Age, Growth, and Spawning Period of Bamboo Sole

*Heteromycteris japonica* in the Seto Inland Sea, Japan

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**Abstract:** The bamboo sole *Heteromycteris japonica* is a small and dominant species found on sandy seabeds in south Japan. To examine the age, growth, and spawning period, juveniles and adults were collected with sledge-nets and beam trawl-nets, respectively, in the Seto Inland Sea, Japan. This species spawned from May to June, as suggested by the high gonadal somatic index in May and June and by the occurrence of newly settled juveniles [<10 mm in total length (TL)] from June to July. Using otolith sectioning and etching methods, the annual formation of opaque zones in sagittally sectioned otoliths was confirmed. The ages of fish between 76 and 158 mm in TL were determined, and the von Bertalanffy growth equations based on age–length relationships were derived for male and female *H. japonica*. The maximum ages encountered were 9 years for males and 11 years for females.

**Key words:** Bamboo sole; Age and growth; Otolith; Seto Inland Sea

Soles are commercially important species in the Pacific Ocean (Wilderbuer et al. 2005). In the central Seto Inland Sea, Japan, *Cynoglossus abbreviatus*, *C. joyneri*, and *C. robustus* are landed by bottom trawls (Mototani 2011). Their age, growth, and spawning period have been studied for stock management purpose, and their asymptotic total length for growth equations ranged between 250 and 420 mm (Yamamoto et al. 2008, 2009; Katayama et al. 2010; Mototani 2011; Katayama and Yamamoto 2012). These soles grow rapidly until they are 2 years old (Yamamoto et al. 2008, 2009; Katayama et al. 2010; Kusakabe 2011; Katayama and Yamamoto 2012) and are mature at 1 or 2 years old (Mototani 2011; Katayama and Yamamoto 2012).

The bamboo sole *Heteromycteris japonica* (<150 mm in total length) is distributed off the coast of southern Japan and in the East China and Yellow Seas (Nakabo 2002). It has been reported to be a dominant species on sandy seabeds in the Seto Inland Sea (Fujiishi 1995; Yamamoto and Tominaga 2005), the Sea of Japan (Minami et al. 1977; Hashimoto et al. 1985; Uchida et al. 1998), and Sagami Bay, Kanagawa Prefecture (Katayama et al. 2007). However, little published information (Minami 1981) is available on its life history, such as the age, growth, and spawning period, because the species has no commercial significance. The main stomach contents of the species were found to comprise crustaceans, such as amphipods, ostracods, and copepods, as well as polychaetes (Minami et al. 1977; Minami 1981; Katayama et al. 2007), which suggests that there is potential food competition between this species and commercial flatfish species, such as ridged-eye flounder *Pleuronichthys cornutus* and black tongue fish *Paraplagusia japonica* (Minami et al. 1977). Therefore it is important to determine the abundance of bamboo sole, which requires studies of its life history characteristics.
In the present study, we determined the age using otolith sections and the spawning period of adult bamboo sole in the central Seto Inland Sea. We also estimated the von Bertalanffy growth equation based on the age–length relationships of adults and early juveniles.

Materials and Methods

Sample collection and treatments

Newly settled bamboo sole juveniles were sampled at intervals of 2 weeks to 2 months 28 days between May 2002 and December 2004 off the sandy Ohama Beach in the central Seto Inland Sea, Japan [34°13.6’N, 133°36.5’E; Fig. 1 (area A)]. A small sledge-net (net mouth = 2 m wide and 0.3 m height, mesh size = 2.1 mm) was towed by a boat for 200 m four times each day along the shoreline at a depth of 1–5 m. The samples were fixed in 5% neutral formalin within 2 h of collection and preserved in 90% ethanol. The total length (TL) of individuals was measured to the nearest 0.1 mm with calipers. Minami (1981) showed that metamorphosing larvae with a TL of 9.0 mm settled on sandy sea-beds after a pelagic period of 1–2 weeks based on field and laboratory observations. Thus, we considered that fish with a TL of <10 mm were newly settled juveniles.

