_n - r_{n-1}},$$

where R is the radius of otolith, r_n is the length of the otolith radius at the time of the nth opaque zone mark. MIR was expressed percentage of otoliths with opaque and translucent margins were plotted by monthly capture.

The following equation was used to relate fish total length (TL) at capture to the otolith radius (OR) at capture: $TL = a + b\ OR$, where a is the intercept and b is the slope from the linear regression. The Frazer-Lee's back-calculation, TL of each specimen at the time of formation of each opaque zone was determined by substituting the measurement to each increment based on the body proportional hypothesis. The relationship between fish age and otolith weight (*OWt*) was estimated to be linear and both the *OWt* and age data were found to conform to assumptions of normality and homogeneous variances among groups (Lou et al., 2007). The following equation is calculated: Fish age= $a + b\ OWt$, where b is the slope of the regression and a the constant. Otoliths which are not used for age validation were estimated to age from the relationship of age and OWt regression.

The von Bertalanffy growth function (VBGF) was fitted to individual length and age data for the conger eel population. VBGF approached with three-ways of equation. Three methods of growth function were calculated by back-calculation, age at mean length data and age at otolith weight regression method. The growth parameters were estimated from three-methods by a non-linear regression, using a software program (SYSTAT 12). The relationship is expressed by the following equation:

$$L_t = L_\infty\{1 - \exp[-K(t - t_0)]\},$$

where L_t is the length at age t, L_∞ is the asymptotic length predicted by the equation, K is the growth coefficient that determines the rate at which L_t approaches L_∞, and t_0 is the hypothetical starting time at zero length if growth follows that predicted by the equation. The length-weight relationship for the population was determined by the power function regression. The weight growth curve makes which can be transfer from length at growth curve.

The estimates of L_∞ and K were used for comparison of growth performance indices (φ') (Pauly & Munro, 1984) among three-ways using the equation: $\varphi' = 2\log_{10}(L_\infty) + \log_{10}(K)$, compare with the three methods of growth parameters by likelihood ratio test.

2-3. Indices of fish condition

The relationship between the weight and length was estimated by power functional regression. As following: $BW = aTL^b$, where a and b are constant, BW is the wet weight (g) and TL the length (cm). A specific weight-length relationship was established for the total sample (n = 635). The condition factor (Kn) was calculated for each individual females using the equation: Kn= BW/aTL^b. The gonadosomatic index (GSI) was calculated as GSI= $(W_g/BW)\times 100$, where W_g is the gonad weight and the BW is the specimen gutted weight. In addition, Hepatosomatic index (HSI) was calculated as HSI= $(W_l/BW)\times 100$, where W_l is the liver weight. This indicator was related to fatness by the lipid accumulation in fish.

2-4. Gonadal development

The juvenile eels were not possible to distinguish the sex and sexual development stage. We examined the sex from the direct visual observation and under the microscope. We separated to all specimens that were divided the female or male (contained indeterminate sex).

Gonadal development of female was estimated from GSI and mean maximum oocyte diameter. Utoh *et al*. (2003) investigated the process and characteristics of oogenesis in the white-spotted conger eel. It indicates that oogenesis is related to increase GSI with oocyte development. We have adapted their result that is referenced. The maximum oocyte diameter (MOD) was calculated as the mean of the 10 oocyte per section (Hood et al., 1988; Utoh et al., 2003).

2-5. Statistical analysis

Differences between left and right otoliths were tested by paired *t*-test ANCOVA was used to test for differences in otolith measurements at total length between two sampling sites, considering fish length as a covariate (Minitab ver.16). Results are presented as mean±standard errors. Significant differences were carried out using one-way ANOVA and then, if there was a significant different between them, Tukey HSD test was performed (SPSS 12.0.1). If the variance of the data was not homogeneous, a nonparametric data were tested by Mann-Whitney *U* test or Kruskal-Wallis test (Minitab ver.16). The differences in the size-frequency distributions between the two sexes were tested by the Kolmogorov-Smirnov two-sample test. The growth curves were compared by the method of Kimura's likelihood ratio test.

3. Results

3-1. Length and weight

The samples of white-spotted conger ranged from 15.6 to 107.5cm in length and from 4.8 to 2,572.7g in weight (Fig. 2). Of 635 individuals, 528 (83.2%) were females, 107 (16.8%) were immature males or indeterminate individuals. Females were observed mainly in the large length classes (Fig. 3). The Kolmogorov-Smirnov two-sample test showed that there was a significant difference between female and male (contained unsexed) length-frequency distributions ($Z=1.88$, $P<0.01$). There was a significant difference in lengths between the two sampling locations (Mann-Whitney U test, $P<0.05$) (Table 1).

Table 1. Length and weight for sample size, location, period and fishing gear for *Conger myriaster* (*n* =635)

Sampling location	Gear	Year	Month	Sample size (*n*)	TL (cm)			BW (g)		
					Minimum	Mean±S.E	Maximum	Minimum	Mean±S.E	Maximum
Coastal water in Geoje Island	Trap	2009	April	31	25.7	40.6±1.2	57.2	35.0	128.7±13.7	351.7
			June	6	37.7	42.5±3.1	57.0	76.8	127.9±34.0	289.1
			August	16	31.3	39.0±1.5	51.0	56.9	105.9±13.8	234.6
			November	41	33.5	41.1±1.1	72.7	55.1	135.1±17.6	760.5
	Shrimp beam trawl	2010	November	16	16.7	19.1±0.3	22.5	5.8	8.4±0.4	13.6
		2011	April	5	18.0	20.5±0.9	22.2	5.7	9.2±1.2	11.8
			November	20	15.6	19.7±0.9	31.6	4.8	11.3±2.0	40.1

Table 1. Continued.

Sampling location	Gear	Year	Month	Sample size (*n*)	TL (cm)			BW (g)		
					Minimum	Mean±S.E	Maximum	Minimum	Mean±S.E	Maximum
Offshore in the Southern Sea of Korea	Bottom trawl	2009	August	53	40.7	56.5±1.9	100.1	100.4	373.5±56.7	2026.4
			September	79	44.6	58.5±1.1	89.0	132.2	339.6±24.5	1238.3
			October	21	77.2	87.8±1.8	106.3	732.3	1402.4±107.4	2572.7
			November	23	60.9	78.3±2.5	101.6	368.8	985.2±119.8	2336.8
			December	52	48.0	62.7±1.4	94.9	175.5	483.6±44.6	1945.0
		2010	January	25	67.6	82.6±2.0	107.5	633.7	1144.6±99.8	2554.7
			February	27	49.2	66.6±2.0	93.3	189.5	669.4±93.0	2526.9
			March	35	53.6	72.1±2.3	103.8	209.1	792.2±97.7	2543.1
			April	50	42.2	54.5±1.3	101.1	113.4	295.5±41.6	2170.0
			May	79	43.6	55.4±0.7	75.1	129.8	278.0±12.5	699.6
			June	28	41.9	56.8±1.4	69.6	117.6	329.6±24.1	561.7
			July	28	36.0	69.0±2.5	104.5	65.5	638.6±79.4	2182.1

