Половой диморфизм и морфометрический анализ Filopaludina martensi martensi (Gastropoda: Viviparidae)

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ABSTRACT. Sexual dimorphism is the condition where individuals of different sex in the same species exhibit different characteristics beyond the differences in their sexual organs. In this study, individuals of a viviparid species Filopaludina martensi from the Kwai Yai River in Kanchanaburi Province (Thailand) were examined for eight shell and operculum characteristics. Sexual differences were observed in size of shell and operculum, with females being larger than males. The results indicated that morphometric analyses are useful to detect subtle differences between sexes in this species.

Sexual dimorphism and morphometric analysis of Filopaludina martensi martensi (Gastropoda: Viviparidae)

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ABSTRACT. Sexual dimorphism is the condition where individuals of different sex in the same species exhibit different characteristics beyond the differences in their sexual organs. In this study, individuals of a viviparid species Filopaludina martensi from the Kwai Yai River in Kanchanaburi Province (Thailand) were examined for eight shell and operculum characteristics. Sexual differences were observed in size of shell and operculum, with females being larger than males. The results indicated that morphometric analyses are useful to detect subtle differences between sexes in this species.

Introduction

Sexual dimorphism describes the systematic differences in phenotypic traits between individuals of different sex in the same species [Shine, 1989], and is widespread among living organisms [Shine, 1989; Abouheif, Pairbairn, 1997], including molluscs [Purchon, 1977; Pastorino, 2007]. Many examples of sexual dimorphism in molluscs have been reported, including unionoid mussels [Jass, Glenn, 2004] and some marine, freshwater, and terrestrial snails [Araikawa, Hayashi, 1972; Jokinen et al., 1982; Cazzaniga, 1990; Raven 1990; Brande et al., 1996; Kenchington, Glass, 1998; Estebenet, 1998; Gofas, 2001; Pastorino, 2007]. Most known cases of sexual dimorphism in molluscs are generally related to size differences, where female shells are usually larger than males [Morse, 1876; Warén, 1983; Simone 1996; Pastorino, 2007 and references therein; Riascos, Guzman, 2010].

Freshwater snails for this study were collected from the Kwai Yai River in the Plant Genetic Protection Area of Srinagarind Dam, Sisawat District, Kanchanaburi Province in September 2009, and later identified as Filopaludina martensi martensi (Frauenfeld, 1865), which is classified in the family Viviparidae Gray, 1847. Although representatives of the family Viviparidae have been well-studied in India and Burma [Brandt, 1974], investigation of this species has been neglected in Thailand. All Thai genera were previously classified using conchological, radula, and embryonic characteristics [Brandt, 1974] and subsequently by genetic criteria and morphological characteristics [Tarbsripair, 1998].

The presence of sexual dimorphism in snails belonging to family Viviparidae was noted only in the right tentacles of males that are modified into a copulatory organ [Brandt, 1974; Tarbsripair, 1998]. However, the sexual dimorphism in shell morphology of this species was not mentioned in the available literature. The objective of this study was to verify statistically if specimens of F. martensi martensi
with relatively larger and heavier shells are females, and whether snail sex can be predicted using shell characteristics alone.

Materials and methods

The specimens of *F. martensi martensi* were collected by hand in September 2009 from a riverbed consisting of gravels and broken concrete near a bank of the Kwai Yai River in the Plant Genetic Protection Area, Srinagarind Dam, Sisawat District, Kanchanaburi Province (14°23'10.3'' N, 99°08'33.7''E). To gain more reliable statistical analysis, we decided to do additional snail collection on 30 July 2020 (14°22'48.7'' N, 99°08'44.4''E). In total, 104 adult snails (52 females and 52 males) were randomly selected to test the hypothesis of sex-related differences in the laboratory. We classified snails as adult based on their shell size i.e. on assumption that shells width ca. 20 mm and shell length ca. 30 mm belong to mature individuals [Brandt, 1974]. We excluded subadult shells from our analyses in order to avoid size discrepancy resulting from age variation and other causes rather than sexual dimorphism. Subadult shells are green to olive green in contrast to brown to black in adult shells [Tarbsripair, 1998]. Piatiratitivorakula, Boonchamoi, 2008]. *Filopaludina* snails are dioecious, having the male and female reproductive organs in separate individuals. Males have a modified right tentacle which acts as a copulatory organ and can be readily identified. The right tentacle is often longer and curved in contrast to the slender and straight shorter left tentacle of the males and both tentacles of the females [Tarbsripair, 1998]. Females are ovoviviparous. Mature females produce eggs that hatch internally, thereby carrying embryos in the uterine brood pouch before giving birth to live young [Jokinen et al., 1982; Stanisic, 1998]. Sex determination was based on the appearance of the modified right tentacles in males as well as the appearance of embryos in the brood pouches of females. Sex identification was confirmed by exposing the soft bodies after boiling the snails in hot water for 30 minutes and checking the reproductive anatomy. Once the soft bodies were removed, shells and opercula were measured with a Vernier caliper to the nearest 0.01 mm using the measurement scheme modified from Cazzaniga [1990] and Andreeva et al. [2017]. Finally, the shells and opercula were weighed to the nearest 0.01 g. Six continuous shell and operculum characteristics were measured (Fig. 1).