To collect adult fish, monthly samplings were carried out using bottom trawls (~2.0 cm mesh at the cod end) of pebble and sandy sea-beds at a depth of 10–20 m between April 2008 and March 2009 [Fig. 1 (area B)]. The fish were packed on ice immediately and frozen at −20°C. In the laboratory, the TL and body weight (BW) of each fish was measured to the nearest 1 mm and 0.01 g, respectively, after which the sex was determined and the gonads were weighed (GW) to the nearest 0.01 g.

Sagittal otoliths were extracted from each fish, cleaned, and embedded in polyester resin. The embedded otoliths were then cut into transverse sections of measuring approximately 0.3 mm using a diamond saw (Leica, sp1600: Leica Microsystems GmbH, Wetzlar, Germany). The sections were mounted on glass slides and their surfaces were ground to sequentially finer grades using carborundum paper. Finally, the sections were etched with 0.2 N HCl for approximately 30 s and examined under a microscope using transmitted light, according to a published technique for observing the fine micro-structure of otolith annuli in the robust tongue sole C. robustus (Katayama and Yamamoto 2012). Similar to a previous reports (Tanaka et al. 2008; Yamamoto et al. 2008), we observed opaque zones and recorded the number of opaque zones and appearance of each zone at the outer margin of each specimen.

Data analyses

The maturational condition was determined using the gonadal somatic index (GSI):

\[
GSI = \frac{GW}{BW} \cdot 10^2.
\]

To estimate the parameters for the von Bertalanffy growth formula, we set a birth date of 30 May when the mean GSI of bamboo sole females was the highest (see result) and determined the male and female ages by counting the number of ring marks observed on the otoliths. The sample size was small for the young-of-the-year, so we utilized the age–length relationships for early juveniles at Ohama Beach. The length data from early juveniles were used for both sexes because their sex was not determined.

Using nonlinear least-squares estimation, we fitted the age–length relationships to the following von Bertalanffy growth equation:

\[
L_t = L_\infty [1 - e^{-k(t-t_0)}],
\]

where \(L_t = TL\) (mm) at age \(t\), \(L_\infty =\) asymptotic
TL, \( k = \) growth coefficient/year, \( t = \) age (year) and \( t_0 = \) hypothetical age when TL is 0. The difference in the growth curves between sexes was determined using an \( F \)-test (Akamine 2010).

**Results**

*Occurrence of newly settled sole off the sandy beach*

A total of 456 bamboo sole (mostly juveniles) were collected by sledge-net off Ohama Beach between April and December, whereas no fish were collected between January and March. TL varied between 7.2 and 143.0 mm. The newly settled juveniles (<10 mm) were present between June and July (Fig. 2). The mode of TL composition increased from June to August, i.e., 12.5 mm in June, 22.5 mm in July, and 27.5 mm in August. From October to December, most fish had a TL of between 35 and 70 mm (mean: 49.7 mm), whereas fish with a TL <35 mm were not observed. Based on the seasonal changes in TL composition, the fish with <70 mm collected (n = 366) were considered to be early juveniles.

**Growth equation**

In total, 181 etched transverse sections of otoliths were produced from adult fish. It was easy to observe the opaque zones of the sections (Fig. 3). The opaque zones at the outer margins of the otoliths occurred from March to July, except April, with >95\% appearing in July, followed by a decline in September (Fig. 4). The results suggest that the bamboo sole produce the annual otolith rings from June to July. The maximum age of bamboo sole was 9 years for males (127 mm TL) and 11 years for females (158 mm TL). Variation in the age determination was observed (Fig. 5).

Based on the age–length relationships of the adults (male, 61; female, 120) and early juveniles (male, 366; female, 366), the von Bertalanffy growth equations for males and females were estimated as follows (Table 1):

- **Male:** \( L_t = 123.6 [1 - e^{-1.15(t+0.03)}], n = 427, \)
- **Female:** \( L_t = 138.1 [1 - e^{-0.99(t+0.03)}], n = 486. \)

Bamboo sole grew rapidly until they were 2 years old, after which their growth rate decreased gradually. The equations differed significantly between males and females (\( F \)-test, \( P < 0.01 \)), which indicated that the females were larger than the males after 2 years.

