A Study on the Reproduction of the Green Snail, *Turbo marmoratus* in the Ryukyu Islands, Southern Japan

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Abstract

The green snail, *Turbo marmoratus* had sex ratios of 1:1 both in Tokunoshima Island and the Yaeyama Island Group, the northern and southern Ryukyu Islands, Japan. No significant differences were found in mean oocyte sizes either between different sections in an ovary or between ovaries with different gonad bulk indices (GI: percentage gonad area to the total cross-sectional area of the gonad-digestive gland) when GI exceeded 50%. The number of oocytes per unit weight was significantly different among different portions in an ovary, and this could be attributed to the difference in the specific gravity of oocytes. The maximum number of oocytes in an ovary was estimated between 1.279×10⁶ and 7.523×10⁶ for females of 12.5-19.3 cm in shell width. The main spawning season was August-November in Tokunoshima I., and July-November in the Yaeyama Island Group. Mature individuals were found during all seasons in both localities. The relationship between the spawning activity and the lunar cycle was examined for *T. marmoratus* from Tokunoshima I., but the relationship was not found.

The largest species of the family Turbinidae, the green snail *Turbo marmoratus* is dioecious, and broadcast its gametes into the water. It can exceed 20 cm in shell width and 3 kg in total weight. It has been exploited from ancient times as one of the most important fishery resources in the Ryukyu Islands, Okinawa, Japan¹. Not only has the meat been eaten, but its nacreous shell has been valued for mother-of-pearl inlay works and various ornaments.

Yamaguchi¹ noted that the over-exploitation of *T. marmoratus* was already in progress in the 1880’s in the Ryukyu Islands. Over-exploitation occurs elsewhere in the western Pacific and Southeast Asia¹,² where *T. marmoratus* is economically important. Today, *T. marmoratus* does not constitute an important fishing target as the animal is just an occasional by-product among others caught by diving fishery in Okinawa.

In order to help facilitate recovery of this resource, a positive approach such as restocking with mass-produced seed may be necessary in addition to closure of *T. marmoratus* harvesting in the Ryukyu Islands and other areas. Compared with other commercially

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important gastropods like *Halotis* spp., *Turbo cornutus* and *Trochus niloticus*, information on the biology and ecology of *T. marmoratus* is limited.\(^1,2,3,4,5,6,\) \(^1,2\)

Mass seed production of *T. marmoratus* has been undertaken at the Okinawa Prefectural Sea Farming Center since 1989.\(^7\) In order to establish restocking methods of *T. marmoratus* with mass produced seed, information on fisheries biology of this species is needed.

To make seed production of *T. marmoratus* predictable, reproductive cycle, sex ratio, fecundity and relationships between spawning activities and the lunar cycle were examined in the present study.

**Materials and Methods**

A total of 530 *T. marmoratus*, of which 80 were whole animal and 450 were soft body with operculum without shell, were collected from different areas in different months from 1985 to 1990 (Table 1). Specimens were collected at Tokunoshima Island belongs to the Amami Island Group, the Yaeyama Island Group and Okinawa Island (Fig. 1). In order to examine a relationship between a lunar phase and a spawning bout, short term changes in gonad conditions were studied. For this purpose, specimens were collected at intervals of 4 to 47 days spanning a lunar cycle during the period from March 29 to July 29, 1989 from Tokunoshima I.

Guts and gonad-digestive glands of 501 *T. marmoratus* were excised, and then fixed and stored in 10% formalin solution for the examinations of sex ratio, fecundity and gonad bulk index mentioned later.

**Measurement of shell size**

In order to estimate the shell sizes, which were necessary for estimation of fecundities, of the specimens without shells, shell widths (SW, after Devambez\(^3\)) and the operculum greater diameters (OGD) of specimens from Tokunoshima I., the Yaeyama Island Group and Okinawa I.: 25, 26 and 29 individuals respectively, ranging 11.3-21.5 cm in SW, were measured (Fig. 2). The correlation between SW and OGD was shown by the equation: \(\text{SW} = -3.8422 + 2.5945 \text{OGD} \ (r = 0.8913, P < 0.05, n = 80)\).

**Gonad bulk index**

Gonads are located along the entire length of the digestive gland, and overlap it laterally (Fig. 3). In gonads of mature individuals, the sexes were distinguishable macroscopically, i.e., ovaries exhibited a granular structure in olive green or dark green just beneath a translucent epithelium, while testes appeared much smoother with a creamy white color.