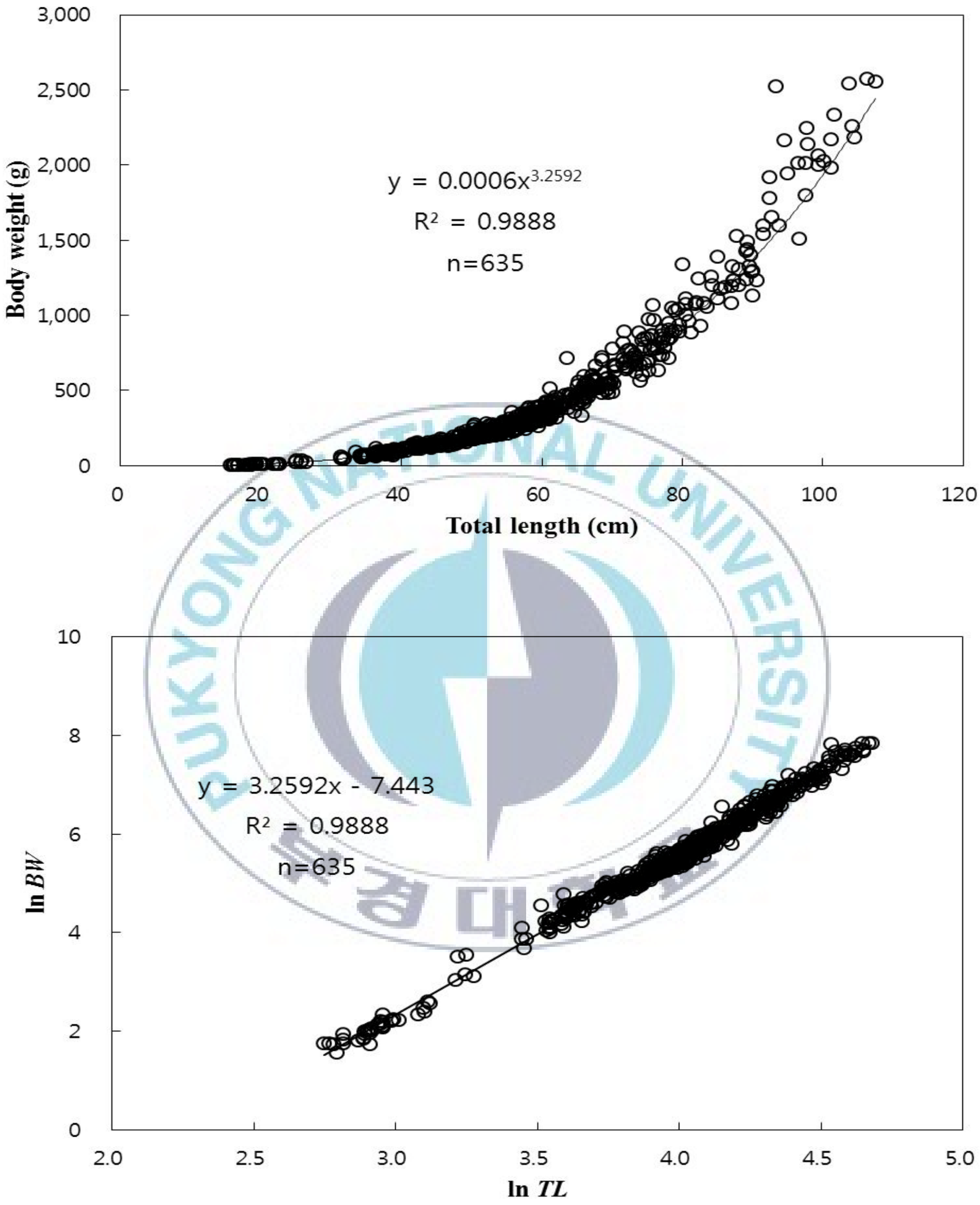


Fig. 2. Relationship between body weight (BW) and total length (TL) of *Conger myriaster* collected in the Southern Sea of Korea (upper graph showed raw data, lower graph showed transferred data).

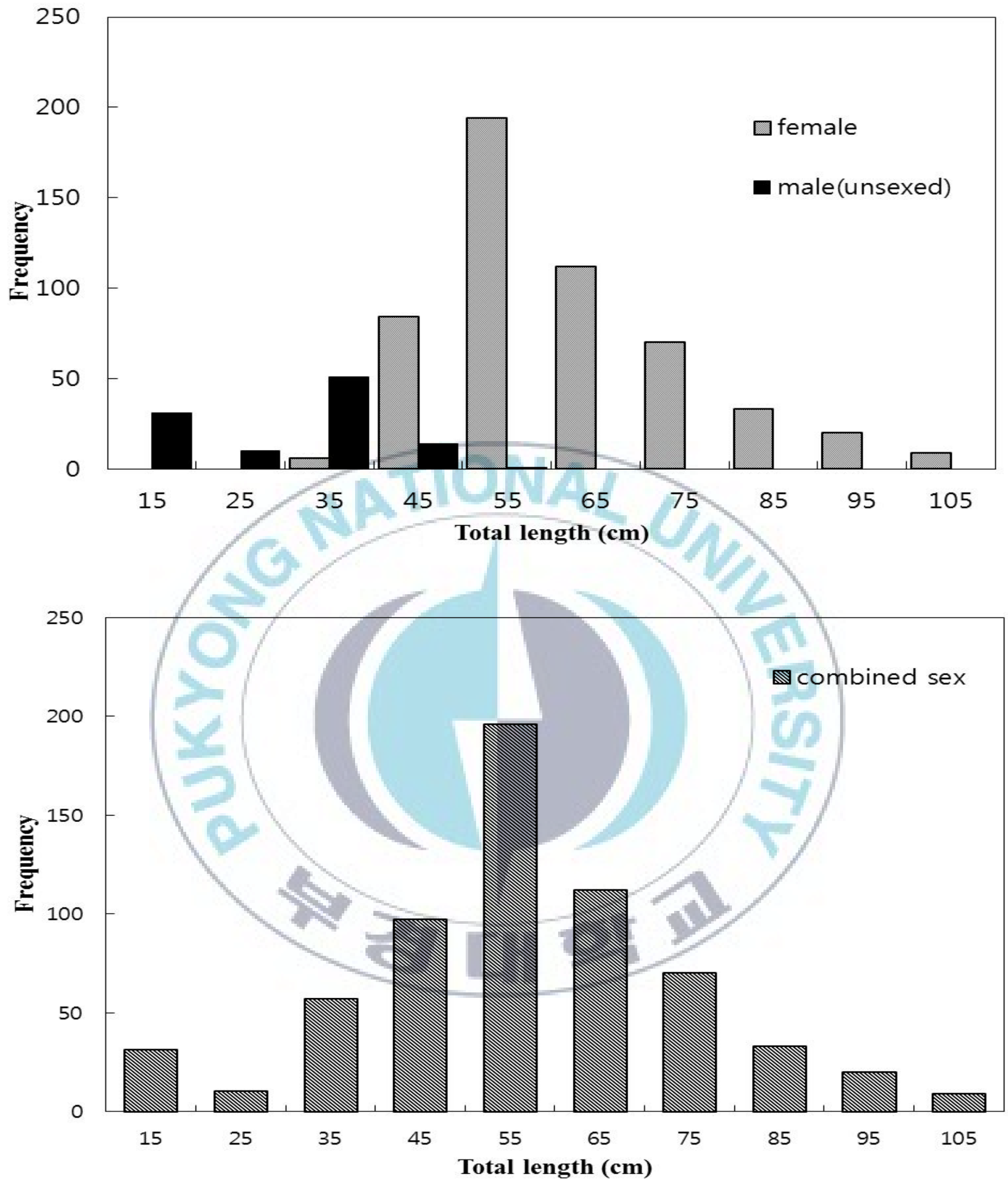
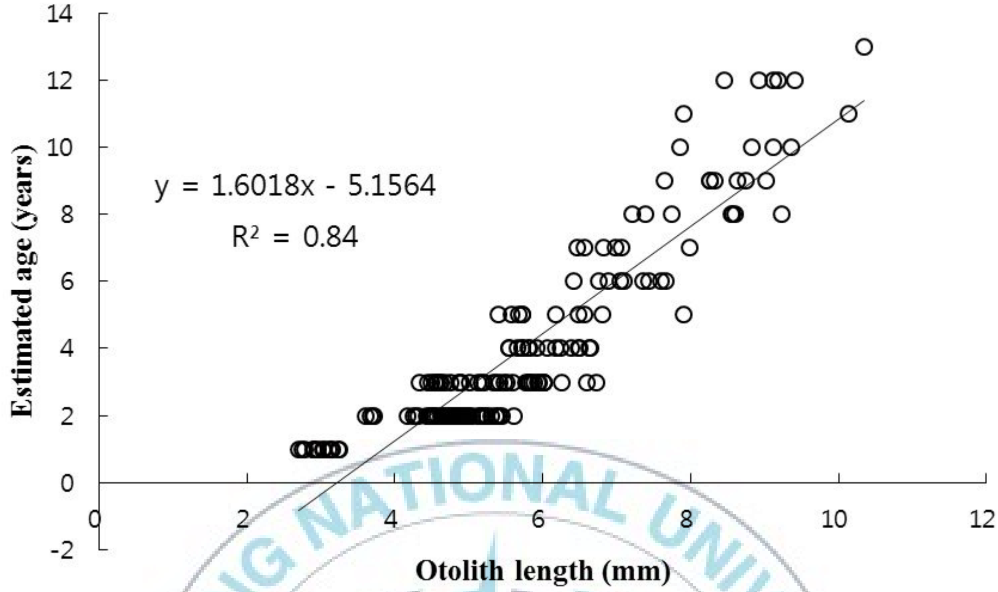


