

Native Indonesian pond mussel *Pilsbryoconcha exilis* and alien *Sinanodonta woodiana*: the potential effect on the future conservation and economic benefit

¹Mochamad C. W. Arief, ^{1,2}Asep Sahidin, ¹Izza M. Apriliani, ¹Isni Nurruhwati, ¹Zahidah Zahidah, ²Gunawan Muhammad, ²Akira Komaru

¹ Department of Fisheries, Faculty of Fisheries and Marine Sciences, Universitas Padjadjaran, Sumedang, West Java, Indonesia; ² Graduate School of Bioresources, Mie University, Mie Prefecture, Tsu, Japan. Corresponding author: M. C. W. Arief, mochamad.candra@unpad.ac.id

Abstract. *Sinanodonta woodiana*, a rapid-growth alien species, coexists with native *Pilsbryoconcha exilis* in the habitats of Indonesia. The biological aspect of biometrics, biomass, and growth was investigated to predict the potential invasiveness of *S. woodiana*, and nacre color was studied to predict economic benefits and to give recommendations for managing both species in nature. The growth analysis showed an allometric negative growth for both species, with the growth ratio of *S. woodiana* being higher than that of *P. exilis*. Similarly, the shell size was longer, and the biomass was heavier for *S. woodiana* than for *P. exilis* (p<0.001). *S. woodiana* had a higher index of convexity and elongation of the shell. Consequently, the species has the potential for invasiveness in the future if it is uncontrolled. Fortunately, the species has a good nacre color to encourage the economic potential for freshwater pearls. Hence, the study recommends community-based management with economic benefits in harvesting periodically to control alien species in the environment.

Key Words: biodiversity, clam, freshwater pearl, Unionid.

Introduction. Indonesia has a mega-diversity of freshwater fauna and is an endemic hotspot, including for freshwater bivalves (Graf & Cummings 2021). Freshwater bivalves are essential in maintaining the freshwater environment, with roles in the ecological balance (Reid et al 2019), ecosystem engineering as biofiltration, sediment oxygenation, and water pollution bioindicators (Zieritz et al 2016), and energy transfer from pelagic to benthic zones (McCasker & Humphries 2021). On the other hand, freshwater mussels are endangered aquatic animals because their habitat is directly influenced by anthropogenic activity (Zieritz et al 2018; Do et al 2019; IPBES 2019), have slow movement (Böhm et al 2020), and are food for many predators, including mammals, birds, reptiles, and fish (Vaughn 2018).

Freshwater mussels (family: Unionid) are the most diverse freshwater bivalves, with 26 species, 45.6% of total freshwater bivalves, found in Indonesia. 12 species are included as endemic and eight as native species. Recently, six alien species were found in Indonesia (Sahidin et al 2021). Aquatic alien species are aquatic organisms that become established beyond their habitat and can increase biodiversity in the new habitat. However, alien species can bring negative impacts on native species, for example, regarding dwelling (Emery-Butcher et al 2020), feeding (Zieritz et al 2016), and reproduction (Tiemann et al 2022), possibly resulting in a decline in the population of a native species in the habitat.

Of particular concern is *Sinanodonta woodiana*, an alien species in Indonesia. This species was initially distributed in the Amur and Yangtze Rivers (southeast China) and expanded worldwide, for example, in East Asia, Russia, Europe, and the USA ()Lopes-Lima et al 2014, including Indonesia (Bolotov et al 2016). Historically, for over 50 years,

S. woodiana has been rapidly expanding in Indonesia after accidentally being introduced in 1969 with tilapia imported from Taiwan (Djajasasmita 1982). A recent study noted that this species has adapted to various habitats from low elevations to over 1500 m above sea level (Sahidin et al 2022). Interestingly, *S. woodiana* is found living together with *Pilsbryoconcha exilis* in a single habitat, such as in fishponds and in certain lakes and rivers in Indonesia (Sahidin et al 2021). *P. exilis* is a native freshwater mussel species in Indonesia, spreading on the main island of Java, Sumatra, and Kalimantan (Sahidin et al 2021). The growth and habitat competition between the two species will affect future conservation.