General descriptive statistics were performed for both male and female shells separately for all characteristics. Student’s t-test was used to determine the significance of characteristics exhibiting sexual...
Sexual dimorphism and morphometric analysis of a viviparid snail

Results

Eight quantitative shell characteristics except three derived characteristics (i.e., SH/SW, AH/AW, and OH/OW) were significantly different between males and females (Table 1). Females possessed longer and wider shells, longer and wider aperture, longer and wider opercula, and heavier shells and opercula than males. PCA analysis revealed that 79.67% of the observed variations could be explained by the first three factors (PC1=50.59%, mostly summarizing variation in general shell size, apertural size, and opercular size; PC2=17.1% mostly reflecting shell height to shell width ratio and shell width; PC3=11.98% mostly reflecting aperture height to aperture width ratio; Table 2).

A plot of individual component scores along the first principal component (x-axis) and the second principal component (y-axis) showed two clusters of individuals overlapping greatly in the four graph quadrants. Male cluster shifted toward the 2nd and 3rd quadrants, while females were mainly clustered in the 1st and 4th quadrants (Fig. 2).

Discussion

Sexual dimorphism involving shell and operculum sizes of *F. martensi martensi* was observed in our study. The earlier studies such as Baker [1928] and Thiele [1929] found similar observation that snails belonging to family Viviparidae are sexually dimorphic. In this study, a consensus of morphologi-

Table 1. Comparative characteristics of shell and operculum quantitative traits of males and females of *F. martensi martensi*.

<table>
<thead>
<tr>
<th>Measurement/ index</th>
<th>Females (n=52)</th>
<th>Males (n=52)</th>
<th>Significance of differences between means (Student’s t-test)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Range</td>
<td>Mean±SD</td>
<td>Range</td>
</tr>
<tr>
<td>Shell height (SH)</td>
<td>33.25-43.45</td>
<td>38.24±2.44</td>
<td>30.50-39.47</td>
</tr>
<tr>
<td>Shell width (SW)</td>
<td>24.61-33.70</td>
<td>28.40±2.56</td>
<td>20.13-30.40</td>
</tr>
<tr>
<td>SH/SW</td>
<td>1.21-1.59</td>
<td>1.35±0.12</td>
<td>1.20-1.83</td>
</tr>
<tr>
<td>Aperture height (AH)</td>
<td>17.60-23.50</td>
<td>20.49±1.63</td>
<td>16.70-29.21</td>
</tr>
<tr>
<td>Aperture width (AW)</td>
<td>14.30-19.87</td>
<td>17.23±1.43</td>
<td>14.50-19.71</td>
</tr>
<tr>
<td>AH/AW</td>
<td>1.10-1.30</td>
<td>1.19±0.05</td>
<td>1.07-1.81</td>
</tr>
<tr>
<td>Operculum height (OH)</td>
<td>14.60-19.59</td>
<td>17.19±1.10</td>
<td>14.00-19.43</td>
</tr>
<tr>
<td>Operculum width (OW)</td>
<td>11.65-16.33</td>
<td>14.34±0.90</td>
<td>10.85-16.30</td>
</tr>
<tr>
<td>OH/OW</td>
<td>1.13-1.28</td>
<td>1.20±0.03</td>
<td>1.10-1.29</td>
</tr>
<tr>
<td>Shell weight (SWe)</td>
<td>2.12-5.74</td>
<td>3.82±0.88</td>
<td>1.72-5.18</td>
</tr>
<tr>
<td>Operculum weight (OWe)</td>
<td>0.03-0.23</td>
<td>0.12±0.04</td>
<td>0.03-0.18</td>
</tr>
</tbody>
</table>

Table 2. Factor loadings for the first three principal axes of a PCA based on eight shell and operculum measurements of *F. martensi martensi*. Characteristics with highest scores are shown in bold.