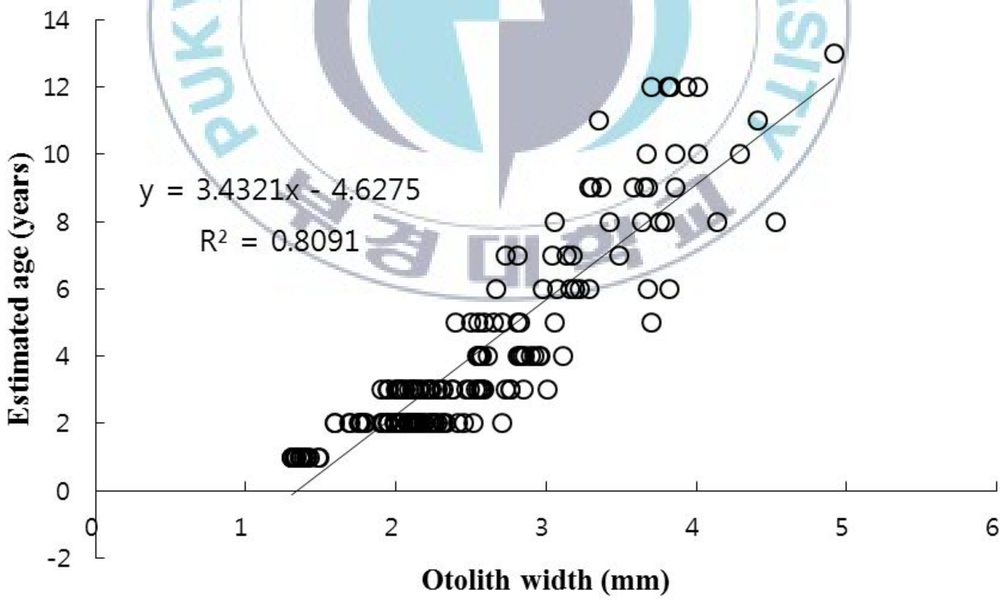
Fig. 3. Frequency distributions of total length (cm) for female and male (upper graph), combined sex (lower graph) of *Conger myriaster* collected in the Southern Sea of Korea.

3-2. Morphological characters of otolith

The otolith of Conger eel was long-elliptic shape and the distal surface was concave (Fig. 4). The anterior was longer because the location of primordium was skewed to the posterior. No significant differences in otolith weight was found between left and right otoliths (paired *t*-test, t=-1.04, P=0.297). The relationships between age and otolith characters (otolith length, width, radius and weight at the validated age) were examined (Fig. 5). All of the regressions were positive linear relationships with age. Fig. 5 showed that each groups of age tend to the overlap with the variables. Of the four regressions, the relationship between the otolith weight and the validated age was best fitted. Therefore, it is used to estimate age composition in population.

Fig. 4. Sagittal whole otolith of *Conger myriaster* (TL: 81.2cm, BW: 884.2g), scale bar=1mm.


y = 1.6018x - 5.1564
R² = 0.84
Estimated age (years)
Otolith length (mm)
14
12
10
8
6
4
2
0
-2
0
2
4
6
8
10
12


y = 3.4321x - 4.6275
R² = 0.8091
Estimated age (years)
Otolith width (mm)
14
12
10
8
6
4
2
0
-2
0
1
2
3
4
5
6

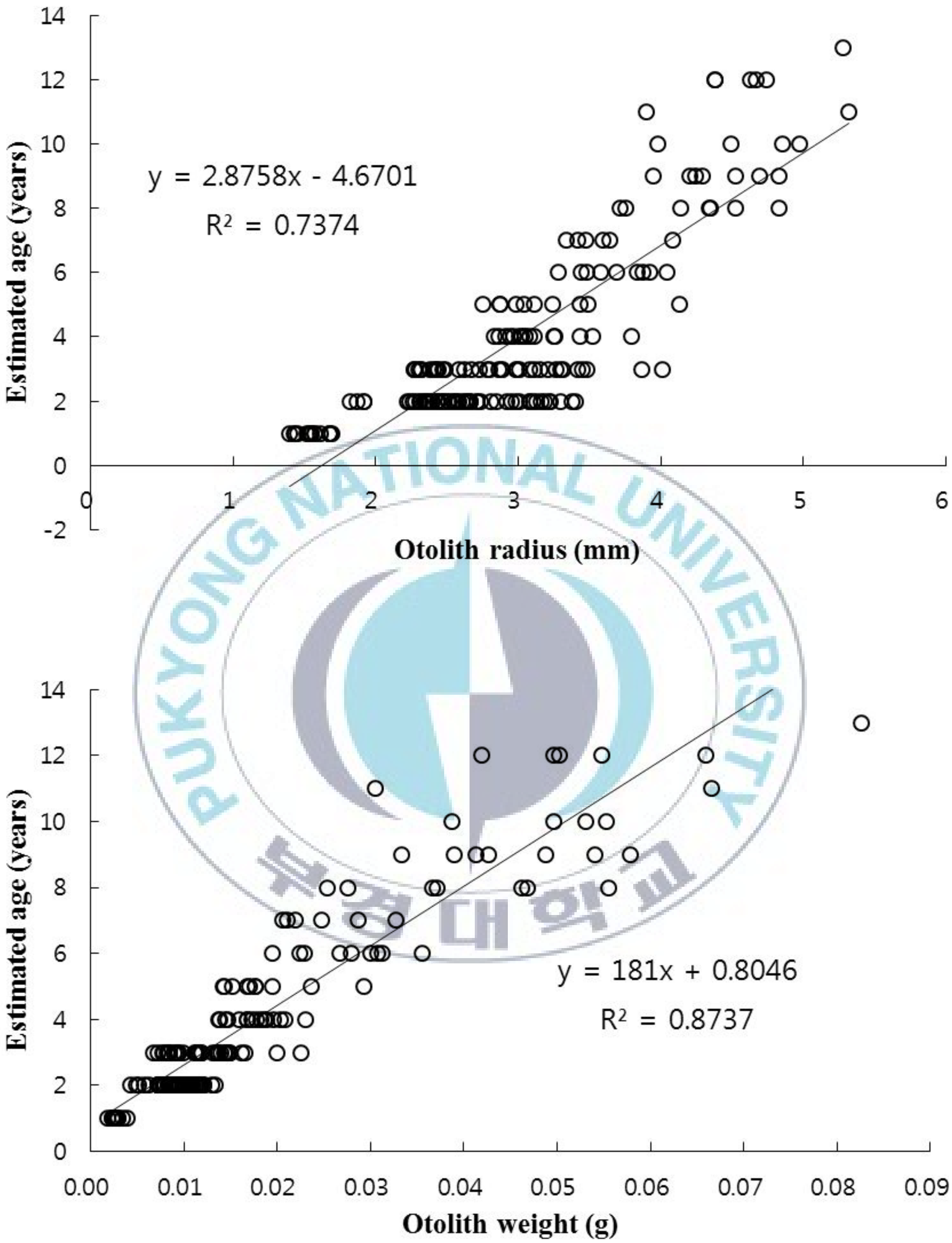


Fig. 5. Relationship between estimated age and otolith measurements (length, width, radius and weight) of *Conger myriaster*.