The growth of an individual mussel is frequently assessed by correlating the evolution of the largest dimension of the individual over time (Caill-Milly et al 2012). The growth of mussels can be represented by biometrics, biomass, and shell shape (Laing 2004). Hence, the growth can predict the competitive strength in the habitat and the conservation status for the future of native and non-native species in the habitat. The shell size of mussels is correlated with the size of the living area is and the quantity of food they need for growth (Caill-Milly et al 2012). In addition, the freshwater mussel has a nacre layer in the inner shell, which can produce pearls that give economic potential to the surrounding community (FAO 2020). This study aims to: 1) analyze the growth, 2) compare biometrics and biomasses of *P. exilis* and *S. woodiana* for future conservation treatment, and 3) analyze the inner shell nacre color for future economic potential.

Material and Method

Mussel shell samples collection. 354 freshwater pond mussel shells were collected from Ciparanje Fisheries Experimental Fishpond at Universitas Padjadjaran, Indonesia (6°54'39"S and 107°46'12"E) from November 2020 to October 2021 and in June 2022. The samples were divided into native mussel *P. exilis* (114 samples) and alien mussel *S. woodiana* (240 samples). Biometric variables including shell length (*L*), height (*H*), and width (*W*), were measured with the digital Mitutoyo caliper, Japan (nearest to 0.01 mm) (Figure 1). The total mussel weight (*Tw*), shell weight (*Sw*), and soft tissue weight (*STw*) were measured using a digital scale (nearest to 0.01 g). The right valve of the clean and dry shell was pointed and marked (1 cm²) in five positions, the posterior, dorsal, anterior, center, and ventral regions (Figure 1). The marked shell at each position was cut using a Struers Discoplan-TS cutting machine.



Figure 1. A and B - biometric measurement of the shell; C - sampling positions on the inner shell surface. *L* - shell length; *W* - width; *H* - height; an - anterior; do - dorsal; po - posterior; ce - central; ve - ventral.

Estimation of morphological shell index and relative growth. Based on shell biometric characteristics, two morphological shell indexes, the shell convexity index (SCI) and the shell elongation index (SEI), were measured using the formula (Bolotov et al 2018):

SCI=(shell width/shell length)*100

SEI=(shell height/shell length)*100

The relative growth was determined using the relationship between L (x variable) and other biometrics (i.e., H and W, as y variable) with linear functions ($y=ax^b$). However, the relationship between L (x variable) and mass (i.e., Tw, Sw, and STw; as y variable) was used in logarithmic functions (Log $y=b \log x + \log a$), where "a'' is intercept and "b'' is coefficient of relative growth (allometry). The growth type, according to the linear function, may be negatively allometric (a<1), positively allometric (a>1), and isometric (a=1). At the same time, the growth type according to the logarithmic function may be negatively allometric (a>3), positively allometric (a>3), and isometric (a=3) (Caill-Milly et al 2012).

Inner shell color estimation. A total of 145 pieces of shell (1 cm^2) from five positions in the posterior, dorsal, anterior, center, and ventral parts of 29 shells (right valves) selected from 234 mussels were used for color estimation. *P. exiles* had 14 shells, with 70 pieces, and *S. woodiana* 15 shells, with 75 pieces. The color of the inner shell in each piece was observed using a CM-700d spectrophotometer (Konica Minolta, Japan) for giving CIE tristimulus (XYZ) values. The specific body color was calculated using the YXY color space following the procedure by Sahidin et al (2022), while the color saturation was estimated from the L*a*b* 3D CIE graph (Wang et al 2020). L* (lightness) is the vertical axis ranging from 0 (black) to 100 (white); a* represents the red to the green axis (-a* represents the green saturation and +a* represents the red saturation); b* represent the blue to the yellow axis (-b* represents blue saturation and +b* represents yellow saturation). In addition, we calculated the yellowness index (YI) to denote gold or silver color in the inner shell (Henderson & Manchanda 2005; Sahidin et al 2022).

Statistical analysis. Shell biometrics of mussels, such as *L*, *W*, *H*, *Sw*, *STw*, and *Tw*, were correlated between species using the Pearson correlation analysis. Similarly, the shell morphological index and relative growth of the shell were compared between species. Body color and color saturation were compared between positions in the anterior, dorsal, posterior, center, and ventral and between species. All comparisons used a Kruskal-Wallis test followed by a post-hoc Pairwise Mann-Whitney U test. All analyses used RStudio open sources software, following codes available from Albert & Rizzo (2012), Chang (2013), and Long & Teetor (2019).

