#### **ORIGINAL ARTICLE**





# Age and growth of damselfish Chromis notata (Temminck & Schlegel, 1843), Jeju Island, Korea

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#### **Summary**

This study investigated the age and growth of damselfish, Chromis notate, from Jeju Island in Korea. Samples were collected monthly by lift net from September 2013 to August 2014. Total lengths of the damselfish ranged from 6.4 to 15.3 cm. The relationship between total length and wet weight was WW = 0.0125TL<sup>3.1631</sup> for females, and WW = 0.0091TL<sup>3.2769</sup> for males. The slopes in the relationship between length and weight were not significantly different between sexes, but were significantly different in the intercepts. There were more female than male specimens (1.3:1). Age determination was conducted using the otoliths. Marginal increment (MI) declined in summer and winter, which suggests that two rings are formed each year. Ages of sampled individuals ranged from 1 to 5 years. Length-at-age data were fitted using the von Bertalanffy growth model. The estimated growth functions were  $L_t$  = 19.93 [1 - $\exp^{-0.21(t+0.811)}$ ] total length and wet weight was females, and  $L_t = 16.47[1 - \exp^{-0.32}$ (t + 0.499)] for males.

# 1 | INTRODUCTION

Common techniques used to determine age in fishes include analyses of growth zones in vertebral bones, fin rays, scales, otoliths, and other skeletal structures (Zhang, 2010). The use of otoliths for age determination and growth estimates is increasing (Campana, 2005). Age determination is necessary to assess parameters such as growth, age at maturity, longevity, and mortality (Bagenal & Tesch, 1978). Assessment of growth parameters is vital for developing strategies to ensure sustainable fisheries (Rochet & Trenkel, 2003). Many previous studies in the ageing of fishes have focused on major commercial species because of their importance in supporting economies and providing sustenance to human populations.

The damselfish, Chromis notata (Temminck & Schlegel, 1843), is not a major commercial species. Due to global warming causing an increase in water temperatures (Myoung, 2002; Shin, Kim, & Choi, 2014) the distribution of C. notata now extends to the northern coast of Korea. Therefore recent catches of this species in Korea have more than doubled, from 78 tonnes (t) in 2005 to 174 t in 2014 (KOSIS, 2015). Consequently, its exploitation is increasing, but in the absence of fishery regulations. Basic information on C. notata is thus needed in order to develop effective management strategies.

The maximum length record of C. notata is 17 cm (Lieske & Myers, 2002). The main breeding season is from May to August (Go & Jeon, 1983; Lee & Lee, 1987), during which both sexes repeatedly undergo many reproductive cycles (Ochi, 1985). No studies of C. notata using otoliths had been made because age determination was thought to be difficult using this method (Go & Jeon, 1983). However, the technique was developed approximately 20 years ago, and the periodicity of annulus formation suggests that the technique is appropriate. Furthermore, most studies involving age determination of C. notata were conducted three decades ago, with results difficult to apply in the current fishery management. Population studies of C. notata are also lacking (Tzioumis & Kingsford, 1999; Shin et al., 2014).

Specific objectives of this study were to: (i) determine the age of C. notata using otolith analysis; and (ii) estimate C. notata growth parameters for fishery management purposes.

#### MATERIALS AND METHODS

#### 2.1 | Sampling method

The sampling area was located near Jeju Island, Korea (126.51°E, 33.23°S), whereby damselfish were collected using a 25.3 mm mesh size lift net once a month from September 2013 to August 2014, except in January 2014. All samples were collected randomly and brought in thermos cool boxes to the laboratory.

## 2.2 | Biological characteristics

Total length (TL) was measured in the laboratory to the nearest 0.1 cm for all fish samples. Wet weight (WW) and gonad weight (GW) were measured with an electronic balance to the nearest 0.01 g and 0.0001 g, respectively.