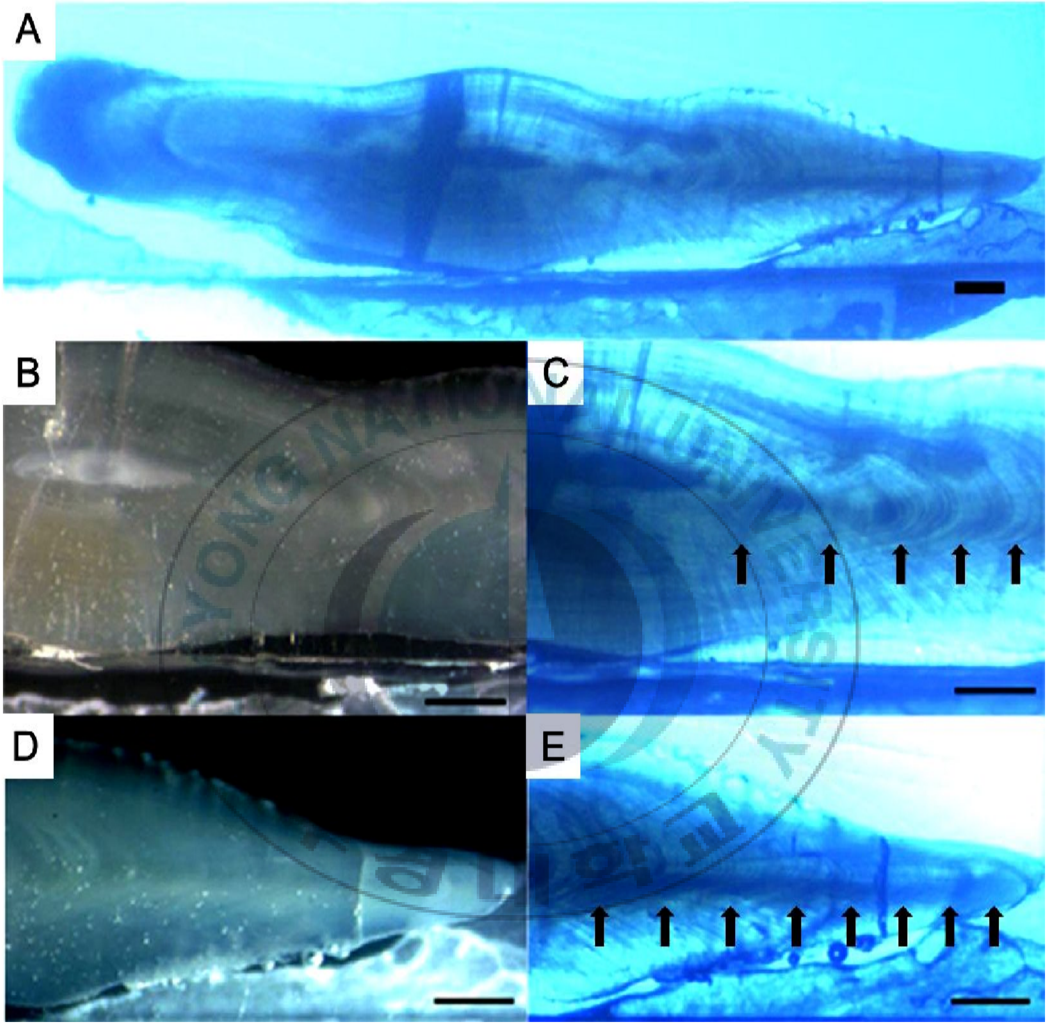


Fig. 6. Transverse otolith section of *Conger myriaster* of 13 years old (Feb. 2010 TL: 93.3 cm) using UV (A, C, E) and normal light (B, D) observation. The solid arrows indicate annual marks. Scale bar= 500㎛.

3-3. Age validation and composition

Age determination was progressed by a new ageing technique using UV light. The burnt otolith under transmitted UV light is showed in Fig. 6. The opaque zones are clear and obvious to observe. Therefore, it made larger individuals determine age easily. Ages were determined from 216 right otoliths among 635 otoliths examined. Age of the conger eel examined ranged from 1 to 13 years. On the assumption that *C. myriaster* hatch on 1 December (Lee and Byun, 1996), we estimated the year class by counting annuli. Of the 635 otoliths were conducted by morphological measurement.

Marginal increment ratio (MIR) was used to validate annual pattern in the deposition of opaque zones in the otolith. Fig. 7 showed the seasonal change in the proportions of annulus (opaque and translucent zone). It was described at time of monthly of annual. MIR increased from spring to summer and decreased in autumn, indicating that there is a single annual band of deposition per year.

The relationship between the age and otolith weight data (*OWt*) was analyzed. Overall, the mean otolilth weight almost invariably increased with age, although there was a considerable variation in *OWt* both within and between age groups (Fig. 5). The relationship was determined by the linear regression: *Age*=

181*OWt*+0.8046 (r^2=0.8737, P<0.05). Relationship between *OWt* and observed age are presented in Fig. 8.

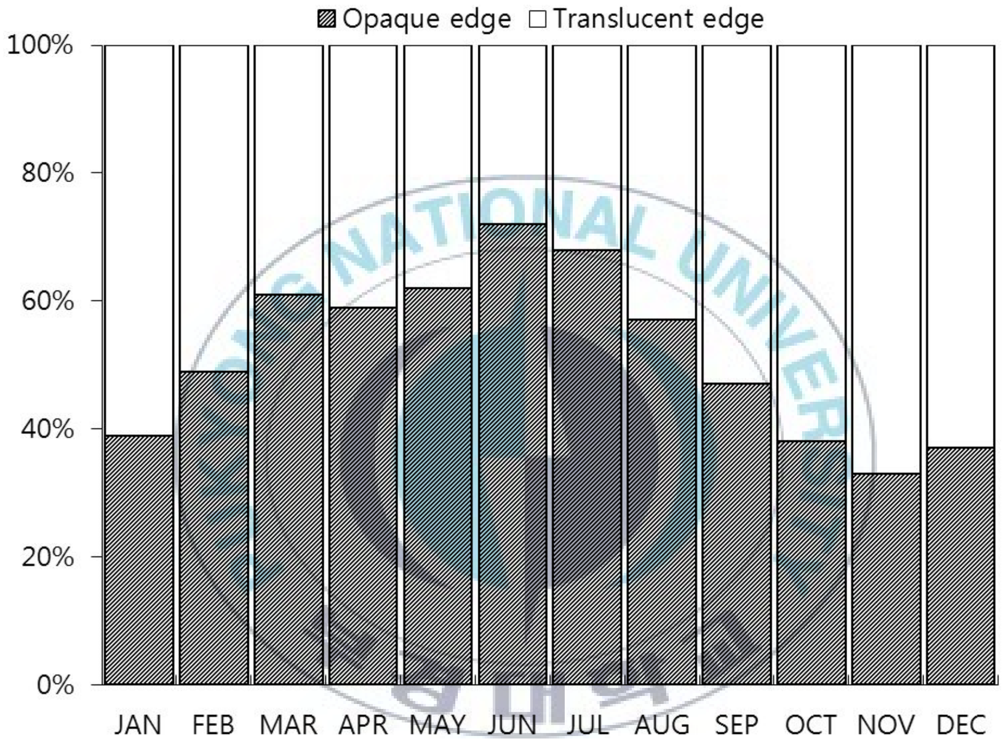


Fig. 7. Seasonal change in the proportion of each otolith marginal structure considered for *Conger myriaster*.

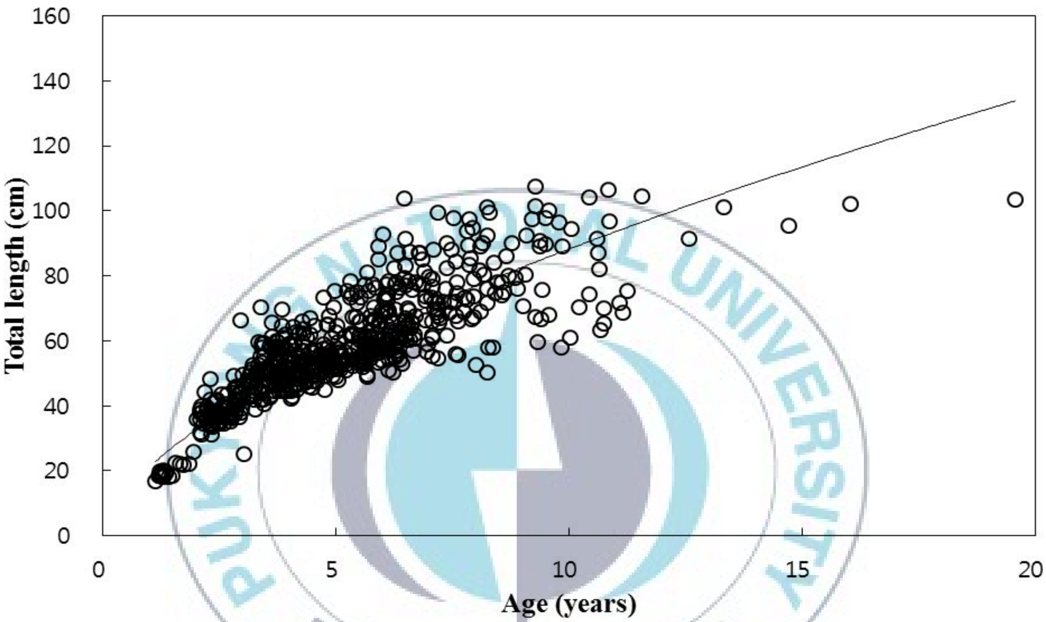


Fig. 8. Relationship between the estimated age and total length (cm) from the OWt-age equation of *Conger myriaster*.

3-4. Comparison of population growth

The growth parameters were estimated using three methods. Firstly, age was determined by using a back-calculation method. There was a linear relationship between *TL* and *OR* (Fig. 9): $TL = 20.162OR - 11.72$ (r^2=0.72, n=635). The total length of each fish was back-calculated to the time of formation of the *n*th annulus from each radius using the BPH method (Francis, 1990). The weighted mean back-calculated total length of each successive annulus showed accurate estimation for total length at age (Table 2).

The von Bertalanffy growth parameters were estimated from the back-calculation of annuli at otoliths by the non-linear regression model. On the other hand, we used that mean total length of collected samples at age. The parameters were estimated from previous procedure. Finally, we have shown a variance was estimated from otolith weight at age (Table 3). In the growth curves no significant differences between the back-calculation, mean length at age and otolith weight at age method was found (Likelihood ratio test, F=0.11, df=2, P=0.956).