<table>
<thead>
<tr>
<th>Character</th>
<th>PC1</th>
<th>PC2</th>
<th>PC3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Shell height (SH)</td>
<td>0.94</td>
<td>0.00</td>
<td>-0.01</td>
</tr>
<tr>
<td>Shell width (SW)</td>
<td>0.47</td>
<td>-0.83</td>
<td>0.22</td>
</tr>
<tr>
<td>SH/SW</td>
<td>0.23</td>
<td>0.90</td>
<td>-0.27</td>
</tr>
<tr>
<td>Aperture height (AH)</td>
<td>0.80</td>
<td>0.29</td>
<td>0.44</td>
</tr>
<tr>
<td>Aperture width (AW)</td>
<td>0.85</td>
<td>0.13</td>
<td>-0.23</td>
</tr>
<tr>
<td>AH/AW</td>
<td>0.11</td>
<td>0.25</td>
<td>0.89</td>
</tr>
<tr>
<td>Operculum height (OH)</td>
<td>0.92</td>
<td>0.15</td>
<td>-0.00</td>
</tr>
<tr>
<td>Operculum width (OW)</td>
<td>0.94</td>
<td>0.05</td>
<td>-0.13</td>
</tr>
<tr>
<td>OH/OW</td>
<td>-0.00</td>
<td>0.26</td>
<td>0.33</td>
</tr>
<tr>
<td>Shell weight (SWe)</td>
<td>0.78</td>
<td>-0.33</td>
<td>-0.06</td>
</tr>
<tr>
<td>Operculum weight (OWe)</td>
<td>0.83</td>
<td>-0.15</td>
<td>-0.14</td>
</tr>
<tr>
<td>Eigenvalue</td>
<td>5.57</td>
<td>1.88</td>
<td>1.32</td>
</tr>
<tr>
<td>Cumulative variance (%)</td>
<td>50.59</td>
<td>67.69</td>
<td>79.67</td>
</tr>
</tbody>
</table>
cal differences between males and females (Table 1) confirmed that shells of females were relatively larger than those of males. Tarbsripair [1998] studied systematic relationships of Thai viviparid snails of the genus *Filopaludina* using comparative morphology, anatomy and biochemistry but failed to show sexual dimorphism because of a lack of morphometric comparison between male and female individuals. In spite of sexual difference, sex identification of empty shells using shell and operculum characters is difficult since the values of all measured parameters overlap.

*Filopaludina martensi martensi* is one of a few species of viviparid snails in which a sex difference has been observed. Similar shell dimorphism was also observed in other viviparid snails such as *Lioplax subcarinata occidentalis* Pilsbry, 1935 [Cleave, Chambers, 1935], *Viviparus georgianus* (I. Lea, 1834) [Jokinen *et al.*, 1982], *Sinotaia angularis* (O.F. Müller, 1774) [Pagulayan, Cepillo, 1991], *Cipangopaludina chinensis* (Gray, 1833) [Jokinen *et al.*, 1982; Chiu *et al.*, 2002], *Viviparus subpurpureus* (Say, 1829) [Minton, Wang, 2011], and *V. viviparus* (Linnaeus, 1758) [Berezkina, Zotin, 2013]. Other gastropod examples are *Ficus subintermedia* (d’Orbigny, 1852) [Arakawa, Hayashi, 1972], *Melanoides tuberculata* (O. F. Müller, 1774) [Brandre *et al.*, 1996], *Pomacea canaliculata* (Lamarck, 1819) [Estebelet, 1998], and *Olivella plata* (Ihering, 1909) [Pastorino, 2007].

FIG. 2. Principal component analysis of eight shell and operculum characteristics from 104 specimens representing males and females of *F. martensi martensi*. The first two axes (PC1 and PC2) represent 67.69% of the variations. The encircled surfaces represent 95% confidence level of the sample means with the center of each cluster marked by a larger symbol. Factor loadings are presented in Table 2.

РИС. 2. Анализ главных компонент восьми признаков раковины и крышки 104 экземпляров самцов и самок *F. martensi martensi*. Перые две оси (PC1 и PC2) объясняют 67.69% дисперсии. Обозначенные эллипсами области соответствуют 95% доверительному интервалу, центр каждого кластера обозначен более крупным значком. Факторные нагрузки представлены в Табл. 2.
The morphological variation documented here may represent a sexual dimorphism of subspecies *F. martensi martensi* because shells and opercula of adult snails were analysed. Thus, we have reason to believe that morphological difference results from sexual dimorphism. Sexual dimorphism is expressed in the development of distinctive shell features that are generally related to reproduction or spawning in viviparid snails [Pastorino, 2007]. In viviparid snails, females bear eggs and brood embryos inside the uterus brood pouch until they are fully developed. Females usually have a larger body volume than males when mature because body volume relates directly to fecundity [Jakubik 2006; Boeckstael, Strong, 2016].

The study was approved by Mahidol University-Institute Animal Care and Use Committee (MU-IACUC): Protocol No. F01-64-001

Acknowledgements

The authors express gratitude to Mahidol University, the Plant Genetic Protection Project under the Royal Initiative of Her Royal Highness Princess Maha Chakri Sirindhorn at Sirinaradit Dam, and the Electricity Generating Authority of Thailand (EGAT) for financial support.

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Pastorino G. 2007. Sexual dimorphism in shells of the southwestern Atlantic gastropod *Olivella plata*