The sex was determined using the gonad morphological shape of each specimen. Gonads were staged visually in the laboratory and assigned to one of the following classes (Miller & Kendall, 2009): Immature, Developing, Mature, Spawning, and Spent. The gonad somatic index (GSI) was calculated for each sex as:

$$GSI = \frac{gonad\ weight}{total\ weight\ of\ fish} \times 100$$

The length-weight relationship was described by the equation

$$WW = aTL^b$$

where a (y-intercept) is the initial growth coefficient, and b (slope) is the growth coefficient. After logarithmic transformation, the equation above can be expressed as

$$logW = loga + blogL$$

Prior to regression analysis of log W on log L, log-log plots of length and weight values were performed for visual inspection of outliers (Froese, 2006). Only extreme outliers attributed to data error were omitted from analyses. The 95% confidence interval, CI of b was computed using the equation:

$$CI = b \pm (1.96 \times SE)$$

where SE is the standard error of b.

#### 2.3 | Otolith preparation and reading techniques

Sagittal otoliths were extracted from each fish head, soaked in 10% KOH solution for 5 min and dried for further observation. The prepared otoliths were embedded in epoxy resin and sectioned with a diamond saw along the longest axis across the core at about 1 mm intervals. Otoliths were viewed using a binocular stereoscopic microscope (Carl Zeiss Discovery v.8) with reflected light and a black background. Microscope magnification was 16×.

A growth ring was defined as a pair of bands consisting of one opaque zone (assumed to be fast growth) and one translucent zone (assumed to be slow growth). Growth rings observed in the translucent zone of each otolith were counted.

# 2.4 | Precision of age and annuli formation periodicity

To compare reading precision, the same reader read each structure at least twice. The average percentage error (APE) was used to compare

the initial age reading with the second reading (Beamish & Fournier, 1981):

$$APE_{j}(\%) = 100 \times \frac{1}{R} \sum_{i=1}^{R} \frac{\left|X_{ij} - X_{j}\right|}{X_{j}}$$

where R is the number of times each fish is aged,  $X_{ij}$  is i (th) age determination of the j (th) fish, and  $X_j$  is the mean age calculated for the j (th) fish.

Ring formation was done to ascertain whether age structures actually took place and to detect errors in the checked growth ring. Marginal increment (MI) analysis was used to validate the periodicity of growth:

$$MI = \frac{R - r_i}{r_i - r_{i-1}}$$

where R represents structure radius,  $r_i$  and  $r_{i-1}$  are annular radii of the last and penultimate annuli. MI was expressed as the percentage of structure with opaque and translucent margins and was plotted by monthly capture.

#### 2.5 | Growth parameters

Regression analysis was used to back-calculate total length from fish total length and otolith radius (Francis, 1990). The von Bertalanffy growth function (VBGF) was fitted to individual length and age data for the damselfish population and expressed by the equation:

$$L_t = L_{\infty} [1 - e^{-k(t-t_{0)}}]$$

where  $L_{\rm t}$  is the length (cm) at age t,  $L_{\infty}$  the asymptotic length (cm), k the rate at which the growth curve approaches the asymptotic length (year–<sup>1</sup>),  $t_0$  the hypothetical age of the fish at zero length.

For comparisons with past studies, the growth performance index was estimated in each case according to the formula of Munro and Pauly (1983):

Growth performance Index 
$$(\phi) = 2 \log L_{\infty} + \log k$$

where  $L_{\infty}$  is the asymptotic length, and k the growth constant of the von Bertanlanffy growth parameter.

#### 2.6 | Statistical analyses

The sex ratio difference was examined using a Chi-square test. Differences in size and wet weight between sexes were tested by one-way analysis of variance (ANOVA). The Kolmogorov-Smirnov two-sample test (K-S test) was applied to test differences in the size-frequency distributions between sexes. Analysis of covariance (ANCOVA) was applied to test differences in length-weight relationships in both sexes. A paired t-test was performed to compare first and second readings. The Kruskal-Wallis test was performed to test differences in monthly MI values. Nonparametric multiple comparisons were used as post-hoc test. Differences in growth parameters between sexes were compared by the method of Kimura's likelihood ratio test. These statistical tests were performed using Minitab v12.