Their BW increases exponentially with TL, and the relationship between TL (mm) and BW (g) was expressed as follows:

**Fig. 2.** Seasonal changes in size of bamboo sole collected by sledge-nets at Ohama Beach (area A) from May 2002 to December 2004. Fish smaller than 10 mm TL were considered as newly settled juveniles.
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Male:
\[ W = 1.70 \times 10^{-5} L^{2.91}, \quad r^2 = 0.91, \quad n = 61, \]
Female:
\[ W = 1.19 \times 10^{-5} L^{3.00}, \quad r^2 = 0.89, \quad n = 120, \]
where \( W \) and \( L \) are BW and TL, respectively.

Seasonal change in GSI

A total of 181 fish (mostly adults) were collected by bottom trawls in all the months except August and October. The mean GSI of males \( (n = 61, \text{86–139 mm TL}) \) remained at a low level \(<0.4\) throughout the year and seasonal variation was not observed \((\text{ANOVA}, P = 0.86; \text{Fig. 6})\), whereas the mean GSI of females \( (n = 120, \text{76–158 mm TL}) \) varied seasonally \((P < 0.01)\). The mean GSI increased from April to May and peaked highest on 30 May. There were

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**Fig. 3.** Transverse sections of otolith of bamboo sole (A, male 117 mm TL; B, female 118 mm TL) collected on 30 May, 2008. Arrows indicate opaque zones.

**Fig. 4.** Seasonal change in the proportion of a opaque zone on the outer margin of sectioned otolith. Numerals above figure indicate the numbers of the specimens.

**Fig. 5.** Von Bertalanffy growth curves fitted age-length relationships in bamboo sole. Open circles, closed circles and open triangles indicate female, male and unidentified, respectively.

**Table 1.** Based on the growth equations, estimated total length of bamboo sole (TL) at each age in the Seto Inland Sea, Japan

<table>
<thead>
<tr>
<th>Age</th>
<th>Male</th>
<th>Female</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.5</td>
<td>56.2</td>
<td>56.4</td>
</tr>
<tr>
<td>1</td>
<td>85.7</td>
<td>88.3</td>
</tr>
<tr>
<td>2</td>
<td>111.6</td>
<td>119.6</td>
</tr>
<tr>
<td>3</td>
<td>119.8</td>
<td>131.2</td>
</tr>
<tr>
<td>4</td>
<td>122.4</td>
<td>135.5</td>
</tr>
<tr>
<td>5</td>
<td>123.2</td>
<td>137.1</td>
</tr>
<tr>
<td>6</td>
<td>123.5</td>
<td>137.7</td>
</tr>
<tr>
<td>7</td>
<td>123.5</td>
<td>137.9</td>
</tr>
<tr>
<td>Asymptotic TL</td>
<td>123.6</td>
<td>138.1</td>
</tr>
</tbody>
</table>

**Fig. 6.** Seasonal change in the mean gonad somatic index (GSI) of adult female (A) and male (B) bamboo sole collected by a beam trawl (area B) from April 2008 to March 2009. Vertical bars and different superscripts indicate standard deviation and significant differences (Scheffe’s \( F \)-test), respectively. Numerals above figure indicate the numbers of the specimens.

Male: \( W = 1.70 \times 10^{-5} L^{2.91}, \quad r^2 = 0.91, \quad n = 61, \)
Female: \( W = 1.19 \times 10^{-5} L^{3.00}, \quad r^2 = 0.89, \quad n = 120, \)
where \( W \) and \( L \) are BW and TL, respectively.

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higher values (>5.0) in May and June compared with other months (Scheffe’s F-test, \( P < 0.05 \)). Subsequently, the GSI decreased and remained at a low level (<2.0) from September to March. The female GSI–age relationship suggested that the age at first maturation was 2 years old (Fig. 7).

**Discussion**

This species spawns from May to June, based on the high GSI from May to June and the occurrence of newly settled juveniles from June to July. The spawning period of bamboo sole was shorter than those of the three commercial sole species that inhabit the same study area of the Seto Inland Sea, *C. abbreviatus*, *C. joyneri* (Katayama et al. 2010; Mototani 2011), and *C. robustus* (Kusakabe 2011; Mototani 2011; Katayama and Yamamoto 2012). However, the first ring was not a spawning check in bamboo sole, *C. abbreviatus* and *C. robustus*, because the ring was formed in immature fish. Therefore, the ring formations might be related to stresses other than spawning.

The age determination data varied widely. However, the results were valid because opaque zones were formed on the otoliths. Thus the dispersion of the age–length relationships was not an assessment error.