Results and Discussion

Morphometrics and biometrics of freshwater mussels. The length of *P. exilis* was significantly longer than that of *S. woodiana* (p<0.001; Table 1), but shell height and width of *P. exilis* were significantly lower than those of *S. woodiana* (p<0.001; p<0.01, respectively; Table 1). The shell weight (*Sw*) of *P. exilis* was higher than that of *S. woodiana*, in contrast with the total weight (*Tw*), which was lower for *P. exilis* than for *S. woodiana* (p<0.001, p<0.01, respectively; Table 1). The spectively; Table 1). The weight (*Sw*) of *P. exilis* was lower for *P. exilis* than for *S. woodiana* (p<0.001, p<0.01, respectively; Table 1). The weight (*STw*) presented no significant differences (t=-0.79, p>0.05).

Table 1

Mean of shell biometrics and weights of Pilsbryochonca exilis and Sinanodonta woodiana

| Shell variables | P. exilis | | | | S. woodiai | t-test | p- value | |
|--------------------|-----------|------------|----------|-----|------------|-----------------------|-------------|------|
| | n | range | mean±SD | n | range | <i>mean<u>+</u>SD</i> | | |
| L (mm) | 114 | 76.1-107.2 | 89.2±5.5 | 120 | 47.2-111.4 | 76.8±13.5 | 8.64 | *** |
| W (mm) | 114 | 17.2-25.3 | 20.4±1.7 | 120 | 20.1-44.3 | 33.4±5.8 | -22.67 | *** |
| H (mm) | 114 | 36.4-56.5 | 45.6±3.5 | 120 | 33.1-68.2 | 50.6±7.9 | -7.12 | ** |
| Sw (g) | 114 | 8.8-28.4 | 15.2±3.2 | 120 | 3.5-31.2 | 11.3 ± 5.2 | 7.13 | ** |
| STw (g) | 114 | 8.7-22.2 | 14.9±2.6 | 120 | 1.3-42.5 | 15.4 ± 7.0 | -0.79 | n.s. |
| Tw (g) | 114 | 20.5-74.1 | 44.1±8.7 | 120 | 12.2-131.3 | 50.7±23.5 | -2.78 | ** |

Note: L - length; H - height; W - width; Sw - shell weight; STw - soft tissue weight; Tw - total weight; n - number of samples; *** - p<0.001; ** - p<0.01; n.s. - not significant (p>0.05).

Morphometric and biometric relationships of freshwater mussels. The relationship of the morphometric between various characteristics shell length, shell width, shell height, shell wet mass and wet mass of soft tissue in both species of freshwater mussels are presented in Figure 2. The SCI of *S. woodiana* was significantly greater than that of *P. exilis* (66.2±4 and 50±2.7, respectively; p<0.001). The relationship between SCI and SEI showed that *S. woodiana* had significantly higher values than *P. exilis* (Figure 3A).



Figure 2. Shell morphometry of *Pilsbryochonca exilis* (n=114) and *Sinanodonta woodiana* (n=120). Scatterplot correlation: A) shell length vs shell height; B) shell length vs shell width; C) shell length vs wet shell mass; D) shell height vs wet shell mass; E) shell length vs wet shell mass; F) shell length vs wet mass of soft tissue; ** - p<0.01; *** - p<0.001.</p>

Relative growth of freshwater mussels. *W* and *H* and *Sw*, *STw*, and *Tw* were significantly correlated with *L*, with a correlation coefficient r between 0.59-0.95 (p<0.001). Furthermore, the growth analysis showed that *W* and *H* had negative allometries with regard to *L* (Table 2). Similarly, masses growth in *Sw*, *STw*, and *Tw* had negative allometries with *L* (Table 2). It means the *L* grew faster than *W* and *H* for both species. *L* grew faster than variables in *Sw*, *STw*, and *Tw* (Table 2). The growth rate of *S*. woodiana was faster than that of *P*. exilis (Table 2).



Figure 3. The shell morphological index of *Pilsbryochonca exilis* and *Sinanodonta woodiana*; A) shell elongation index vs shell convexity index scatterplot; B) statistical comparison of shell convexity index; and C) statistical comparison of shell elongation index; *** - p<0.001.