#### 3 | RESULTS

### 3.1 | Biological characteristics

In total, 772 *C. notata* specimens were captured during the sampling period. The overall sex ratio of females (408) to males (319) was 1.3:1, significantly different from 1:1 ( $\chi^2$  = 10.681, df = 1, p < .01). Lengths of the sampled fish ranged from 6.4 to 15.3 cm, with no significant differences in mean lengths between sexes (F = 0.49, df = 1, p > .05) Wet weights were 3.73–71.1 g and 3.25–72.17 g in females and males, respectively. There was no significant difference in the mean wet weight of the two sexes (F = 0.05, df = 1, p > .05), or in their length frequency distributions (Kolmogorov–Smirnov test, p > .05).

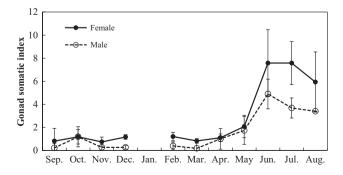
For females, the gonadosomatic index (GSI) peaked in June and July, and began to decline in August. For males, the GSI peaked in June, and declined in July (Fig. 1). Thus, the spawning season for *C. notata* was from June to August, and the major spawning period was from June to July.

The relationship between total length (TL) and wet weight (WW) was WW = 0.0125TL $^{3.1631}$  ( $r^2$  = .95, n = 342, p < .001) for females, and WW = 0.0091TL $^{3.2769}$  ( $r^2$  = .95, n = 295, p < .001) for males (Fig. 2). The 95% confidence interval of b was from 3.09 to 3.24 for females, and from 3.19 to 3.36 for males. This is within the expected range of 2.5 < b < 3.5. The slope in the relationship between TL and WW was not significantly different between the sexes (ANCOVA F = 3.75, p > .05), but there was a significant difference in the intercept (ANCOVA, F = 34.94, p < .001).

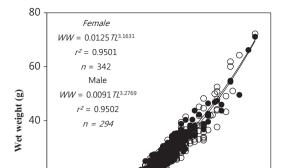
#### 3.2 | Age precision and annuli formation periodicity

Increments in otolith growth showed the typical pattern of translucent and opaque zones. Vertical lengths of the otoliths ranged from 2.9 to 6.89 mm. The core-to-increment distance increased as the otolith radius increased (Fig. 3). Of the 398 otoliths examined, similar replicate measurements were 76.05%, the average percentage error between the first and second measurements was very low (6.74%), and there was no significant difference between measurements (t = 0.726, p > .05).

The marginal increment (MI) was calculated for each month (Fig. 4), declining in summer (July) and winter (February), suggesting



**FIGURE 1** Monthly changes and mean gonado-somatic index (± standard deviation = bars), *Chromis notata* (*n*=398) by sexes. Samples collected from Jeju Island, September 2013 to August 2014



20

0

**FIGURE 2** Relationship between total length (TL) and wet weight (WW), *Chromis notate*, September 2013 to August 2014, collected using 25.3 mm mesh size lift nets. Solid circles and dashed line = females; Open circles and solid line = males

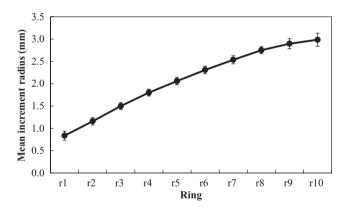
10

12

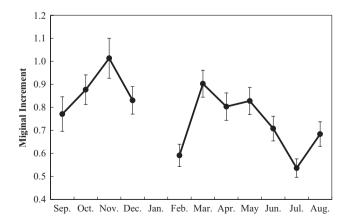
Total length (cm)

14

16



**FIGURE 3** Mean increment radius (± standard deviation = bars) on otolith of *Chromis notata* (*n* = 398). Samples collected from Jeju Island, September 2013 to August 2014



**FIGURE 4** Monthly change of marginal increment (MI ± standard deviation), *Chromis notata* (*n* = 398), September 2013 to August 2014, Jeju Island. MI declined in summer (July) and winter (February)

that two rings formed each year. Significant differences were found in the MI values among months (Kruskal–Wallis test, H = 52.51, p < .001). Nonparametric multiple comparisons revealed that the peak value in November differed from those in February, July, and August. The peak value in March differed from those in February and July. The lowest value in February differed from those in March, October, and November, and the lowest value in July differed from those in March, May, October, November, and December (Table 1). These results show that MI had a periodicity involving two increases and two decreases each year. The number of increments observed for *C. notata* ranged from 1 to 10, indicating that fish ranged in age from 1 to 5 years.