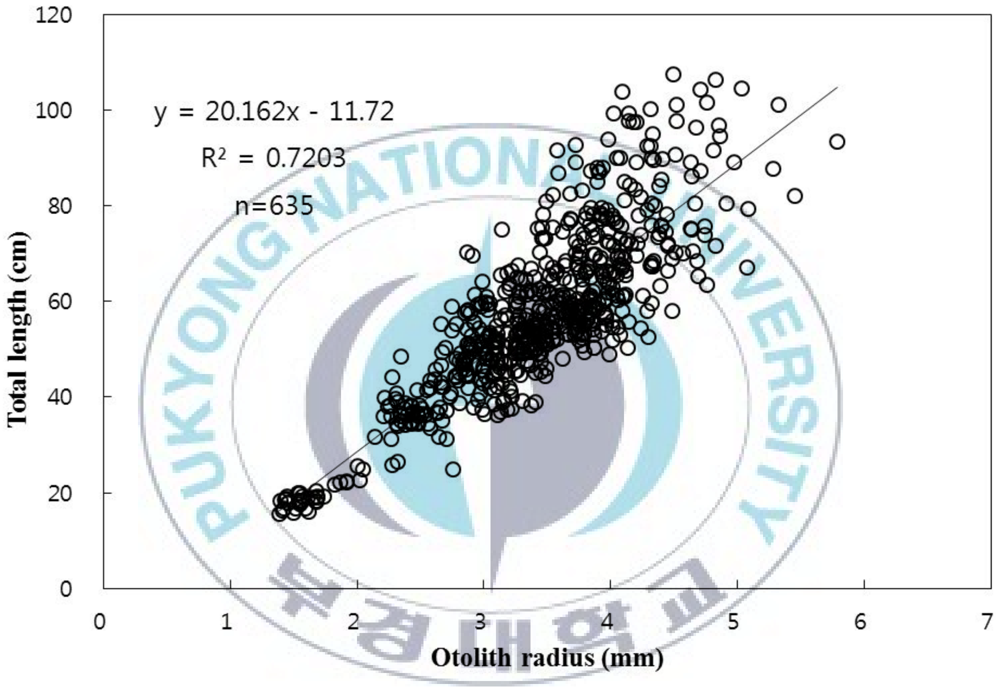


Fig. 9. Relationship between total length (cm) and otolith radius (mm) of *Conger myriaster*.

Table 2. Mean back-calculated total length (cm) for each ring group of *Conger myriaster*

Age (year)	Age validation (n)	Mean TL±S.E (cm)	Mean length of annulus (mm)													
			R	r^1	r^2	r^3	r^4	r^5	r^6	r^7	r^8	r^9	r^{10}	r^{11}	r^{12}	r^{13}
1	32	18.8±0.9	1.55	1.32												
2	33	35.8±5.7	2.37	1.28	2.15											
3	38	44.5±7.7	2.75	1.25	2.16	2.63										
4	48	52.8±6.2	3.18	1.22	2.11	2.65	3.10									
5	15	56.5±7.3	3.58	1.26	2.02	2.66	3.12	3.48								
6	12	63.2±9.1	3.89	1.25	1.91	2.58	3.02	3.44	3.81							
7	8	71.9±10.7	4.22	1.21	1.88	2.54	3.03	3.39	3.79	4.12						
8	7	76.9±13.5	4.52	1.18	1.82	2.49	2.98	3.41	3.82	4.13	4.39					
9	6	81.6±12.1	4.77	1.17	1.79	2.48	2.86	3.37	3.76	4.08	4.42	4.67				
10	6	85.5±16.3	5.08	1.15	1.77	2.39	2.88	3.36	3.72	4.00	4.42	4.73	5.01			
11	3	87.1±14.1	5.22	1.11	1.78	2.41	2.84	3.32	3.70	3.91	4.45	4.83	4.99	5.11		
12	5	90.7±17.9	5.28	1.10	1.72	2.39	2.77	3.31	3.71	3.96	4.29	4.61	4.83	5.08	5.22	
13	3	94.5±10.1	5.33	1.06	1.81	2.42	2.81	3.34	3.69	3.92	4.30	4.59	4.82	5.05	5.11	5.23
Total number	216	Weighted mean		1.20	1.91	2.51	2.94	3.38	3.75	4.02	4.38	4.69	4.91	5.08	5.17	5.23

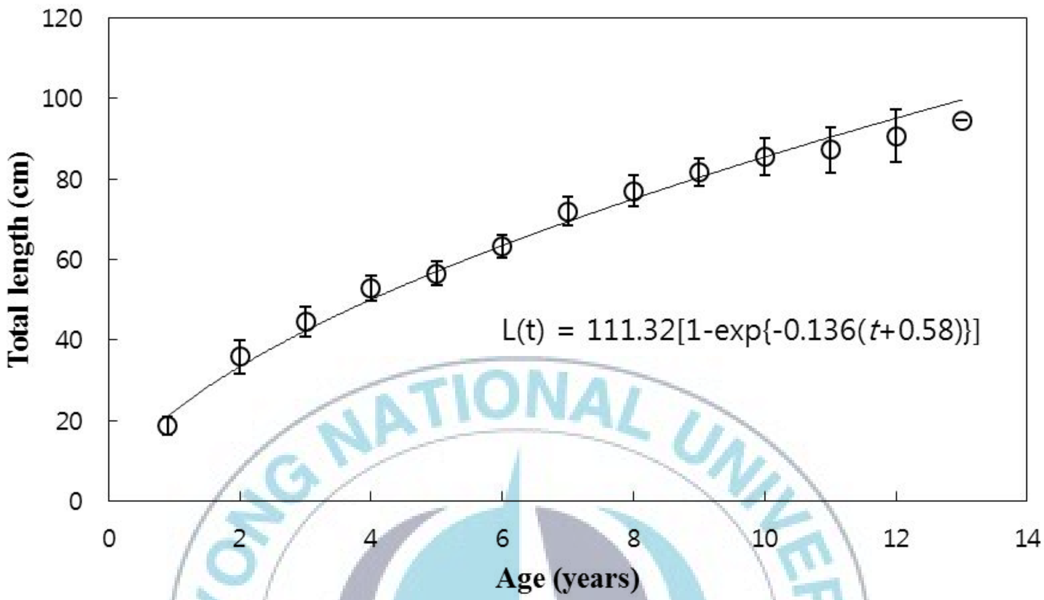
Table 2. Continued

Age (year)	Age validation (n)	Mean back-calculated total length (cm)												
		TL[1]	TL[2]	TL[3]	TL[4]	TL[5]	TL[6]	TL[7]	TL[8]	TL[9]	TL[10]	TL[11]	TL[12]	TL[13]
1	32	14.9												
2	33	14.1	31.6											
3	38	13.5	31.8	41.3										
4	48	12.9	30.8	41.7	50.8									
5	15	13.7	29.0	41.9	51.2	58.4								
6	12	13.5	26.8	40.3	49.2	57.6	65.1							
7	8	12.7	26.2	39.5	49.4	56.6	64.7	71.3						
8	7	12.1	25.0	38.5	48.4	57.0	65.3	71.5	76.8					
9	6	11.9	24.4	38.3	45.9	56.2	64.1	70.5	77.4	82.4				
10	6	11.5	24.0	36.5	46.3	56.0	63.3	68.9	77.4	83.6	89.3			
11	3	10.7	24.2	36.9	45.5	55.2	62.9	67.1	78.0	85.7	88.9	91.3		
12	5	10.5	23.0	36.5	44.1	55.0	63.1	68.1	74.8	81.2	85.7	90.7	93.5	
13	3	9.7	24.8	37.1	44.9	55.6	62.7	67.3	75.0	80.8	85.5	90.1	91.3	93.7
Weighted mean	216	13.3	29.5	40.5	49.3	57.0	64.2	69.8	76.6	82.7	87.5	90.7	92.7	93.7

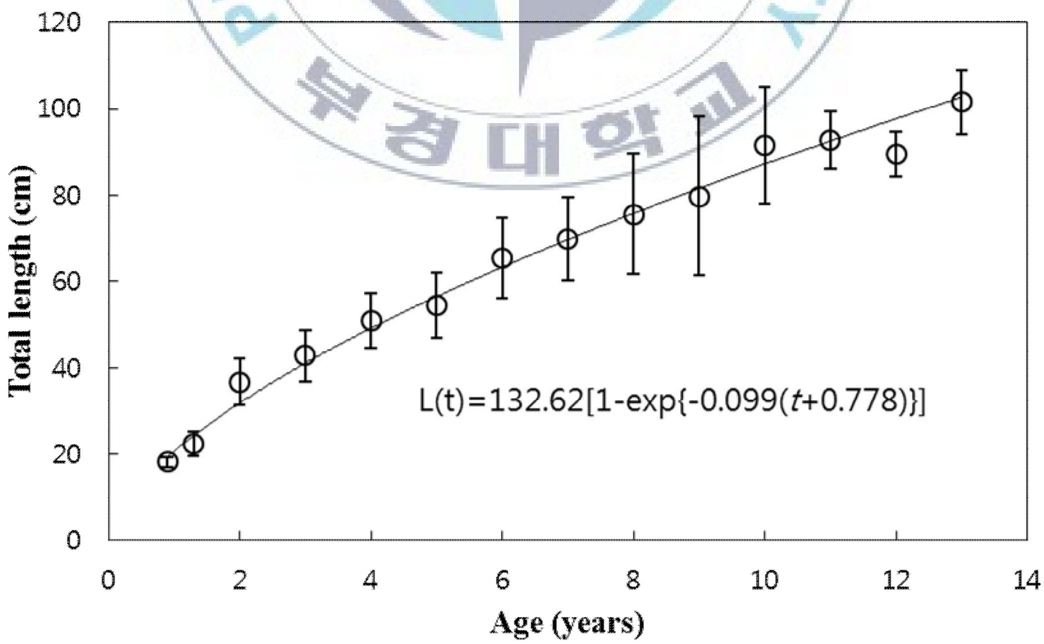
Table 3. Comparison of von Bertalanffy growth parameters from the estimated age models of *Conger myriaster*