The gonad bulk index (GI) of Newman (1967, cited in Wells and Keesing\(^8\)) and Hahn\(^9\) was used to

<table>
<thead>
<tr>
<th>Collecting area</th>
<th>Number of specimens</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tokunoshima I. (Amami Group)</td>
<td>1985</td>
</tr>
<tr>
<td></td>
<td>1986</td>
</tr>
<tr>
<td></td>
<td>1987</td>
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<tr>
<td></td>
<td>1988</td>
</tr>
<tr>
<td>Total</td>
<td>1989</td>
</tr>
<tr>
<td>Yaeyama Group</td>
<td></td>
</tr>
<tr>
<td></td>
<td>1987</td>
</tr>
<tr>
<td></td>
<td>1988</td>
</tr>
<tr>
<td>Total</td>
<td></td>
</tr>
<tr>
<td>Okinawa I.*</td>
<td></td>
</tr>
</tbody>
</table>

* collected for mass seed production\(^7\), used only for shell size measurement in this study.


Reproduction of *Turbo marmoratus* in the Ryukyus

Fig. 1. Map of the Ryukyu Islands. Areas enclosed by broken lines are where *Turbo marmoratus* sampled.

Fig. 2. Measurements of shell width (SW, after Devambez) and operculum greater diameter (OGD) of *Turbo marmoratus*.

describe gonad conditions. Gonad-digestive glands were cut at the three positions described by Newman (1967, cited in Giorgi and DeMartini), and GI were monitored at Section A, where is just behind the gastric caecum. (Fig. 3). The diameters of the total gonad-digestive gland and that of the digestive gland at Section A were measured to the nearest 0.1 mm with vernier calipers.

Gonad bulk index obtained by these measurements (GI') were calculated from $GI' = 100 \left( \frac{R^2 - r^2}{R^2} \right)$, where R and r are the radii of the total gonad-digestive gland and that of the digestive gland respectively. The GI' values were correlated with GI values which were obtained preliminarily based on direct measurements of the cross-sectional areas of gonad-digestive gland and that of gonadal portion. Therefore, the GI' values converted into GI values by the following regression equations: $GI = -1.7115 + 0.9349GI'$ ($r = 0.9693, P < 0.05, n = 78$) for female and $GI = 0.5367 + 0.842GI'$ ($r = 0.9478, P < 0.05, n = 77$) for male.

Presumptive fecundity

Twenty female *T. marmoratus* (GI > 50 %) from the specimens collected at Tokunoshima I. in 1989 were assessed for fecundity by means of the number of eggs present in ovaries, which was called as presumptive fecundity. Ovaries were excised, and their total wet weights were measured individually. A small piece of ovary, approximately 0.01 g in wet weight, was dissected out from each of the three sections in an ovary, i.e., just behind the gastric caecum (Section A) and at the distances of 1/2 (Section B) and 3/4 (Section C) of the length between the posterior end of the gastric caecum whorl and the tip of the gonad. g: gonad; dg: digestive gland.

For 10 females selected from every GI level of 50, 60 and 70 %, sizes of 50 oocytes were determined with a profile projector and vernier calipers under magnification of × 50. Most of the oocytes measured, however, appeared square or rectangular in profile, so that two neighboring sides of a quadrilateral oocyte profile were measured. The area of the oocyte profile was calculated, and then it was calculated into a diameter of a circle, which was considered to be oocyte size in the present study.

**Results**

**Sex ratio**

Twenty-six out of 372 individuals from Tokunoshima I. were spent. Of the remaining 346 snails, 175 (50.6 %) were female and 171 (49.4 %) were male. For the specimens from the Yaeyama Island Group, 67 specimens (52.3 %) were female and 61 (47.7 %) were male but sex could not be determined in one specimen. In both localities, sex ratios were not significantly different (χ² test, P > 0.05) compared to the expected 1:1 ratio.

**Gonad bulk index**

(1) seasonal changes

Fig. 4 is the monthly GI changing patterns of *T. marmoratus* from Tokunoshima I. for the five years combined (1985-1989). The mean GI for females was maintained above 30 % -level most of the year. The mean GI of 60 % was marked in August, and such the level was assumed to be continued, without conspicuous peak, until November. Then, GI declined steeply from 60 % in November to 25 % in December. The maximum GI indicated the values larger than 60 % most of the year, and this suggested that ripe females existed in the population throughout the year (Fig. 4). The minimum GI in August and November were above 40 %. The rest of the year except January and October, individuals with GI less than 12 % appeared consistently. The mean GI of the males did not change significantly, and indicated the values between 15 and 25
Reproduction of *Turbo marmoratus* in the Ryukyus

% from January to June and in December. The males indicated smaller GI ranges than those of the females except for August. The GI declined steeply from 54% in November to 15% in December.