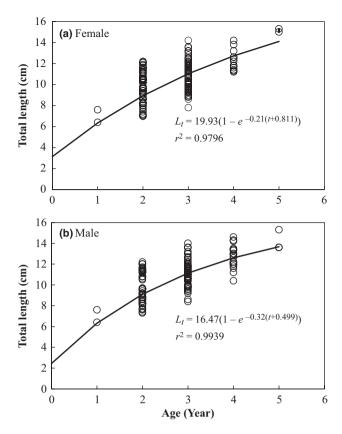
#### 3.3 | Growth parameters

The von Bertalanffy growth model for *C. notata* was estimated from back-calculation of the mean otolith annulus at age (Fig. 5). The von Bertalanffy growth functions for males and females were expressed with nonlinear regression according to the equations:

$$\begin{split} \text{Female:} \ & L_t = 19.93 \, \left( \, 1 - e^{-0.21(t + 0.811)} \, \right) \\ \\ \text{Male:} \ & L_t = 16.47 \, \left( \, 1 - e^{-0.32(t + 0.499)} \, \right) \\ \\ \text{Combined:} \ & L_t = 17.51 \, \left( \, 1 - e^{-0.28(t + 0.646)} \, \right) \end{split}$$

where  $L_t$  is the total length at age t.

A likelihood test was used to assess the differences in the growth curves for the two sexes. The likelihood test for the nonlinear



**FIGURE 5** The von Bertalanffy growth function (VBGF) for female and male *Chromis notata* (non-linear regression analysis). Samples collected from Jeju Island, September 2013 to August 2014. Circles are experimental values; lines are estimated trend lines

**TABLE 1** Nonparametric Multiple Comparisons between monthly marginal increment (MI) values of *Chromis notata*. Samples collected September 2013 to August 2014, Jeju Island. (n: sample size, *SD*: standard deviation)

	Sept.	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May	June	July	Aug.
Sept.												
Oct.												
Nov.												
Dec.												
Jan.												
Feb.		*	***									
Mar.						**						
Apr.												
May												
June												
July		**	***	*			***					
Aug.			*						*			
n	24	38	39	41		42	39	48	40	40	40	40
Mean MI	0.77	0.87	1.01	0.83	-	0.59	0.9	0.8	0.82	0.71	0.54	0.68
SD	0.08	0.06	0.09	0.06	-	0.05	0.06	0.06	0.06	0.06	0.04	0.05

<sup>\*</sup>p < .05.

<sup>\*\*</sup>p < .01.

<sup>\*\*\*</sup>p < .001.

**TABLE 2** Likelihood tests comparing von Bertalanffy growth parameters by non-linear regression estimates for *Chromis notata* (1) females and (2) males

Hypothesis	Linear constraints	L <sub>∞1</sub>	L <sub>∞2</sub>	K <sub>1</sub>	K <sub>2</sub>	t <sub>01</sub>	t <sub>02</sub>	Residual sum of squares (1)	Residual sum of squares (2)	X <sup>2</sup>	Р
$H_0$	None	19.93	16.47	0.21	0.32	-0.81	-0.5				
H <sub>1</sub>	$L_{\infty 1} = L_{\infty 2}$	17.93	17.93	0.27	0.26	-0.59	-0.71	0.07	0.16	11.75	< 0.001
$H_2$	$K_1 = K_2$	18.1	17.68	0.27	0.27	-0.6	-0.7	0.06	0.15	11.99	< 0.001
H <sub>3</sub>	$t_{01} = t_{02}$	18.65	17.18	0.25	0.28	-0.64	-0.64	0.05	0.13	13.21	< 0.001
H <sub>4</sub>	$L_{\infty 1} = L_{\infty 2} K_1 = K_2 t_{01} = t_{02}$	17.86	17.86	0.27	0.27	-0.64	-0.64	0.09	0.17	10.56	< 0.05