Method	Parameters			
	L_∞	K	t_0	φ'
Back-Calculation	111.32cm	0.136	-0.58	3.23
Mean length at Age	132.62cm	0.099	-0.778	3.24
Otolith weight at Age	143.76cm	0.081	-1.285	3.22

Back-Calculation



Mean length at Age



Otolith weight at Age

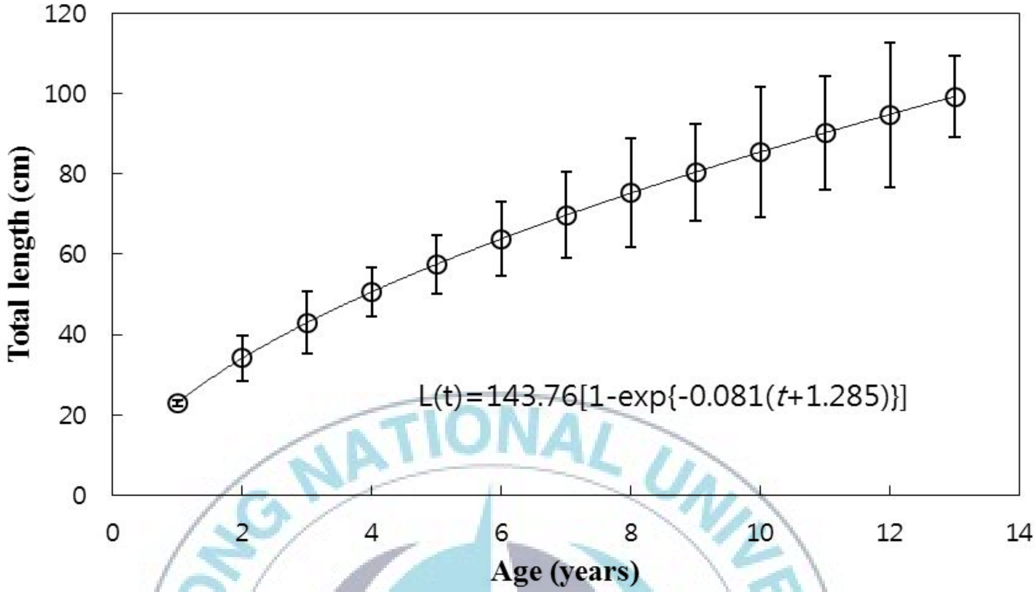


Fig. 10. Age-total length relationships with von Bertalanffy growth curves for three methods of *Conger myriaster*.

3-5. Condition factors

The condition factor (Kn) ranged from 0.67 to 1.63 for the conger eels (Fig. 11). There were significant mean differences in Kn among seasons (One-way ANOVA, $F_{499,\ 3}$=2.622, P<0.05), namely between summer and winter (Tukey HSD test, P<0.05). The hepatosomatic index (HSI) ranged from 0.50 to 3.80. There were no significant mean differences among seasons (One-way ANOVA, P=0.07). The gonadosomatic index (GSI) ranged from 0.06 to 8.18 and varied significantly among seasons (One-way ANOVA, $F_{499,\ 3}$=24.718, P<0.05), namely between summer and autumn (Tukey HSD test, P <0.05).

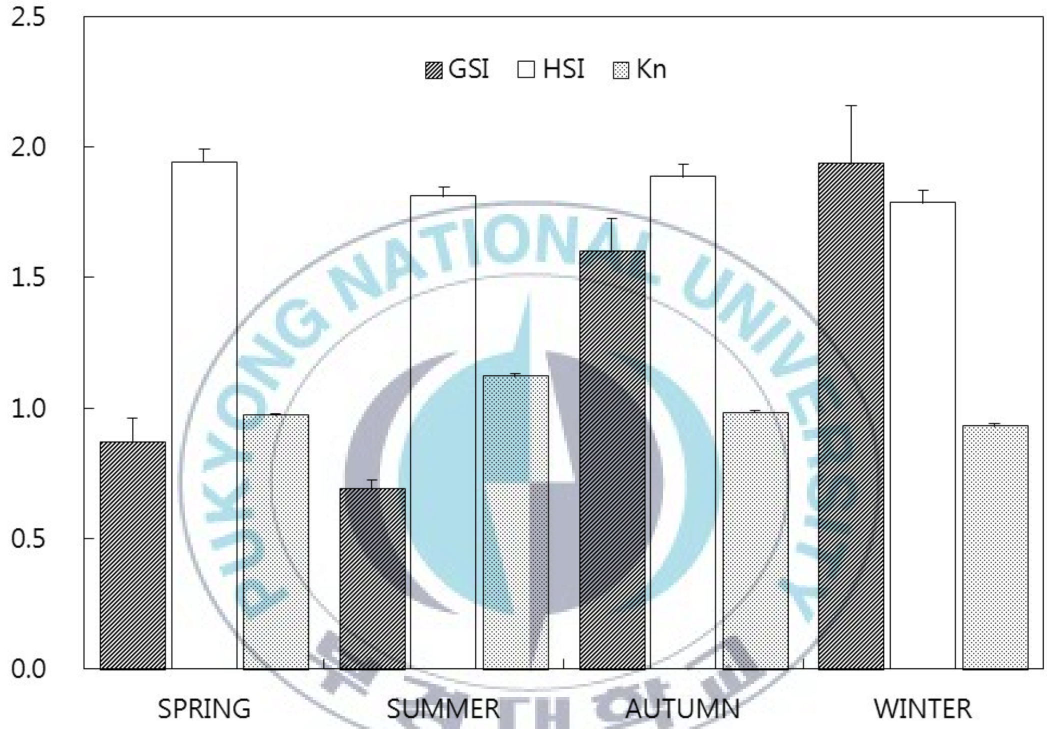


Fig. 11. Seasonal variation of the relative condition factor (Kn), the hepatosomatic index (HSI) and the gonadosomatic index (GSI) for the females of *Conger myriaster* collected in the Southern Sea of Korea.

3-6. Reproductive traits of ovaries

The gonads are paired organs that are attached to the peritoneum on either side of the swim bladder and longitudinal stretches of the body cavity. The gonads were observed line-like structure of translucent white color in the juvenile eels. The larger eels were emerged ribbon or curtain-like structure of ivory (sometimes pinkish) in color. Females were either immature or developing stage which did not appear to be sexual mature specimen. The ovaries contained oocytes in various stages of development. Fig. 12 showed positive correlation between the mean maximum oocyte diameter (MOD) and GSI.

GSI was more suitable with the power function from MOD. We observed female conger eels of about 75 specimens that total length and body weight ranged from 44.7 to 107.5cm and 130.2 to 2,554.7g, respectively. The mean maximum oocyte diameters ranged approximately from 50 to 430 ㎛ (average =201.4±13.1) under the stereoscopic microscope in fresh samples. We would like to consider the start time of the oogenesis that was estimated TL 45cm and the age from 4 years old (Fig. 13).