Seasonal GI changes of *T. marmoratus* from the Yaeyama Island Group are shown in Fig. 5. The mean GI for the females was 37% in August, 1987, and this level was maintained until October (36%). The mean GI rapidly decreased from 30% in November to 9% in December. Recovery of gonad condition appeared to commence between April and May, and the mean GI reached 35% in July 1988. The mean GI of the males fluctuated between August and November 1987 while that of the females did not.

(2) short term changes

The mean GI of the females (Fig. 6A) demonstrated fluctuations within the period from March 29 to July 29 in 1989. Abrupt declines of GI occurred between April 5 and 9, and between May 10 and 22. On the other hand, the mean GI of the males shifted without fluctuations from March 29 through May 10 (Fig. 6B). Short term changes in mean GI are also represented together with the lunar cycle (Fig. 6). Evidences which showed that spawning activities related to a certain lunar phase were not found.

![Fig. 4. Seasonal changes in mean gonad indices combined for 5 years (1985-1989) in *Turbo marmoratus* from Tokunoshima I. Perpendicular bars indicate ranges. A: female; B: male.](image)

![Fig. 5. Seasonal changes in mean gonad indices in *Turbo marmoratus* in the Yaeyama Island Group (1987-1988). Perpendicular bars indicate ranges. A: female; B: male.](image)

![Fig. 6. Short term fluctuation of mean gonad indices in *Turbo marmoratus* in Tokunoshima I. (1989). The lunar phases are indicated together. Perpendicular bars indicate ranges. A: female; B: male.](image)

Presumptive fecundity

(1) oocyte size

The oocyte size ranged from 180 to 350 μm, and the average was 255 μm. Mean oocyte sizes for three
gonad positions (Fig. 3) within the same ovary were not significantly different for 10 individuals measured (GI ranging from 51 to 73 %, ANOVA, P > 0.05). There was no significant correlation between mean oocyte size and gonad index (GI > 50 %, r = 0.0429, P > 0.05, n = 10).

(2) number of oocytes in ovaries

Number of oocytes per 0.01 g counted for three positions of ovaries were significantly different (ANOVA, P < 0.05). Mean number of 787 oocytes/0.01 g at Section C (Fig. 3) was 13 to 16 % greater compared to those of 679 and 697 oocytes/0.01 g at more anterior Sections A and B respectively.

There was a significant positive correlation between shell width of females and the weight of their ovaries (GI = 62-73 %, r = 0.9789, P < 0.05, n = 8). Ovarian weight of those eight T. marmoratus ranged from 15.8 g for a 12.5 cm individual to 121.2 g for a 19.2 cm individual (Table 2). Presumptive fecundities were ranged from 1,279,000 oocytes for a female of 12.5 cm in shell width with GI of 62 % to 7,523,000 oocytes for a female of 19.3 cm with GI of 73 % (Table 2). The presumptive fecundities were significantly correlated to shell width (r = 0.9821, P < 0.05, n = 8) with the equation: F = 64502.9e (0.2428SW), where F is presumptive fecundity and SW is shell width in cm.

Discussion

It is necessary to confirm whether the year-round appearance of ripe ovaries is of retention of unspawned oocytes or the results of ordinary process of oogenesis in T. marmoratus. The fact that ripe females existed all year round has been reported for Haliotis rufescens, H. cracheroides, and H. kamtschatkana. Trochus niloticus in Palau and in the Andaman Islands spawned all year round. T. niloticus in the Great Barrier Reef appeared to spawn, as a population, at approximately monthly intervals all year round but individual females may spawn once every two to four months. Giorgi and DeMartini reported that within a spawning season, there were three types of females that have different spawning patterns, such as complete spawning, incomplete spawning—this pattern, GI does not indicate full maturation of ovaries but these are packed with ripe oocytes, and nonspawning in the same population in H. rufescens. It was presumed by histological examination that the residual oocytes retained by incomplete spawner or nonspawner of female H. rufescens became necrotic, and eventually, resorption of the oocytes would be occurred. Such the phenomenon was also suggested for T. marmoratus by unpublished data of histological examination of ovaries by the authors, and this may suggest the existence of females which do not participate in reproduction under certain conditions.