TABLE 3 Total length (TL) at estimated age for Chromis notata

Age (yr)	Females			Males			
	Estimated TL (cm)	Mean measured TL ± SD (cm)	Range (cm)	Estimated TL (cm)	Mean measured TL ± SD (cm)	Range (cm)	
1	6.4	7 ± 0.85	6.4-7.6	6.37	7 ± 05	6.4-7.6	
2	9.06	9.8 ± 19	7-12.2	8.98	9.7 ± 16	7.3-12.2	
3	11.02	11 ± 16	7.8-14.2	11.10	11.1 ± 14	8.4-14.0	
4	12.81	12.3 ± 05	11.2-14.2	12.89	12.7 ± 12	10.4-14.6	
5	13.75	15.2 ± 01	15-15.3	13.52	14.2 ± 08	13.6-15.3	

regression showed that there were significant differences between the sexes in the growth parameters asymptotic length ( $L_{\infty}$ ; p < .001), growth rate (K; p < .001), and hypothetical age ( $t_0$ ; p < .001) (Table 2). The asymptotic length value for females was larger than that for males (Fig. 4), and the growth rate for females was lower than that for males. The estimated and measured total lengths at age for C. notata are shown in Table 3.

The growth performance indices  $(\phi)$  for females and males were 3.92 and 3.94, respectively, with a combined value of 3.93 (Table 4).

#### 4 | DISCUSSION

An unequal sex ratio was observed for *C. notata*, probably attributable to the reproductive behavior of pomacentrid fish. Males migrate to the bottom and are involved in parental care during the spawning period (Ochi, 1985; Bracciali, Piovano, Sara, & Giacoma, 2014). The spawning period for *C. notata* was from June to August, as reported in Go and Jeon (1983); Suzuki, Hioki, and Kurita (1985); and Lee and Lee (1987). Gonads began to develop from April onwards. Gonad development is affected by the water temperature and photoperiod, and gonads begin to mature when the water temperature reaches 15°C and the photoperiod exceeds 12 hr (Lee & Lee, 1987).

For *C. notata*, there was a periodicity in the formation of the annulus, centering on February (winter) and July (summer), which suggests that two rings are formed each year. Annuli formation is associated with environmental factors and endogenous rhythms, and is affected by water temperature in winter. Incremental growth has been

correlated approximately with the rise and fall of the water temperature (Fowler & Doherty, 1992; Zekeria, Weertman, Samuel, Kale-ab, & Videler, 2006). Abrupt fluctuations in water temperatures can cause changes in the metabolic activity of fish, resulting in the deposition of opaque zones (Huo, Xie, Ma, Yang, & Huang, 2012). The growth rate of *C. notata* has also been reported to be slow during winter (Go & Jeon, 1983). Incremental growth during summer is affected by the spawning season, and is consistent with the period of peak GSI. The growth rate slows during the spawning season because energy is diverted from growth to reproduction (Karino, 1999). Smith and Deguara (2003) suggested that the formation of opaque and hyaline zones in the otolith is related to water temperature and spawning activity. Thus, the formation of annuli in the otoliths of *C. notata* could result from an interaction between water temperature and reproduction.

Chromis notata was estimated to have five age classes (ages 1–5 years). The number of growth rings ranged from 1 to 10. A 9-year-old *C. chromis* was found in a temperate area, based on counts of the growth rings in the scales (Dulcic & Kraljevic, 1995). Parma microlepis in Australia was estimated to have a maximum age of 37 years (Tzioumis & Kingsford, 1999). Studies of the ages of tropical pomacentrid fishes have been more common than those of temperate pomacentrids. Maximum ages in the family Pomacentridae determined by otolith analyses include: 10 years for Parma wardi (Fowler & Doherty, 1992); 32 and 27 years for Stegastes arcifrons and S. leucorus beebei, respectively (Meekan, Ackerman, & Wellington, 2001); 11 years for Acanthochromis polyacanthus (Kingsford & Hughes, 2005); 15 years for S. fuscus (Schwamborn & Ferreira, 2002); and as low as 4 years for S. flaviatus from the eastern Pacific Ocean (Meekan et al., 2001). Age