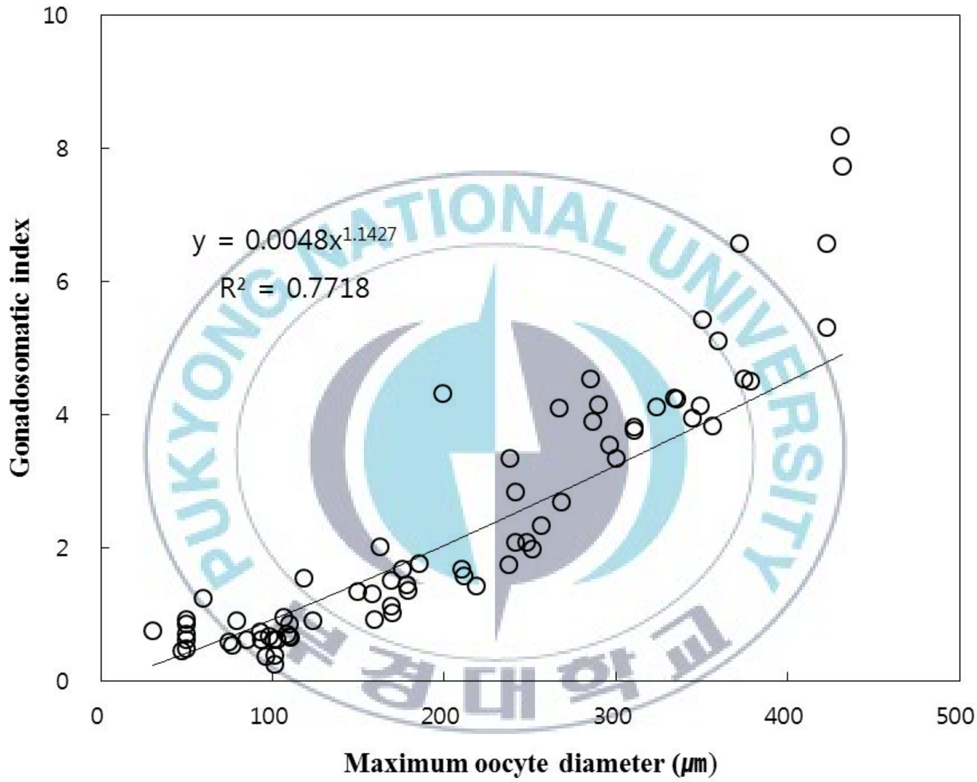


Fig. 12. Relationship between gonadosomatic index (GSI) and the maximum oocyte diameter (MOD) in females of *Conger myriaster* (*n*=75).

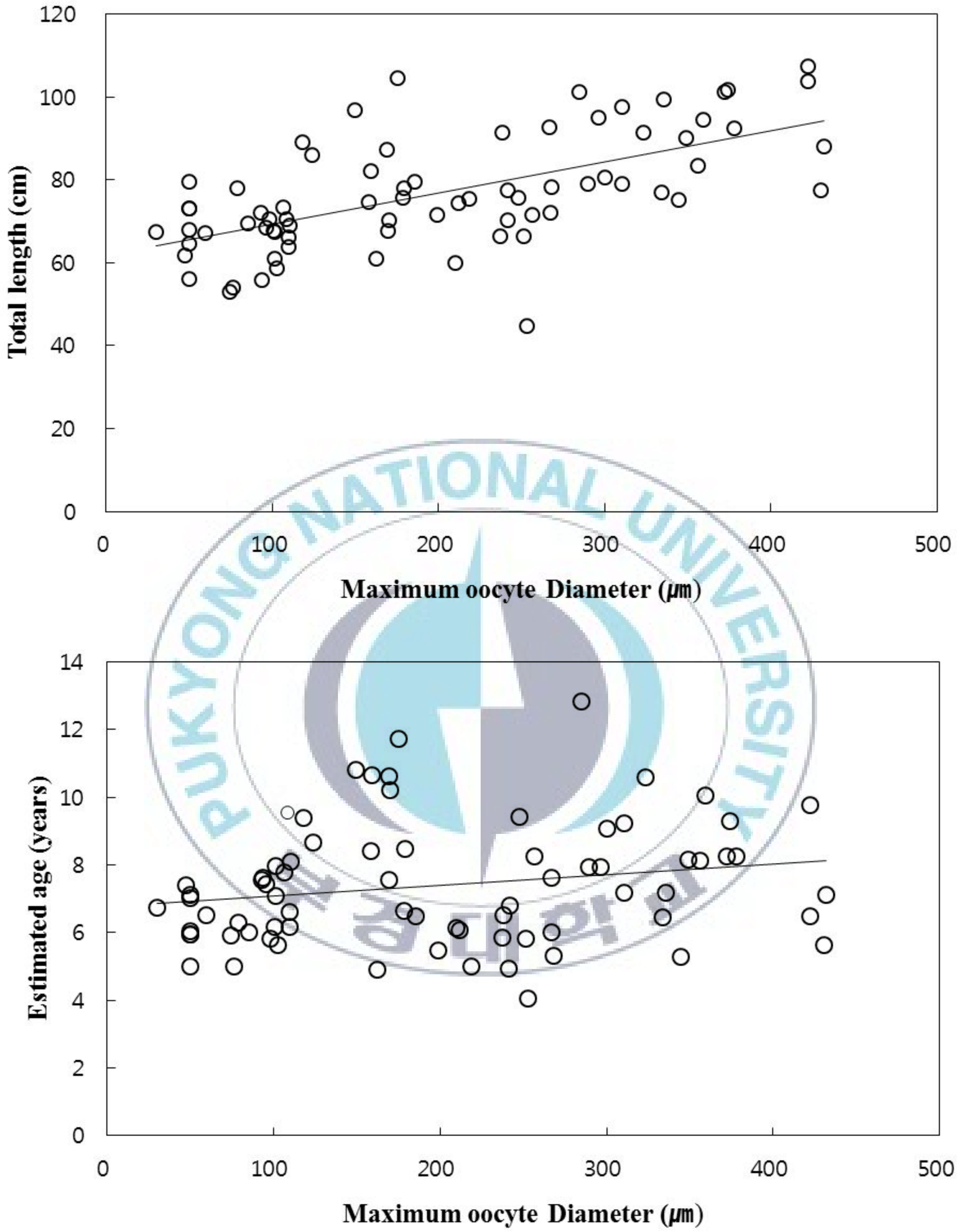


Fig. 13. Relationship between maximum oocyte diameter (MOD) and total length (upper graph), age (lower graph) in females of *Conger myriaster* (*n*=75).

4. Discussion

The white-spotted Conger eel, *Conger myriaster* arrives in coastal waters while they are undergoing metamorphosis (Lee and Byun, 1996; Wang, 2000; Kimura et al., 2004; Yagi et al., 2010). This study covers from after metamorphosis to adult stage. Samples collected in this study ranged from 15.6 to 107.5 cm TL and body weight ranged from 4.8 to 2,572.7 g, which covers the range that Gorie *et al*. (2004) reported.

In this study, 635 individuals were observed, among which 528 (83.2%) were females, 107 (16.8%) were males or immature individuals. Total length of female and male ranged from 34.4 to 107.5 cm and from 15.6 to 50.6 cm, respectively. The dominant of females were observed in the large length classes, relatively (Fig. 3). There were significant differences between female and male length-frequency distributions, two sampling and the sampling gears ($P<0.05$). Although the selectivity of the fishing gear was outside the scope of this study, different types of gears may have an effect in the size of collected fishes. A few of males have been captured exclusively in coastal waters. None of them are mature. These data are in agreement with the known sex-ratio fluctuation in function of depth for this species. The size and sex distribution in a collection could be influenced by gear selectivity, net avoidance, depth range sampled and trawl pattern. In case of the American conger eel, *C. oceanicus*, both males and females were size-selected from different fishing gears (Hood et al., 1988). For these reasons, we examined growth parameters by combining two sexes, not dividing them.

In recent years, there has been high demand for more sophisticated population studies aimed at providing detailed information on the population dynamics and demography of conger eels (Correia et al., 2009). Parameters for which specific estimates are required are growth rates, age structures and longevity. This information can be most conveniently obtained by the determination of age from otoliths (Fowler, 1990), provided the otoliths contain internal structures laid down according to a regular time able. In the present study, the validation of age was assessed for white-spotted conger in the coastal areas of Korea by using a specific method.

Transverse sectioned otoliths contained an alternating sequence of opaque and translucent bands that formed and interpretable pattern of increments. However, some congers have been wrong annulus of otolith caused confusing for age determination. Problem of age determination was observed under the microscope which uses a normal light. Katayama *et al*. (2002) devised that the burnt and UV light observation of otolith. Also, European conger, *Conger conger* has similar problem, but it is solved by using a UV light (Correia et al., 2009). Although UV light causes more time-consuming and dangerous work, it helps to distinguish the false rings (Fig. 6). Despite these efforts, some otoliths are difficult to read. The unclear increment part of otolith could represent a period of very slow growth. The several old annual rings are too thin to be distinguished and counted.