The growth equation fitted to the age–length relationships from early juveniles to adults can be used to estimate the growth rate of juveniles. In general, given that the value of \( t_0 \) (hypothetical age when TL = 0) used in the growth equations was estimated based on the age–length relationships of adults only, the body lengths estimated using these equations were not suitable for small juveniles (e.g., Masuda et al. 2003; Shimose and Tachihara 2005; Yamamoto et al. 2009). In contrast to our growth equations based on the age–length relationships of adults and early juveniles, the value of \( t_0 \) was almost zero and the equations described the growth rate during the small juvenile to adult stage. Our method might be a simple and a useful tool for determining age-length relationships.

The bamboo sole females grew larger than the males. This growth characteristic has also been reported in the commercial sole species, *C. abbreviatus* (Yamamoto et al. 2008; Katayama et al. 2010; Mototani 2011) and *C. robustus* (Kusakabe 2011; Mototani 2011; Katayama and Yamamoto 2012). In flatfish there is generally a sexual dimorphism where females grow larger than males (Nash and Geffen 2005).

The maximum age and TL of bamboo sole species will be required to clarify the reproductive ability and particular aspects of spermatogenesis in soles.

The opaque zones of the otoliths formed during the spawning season, from June to July. This relationship has also been reported in the commercial sole species in the Seto Inland Sea, *C. abbreviatus*, *C. joyneri* (Katayama et al. 2010; Mototani 2011), and *C. robustus* (Kusakabe 2011; Mototani 2011; Katayama and Yamamoto 2012). In flatfish there is generally a sexual dimorphism where females grow larger than males (Nash and Geffen 2005).
were 11 years and 158 mm, respectively. The maximum ages (maximum TL) for *C. abbreviatus*, *C. joyneri*, and *C. robustus* in the same study area were 6 years (387 mm; Yamamoto et al. 2008; Katayama et al. 2010), 5 years (281 mm; Yamamoto et al. 2009; Katayama et al. 2010) and 11 years (451 mm; Kusakabe 2011; Katayama and Yamamoto 2012), respectively. The maximum TL of bamboo sole was small compared with these other sole species, but the longevity of this species matched that of *C. robustus*. Nash and Geffen (2005) suggested that there is a relationship between commercial exploitation and longevity. Thus, the long life of bamboo sole might be explained by a low fishing pressure.

As explained previously, the bamboo sole specimens were smaller than the three commercial sole species, *C. abbreviatus*, *C. joyneri* (Katayama et al. 2010; Mototani 2011), and *C. robustus* (Kusakabe 2011; Mototani 2011; Katayama and Yamamoto 2012), and the first maturation size of the bamboo sole was small. However, despite the differences in the body and maturational size, a similar growth rate was observed in the sole species. The bamboo sole grew rapidly until the first maturation age (2 years old). Similarly, the three commercial sole species grew rapidly until the first maturation age (2 years old; Yamamoto et al. 2008, 2009; Katayama et al. 2010; Kusakabe 2011; Katayama and Yamamoto 2012). Thus, rapid growth until first maturation age was a characteristic of sole species in the Seto Inland Sea. The individual growth rate has been reported to affect the allocation of available energy between egg production and somatic growth in adults (Nash and Geffen 2005). Therefore, the low growth rate after the first maturation season might have been caused by the energy expenditure during reproduction.

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**References**


瀬戸内海におけるササウシノシタの年齢、成長、産卵期

山本昌幸・片山知史

浅海域において多く分布するササウシノシタ Heteromycteris japonica の成長と産卵期を調べるため、瀬戸内海において稚魚と成魚をそれぞれ2 m 桁網と小型底びき網を用いて採集した。雌の生殖腺は5月～6月に高く、着底直後の稚魚は6月～7月に採集された。これらの結果から、本種が5月～6月に産卵することが示唆された。耳石（端平石）の横断切片を作成して、耳石の不透明帯を観察し、これが年齢形質であることを確認した。検体（全長: 76～158 mm）のオスとメスの最大年齢はそれぞれ9歳と11歳であった。Von Bertalanffy の成長式は、オスでは $L_t = 123.6 \times [1-e^{-1.15(t+0.03)}]$、メスでは $L_t = 138.1 \times [1-e^{-0.99(t+0.03)}]$ とあった。