TABLE 4 Family Pomacentridae growth parameter comparisons reported from around the world

	Parameters							
Species	L <sub>∞</sub> (mm)	K t <sub>0</sub>		φ <sup>'</sup>	Climate	Region	Authors	
Pomacentruswardi	72.03	0.51	-1.402	3.43	Tropical	Great Barrier Reef, Australia	Fowler and Doherty (1992)	
Chromis chromis	142	0.26	-0.3	3.72	Temperate	Eastern middle Adriatic	Dulcic and Kraljevic (1995)	
Parma microlepis	135.8	0.41	-0.61	3.88	Temperate	Sydney, Australia	Tzioumis and Kingsford (1999)	
Stegastes acapulciebsus	118.8	0.77	-	4.04	Tropical	Eastern Pacific Ocean	Meekan et al. (2001)	
Stegastes arcifrons	103.8	0.75	-	3.91	Tropical	Eastern Pacific Ocean	Meekan et al. (2001)	
Stegastes flavilatus	84.7	1.95	-	4.15	Tropical	Eastern Pacific Ocean	Meekan et al. (2001)	
Stegastes fuscus	109.6	0.19	-	3.36	Tropical	Brazil	Schwamborn and Ferreira (2002)	
Acanthochromis polyacanthus	100.27	1.13	-	4.06	Tropical	Great Barrier Reef, Australia	Kingsford and Hughes (2005)	
Chromis notata	199.3 (female) 164.7 (male) 175.1 (combined)	0.21 0.32 0.28	-0.81 -0.49 -0.65	3.92 3.94 3.93	Temperate	Jeju Island, Korea	This study	

groups estimated here for *C. notata* are younger than the maximum ages determined for other pomacentrid fishes. However, it is possible that *C. notata* live for longer than five years because the maximum length of this species is recorded as 17 cm (Lieske & Myers, 2002). The variations observed in the ages of damselfish populations in different areas could be related to different population structuring processes, including competition for territory and predation, which could have different effects on these populations (Schwamborn & Ferreira, 2002).

The growth rate of pomacentrid fishes was compared using the growth performance index ( $\phi$ ) ecause the asymptotic length ( $L_{\infty}$ ) and the growth coefficient (K) cannot be used to compare growth rates among species. Compared with fish that inhabit temperate areas, C. notata is a fast-growing species, but its growth rate is generally similar to pomacentrid fishes worldwide. The most similar pattern of growth occurs in S. arcifrons and S. acapulcoensus, although the habitat of these species is the tropical eastern Pacific Ocean. Coral reef fishes are generally assumed to have rapid growth in their first year (Buesa, 1987; Sale, 2013). C. notata has been reported to grow 6 cm in their first year after hatching (Go & Jeon, 1983). Reef fish are clearly adapted to rapid growth, and invest their energy in reproduction as soon as possible for biological and physical reasons (Caldow & Wellington, 2003). Their growth is also strongly related to environmental factors, including temperature and food availability (Diana, 1954).

The growth pattern differed between sexes in this study, with significant differences between males and females in all three analyzed growth parameters (Table 4). Results indicate that males grow faster and reach maximum length at a younger age than females. The difference in growth between the sexes seems to be related to the reproductive effort (Vieira et al., 2009). Growth and reproduction of individuals are determined by the interactive effects of two physiological processes, based on the partitioning of net production between

growth and energy reserves (Houston & McNamara, 1999). In males, the absence of an energy switch between growth and reproduction is attributable to their lower energy demand for gonad development, thus most of their energy can be used for growth (Vieira et al., 2009). This study suggests that the von Bertalanffy growth curves for *C. notata* should be used separately for each sex and also indicates that the regulation strategies developed for the fishery management should be based on their different growth patterns.

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