The seasonal variation of otolith opaque and translucent edges shown in this study demonstrated annual opaque zones formation in the otoliths of Conger eel. Therefore, we have found the interpreting annual growth rings in sagittal otoliths to be a valid method in ageing of white-spotted conger eel. The same results, including data on timing of opaque zone formation, have been obtained by Gorie *et al*. (2004) who found a high percentage of congers otoliths with opaque edges during spring and summer months in Japan. However, otolith structures of some specimens were seasonally formed, broadly. This is because that each specimen has different birthday. Some sectioned otoliths exhibited a structure of the edge that was weakly displayed. The check was mainly formed in the spring season; once a year (Fig. 7). Similarly, Katayama *et al*. (2002) observed that the bright zone of the otoliths was formed from April to October in Japan. Also, the European conger, *Conger conger* was similarly formed that opaque zone was deposited each year during the summer season (Correia et al., 2009).

It was difficult to determine the first annulus. Whether obscure ring is true or false annulus can be observed by studying the early life history (Lee and Byun, 1996; Correia et al., 2002). The first annulus formed from an after metamorphosis check. We decided hatch on 1 December from previous research (Lee and Byun, 1996). Estimated maximum ages differ among regions. In this study, we estimated that maximum age was 13 years old. It is different from the previous research (Gorie et al., 2004). It is considered that the difference in maximum ages might be due to geographical differences in life history or the fisheries, different exploitation rates, etc.

The otolith growth is regarded to be related to favorable somatic growth. Nevertheless, the biological relationship of fishes between otolith growth and morphological and ecological events are not fully understood. However, it is found that close relationship from total length at morphological characters of otoliths. Similar changes in otolith measurements with age and fish length have been described for other species of fish (Boehlert, 1985; Anderson et al., 1992).

A linear relationship is established between otolith length, width, radius and weight plotted against fish total length. The regression of total length (TL) and otolith weight (OWt) was best fitted relationship (Fig. 5). There was highest value for the adjusted coefficient of determination (R^2=0.8737). Therefore, we used equation from the estimated age at otolith weight.

It appears that fish length is a poor proxy for age in conger eels (Katayama et al., 2002; Gorie et al., 2004). Many researches in this field presented that OWt is a better age predicator than other otolith measurements, including otolith length, width and radius (Worthington et al., 1995; Newman et al., 2000; Francis and Campana, 2004). In this study, the regression of TL-OWt was determined for age groups. Otolith weight continued to increase linearly with age and it reached a maximum at the age of about 20 years (Fig. 8). This result shows the potential possibility that the life span can be estimated around 20 years old. Unfortunately, this age had not verified from the age validation.

The equation based on the regression of back-calculated length on otolith radius is larger than the mean otolith radius for the same body size. The growth calculations may be biased upwards due to Lee's phenomenon. For this reason, the asymptotic length (L_∞) of the von Bertalanffy parameter from back-calculation could have underestimated the length at age of the virtual value. Therefore, we can overcome handicap that minimized errors from parameters.

We approached with three-ways of equation. Three methods of growth function were calculated by back-calculation, age at mean length data and age at otolith weight regression method. This difference in the estimates of growth parameters is considered to be due to the use of the different equation methods. We compared the von Bertalanffy growth equation with those of each method (Table 3). Further support for our age estimates is given by the comparison of growth curves (Fig. 10). The von Bertalanffy growth curves were compared with likelihood ratio test. There was no significant differences (P=0.956). Therefore, we suggest that combined data showed best relationship between the age at total length (Fig. 14). The growth model was estimated as following VBGF parameters; L_∞=127.95 cm, K=0.102, t_0=-0.922. It is enhanced that accurate estimation of population growth. Also, the relationship of otolith weight at age can be useful to age determination for a conger eel, *C. myriaster*. In contrast, VBGF parameters of the European conger eel have been estimated in the east coast of Ireland, Irish coastal waters and Atlantic Iberian waters, indicated that L_∞= 265cm, K= 0.0633, t_0= -0.3861, L_∞= 271cm, K= 0.037, t_0= -1.396 and L_∞= 265cm, K= 0.07, t_0= -1.20, respectively (Fannon et al., 1990; Sullivan et al., 2003; Correia et al., 2009). It was shown long life span and slower growth than the white-spotted conger eel.

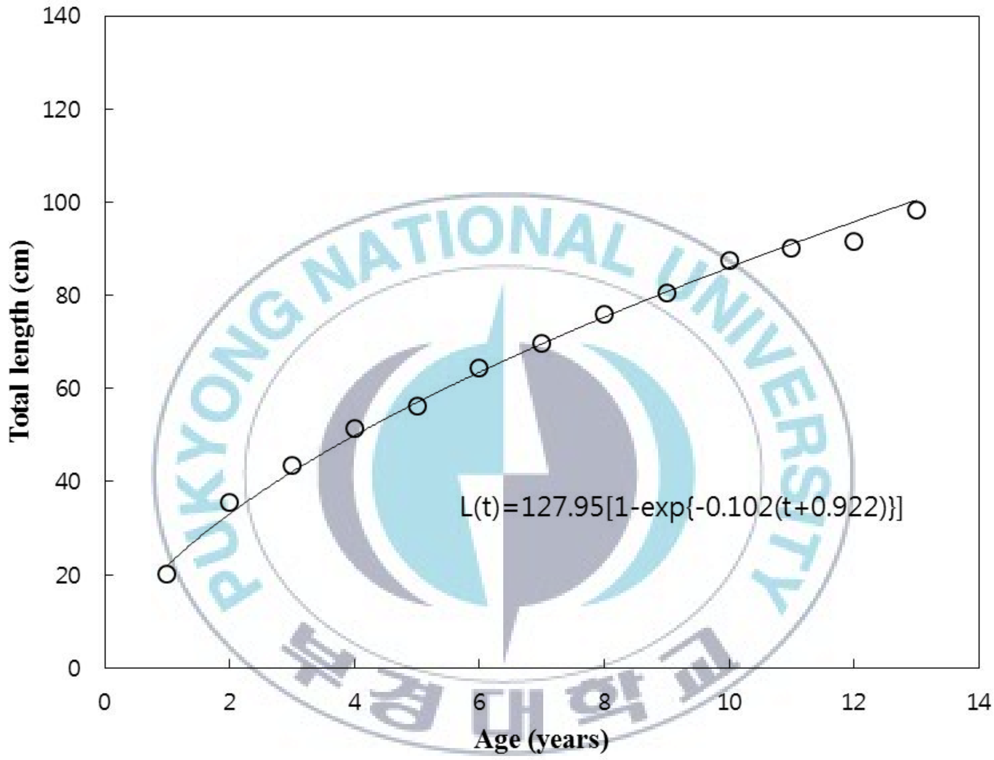


Fig. 14. Age-total length relationship with von Bertalanffy growth curve for combined data of *Conger myriaster*.

Fish condition is one of the most common measures of the nutritional state of a fish and depends on the sex, size, season and reproductive stage of an individual (Froese, 2006). The condition factor (Kn) of the collected samples was highest in summer and another seasons indicated similar values. Unexpectedly, the result from the hepatosomatic index (HSI) showed no significant mean differences among seasons. However, the gonadosomatic index (GSI) was related Kn that following seasons changes (Fig. 11). GSI was increased in autumn and winter, when the ovaries were at their largest size. These results was related at the hatching time that reported from September to February by previous study for a conger eel (Lee and Byun, 1996; Kimura et al., 2004)

In this study, the gonad examination was carried out for the ovaries except immature ones. Utoh *et al*. (2003) showed that detailed investigation process and characteristics of oogenesis in conger eel, *C. myriaster*. A close relationship between the development stage and GSI was also ascertained in the previous study. We obtained these report of development oocyte which was approached with compared our data.

Horie *et al*. (2002) examined that development of embryos were observed after artificial fertilization. It is presented that 1,000 ㎛ is required for fertilized egg of *C. myriaster*. In this study, Maximum oocyte diameter (MOD) ranged approximately from 50 to 430 ㎛ and the largest oocytes were still undergoing the secondary yolk globule stage from previous research. Average of present results was 201.4 ㎛ (±13.1). It is examined for the oil droplet stage (Utoh et al., 2003). We obtained no samples of maturation stage. According to previous study, GSI of maturation stage may be around 50 in conger eel. GSI varies during oogenesis. We suggest that goodness of fit from regression of maximum oocyte diameter (MOD) and GSI by the power function (Fig. 12). It is suggested that oogenesis begins to develop from 4 years old at 45cm TL (Fig. 13). However, gonad developmental biology was not enough to understand for a lifecycle.

In summary, age, growth and reproductive traits of the white-spotted conger eel, *Conger myriaster* were investigated. This is the detailed report clarifying and accuracy the ageing determination using a UV light observation. In addition, from relationship between the otolith measurements and age estimated the age structure of population of conger eel. Moreover, VBGF parameters estimated from three-methods by non-linear regression. There is close relationship between OWt at age. Reproductive traits of ovaries were examined and it showed strong positive relationship between GSI and MOD by the regression of power function. The present study was described to provide a better understanding of the age and growth for a conger eel in the Southern Sea of Korea.

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