The relationship between spawning cycle and the lunar cycle has been reported for Trochus niloticus by direct observations of tank-held animals. However, Grange did not find such relationships in two trochids as Melagraphia aethiops and Zediloma atrovirens and in one turbinid as Lunella smaragda. The relationship between the timing of spawning and the lunar cycle was not clear in T. marmoratus. As a possible reason why the relationship was not detected, it is thought that there probably were individuals that

<table>
<thead>
<tr>
<th>Shell width (cm)</th>
<th>Gonad index (%)</th>
<th>Gonad wet weight (g)</th>
<th>No. of eggs* per 0.01 g wet weight</th>
<th>No. of Eggs** per animal</th>
</tr>
</thead>
<tbody>
<tr>
<td>12.5</td>
<td>62</td>
<td>15.8</td>
<td>808 (± 52)</td>
<td>1,279,000</td>
</tr>
<tr>
<td>12.7</td>
<td>71</td>
<td>23.1</td>
<td>730 (± 77)</td>
<td>1,689,000</td>
</tr>
<tr>
<td>13.9</td>
<td>64</td>
<td>32.8</td>
<td>760 (± 75)</td>
<td>2,491,000</td>
</tr>
<tr>
<td>14.9</td>
<td>62</td>
<td>32.0</td>
<td>790 (± 121)</td>
<td>2,529,000</td>
</tr>
<tr>
<td>16.6</td>
<td>71</td>
<td>47.5</td>
<td>744 (± 119)</td>
<td>3,529,000</td>
</tr>
<tr>
<td>17.6</td>
<td>71</td>
<td>60.7</td>
<td>686 (± 15)</td>
<td>4,165,000</td>
</tr>
<tr>
<td>19.2</td>
<td>63</td>
<td>121.2</td>
<td>531 (± 66)</td>
<td>6,432,000</td>
</tr>
<tr>
<td>19.3</td>
<td>73</td>
<td>119.8</td>
<td>628 (± 95)</td>
<td>7,523,000</td>
</tr>
</tbody>
</table>

* Mean of the three sections A, B and C, (±S. D.).
** Rounded to the nearest thousands.
spawn in different patterns or intensities, that were represented by GI variation, in the same population of T. marmoratus as reported for H. rufescens10). Concerning with the spawning season, patterns and annual frequency of spawning in T. marmoratus should be studied. For the species such as T. marmoratus whose population is seriously declined, however, it should also be necessary to consider whether they still exhibit their genuine reproductive ecology or not.

In the estimation of the number of eggs, there were significant differences in the number of oocytes per 0.01 g of ovary among the different portions in an ovary, especially between the basal part and the tip of the gonad. A similar trend was found in the Californian red abalone H. rufescens10). Since no significant difference was indicated in mean oocyte sizes in each of three positions in the ovary, the difference in the number of oocytes per 0.01 g may indicate the difference in specific gravity of individual oocytes in each portion. Non-synchronous maturation of oocytes in an ovary are suggested for Trochus niloticus21), and Monodonta liniata and Gibbula umbilicalis22). Asano21) reported that maturation of gametes did not occur synchronously throughout a gonad but that it occurred gradually from the periphery of the gonoduct in T. niloticus in Palau. In T. marmoratus, there is also a possibility that its maturation may not be synchronized throughout an ovary on account of qualitatively different groups of oocytes within the same ovary. A more precise examination on the ripening process (e.g., vitellogenesis) is needed to explain the possible differences in the specific gravity of the oocytes among sections in an ovary.

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References

12) Kakuda, N., T. Watanabe, N. Yurano, and M. Jinnouchi (1986): Studies on maturation and spawn-
琉球列島におけるヤコウガイの生殖について

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琉球列島のそれぞれ北部と南部に位置する，徳之島および八重山諸島におけるヤコウガイ，Turbo marmoratus の性比は，それぞれ 1 対 1 を示した。生殖腺指数が 50% を越える雌個体に関しては，卵果内の異なる部位における成熟卵の平均卵径値に有意な差は見られず，また，異なる生殖腺指数を示す個体間における平均卵径値にも有意な差は現れなかった。一方，上述の個体について卵果の単位重量当たりの卵数を比較した結果，有意差が見られたことにより，卵果内の部位の違いによる卵の比重変化が示唆された。成熟した雌1個体の卵果中の平均卵卵数は，殻幅 12.5 cm の最小標本個体において 1,279×10^6 粒，19.3 cm の最大標準個体において 7,523×10^6 粒と推定され，個体サイズの増加とともに卵数は指数関数的に増加した。ヤコウガイの主たる産卵期は，徳之島においては 8 〜 11 月，八重山諸島においては 7 〜 11 月であると推定され，いずれの海域においても雌雄の成熟個体が同年出現した。徳之島産ヤコウガイに関して，産卵行動と月齢周期との関係を調べたが，明確な関係は見出されなかった。