Table 2

Relationships between relative growth variables of native and exotic Indonesian freshwater mussels, *Pilsbryochonca exilis,* and *Sinanodonta woodiana*, from West Java, Indonesia

| Species | <i>Biometric variables</i> | <i>Ratio±SD</i> | $y=ax^b$ log y = b log x + log a | r | t-test | p-value | Growth type |
|--------------------------|--------------------------------|-----------------|---|------|--------|---------|----------------|
| | H with L | 0.5±0.03 | 2.019 x ^{0.478} r=0.75 | 0.75 | 11.11 | *** | - |
| | W with L | 0.23±0.02 | 4.263 $x^{0.182}$ r=0.59 | 0.59 | 7.39 | *** | - |
| Pilsbryochonca exilis | Sw with L | 0.17±0.03 | $\log y = 2.414 \log x - 3.537$ r = 0.73 | 0.73 | 11.17 | *** | - |
| | STw with L | 0.16±0.02 | log y=2.302 log x - 3.323 r=0.79 | 0.79 | 13.23 | *** | - |
| | <i>Tw</i> with <i>L</i> | 0.49±0.07 | log y=2.748 log x - 3.724 r=0.73 | 0.73 | 16.11 | *** | - |
| | H with L | 0.66±0.04 | 7.403 x ^{0.563} r=0.95 | 0.95 | 31.70 | *** | - |
| | W with L | 0.44±0.05 | 5.665 $x^{0.363}$ r=0.83 | 0.83 | 15.05 | *** | - |
| Sinanodonta woodiana | Sw with L | 0.16±0.04 | log y=2.449 log x - 3.534 r=0.91 | 0.91 | 21.90 | *** | - |
| | STw with L | 0.19±0.06 | log y=2.321 log x - 3.223 r=0.86 | 0.86 | 13.33 | *** | - |
| | Tw with L | 0.63±0.19 | $\log y = 2.450 \log x - 2.904$ r = 0.84 | 0.84 | 22.21 | *** | - |

Note: L - shell length; W - shell width; H - shell height; Tw - total wet mass; STw - soft tissue wet mass; Sw - shell wet mass; N - number of individuals; (-) - negative allometric growth; T-test analysis with significant differences for p < 0.001 - ***.

Freshwater mussel shell color variation. Visually, the inner shell surfaces in both freshwater pond mussels, *P. exilis* and *S. woodiana*, displayed various colors, as presented in Figure 4. CIELAB YXY color space analysis showed that the *S. woodiana* inner shell appearance has 3 colors: golden, yellow, and silvery (Sahidin et al 2022). The shell color of *P. exilis* was silvery with little yellow (92.8% and 7.2%, respectively) (Figure 5A). Body-color analysis using the yellowness index (YI) value also showed the same trend, with *S. woodiana* having higher YI values than *P. exilis* (21.45±27.39 and 11.05±12.33, respectively; p<0.05; Table 3). However, in contrast, the hue (h*) and chromatic (c*) values of *S. woodiana* were lower than those of *P. exilis* (p<0.05; Table 3).



Figure 4. The inner shell color variation of *Sinanodonta woodiana* (A, B, and C) and *Pilsbryochonca exilis* (D and E).

Furthermore, the color saturation distribution by CIELAB 3D L*a*b* analysis was dominated by yellow (+b*) and green (-a*) color saturation, while the blue (-b*) and red (+a*) color saturation displayed a small bit portion (Figure 5B). The +b* value in *S. woodiana* inner shell was significantly higher than that of *P. exilis* (13.59±8.86, 5.89±3.74, respectively; p<0.05). In contrast, the +a* value of *S. woodiana* inner shell was significantly lower than that of *P. exilis* (0.65±0.51, 1.19±0.84, respectively; p<0.05) (Table 3). The -a* and -b* values were not significantly different between *S. woodiana* and *P. exilis* (p>0.05; Table 3). The vertical axis in lightness (L*) of *S. woodiana* was higher than that of *P. exilis* (p<0.05; Table 3).

Inner shell color in different areas of the shell. YI, c*, and b* values in *S. woodiana* gradually increased in the anterior, dorsal, and posterior areas and greatly decreased in central and ventral positions. However, this was not the case for *P. exilis*, where YI, c*, and b* had constant values between anterior, dorsal, posterior, central, and ventral positions (Figure 6A-B, F). In both species, the h* value in the anterior, dorsal, and posterior areas had a constant value and gradually increased in the central and ventral positions. However, the L* value was lowest in the central area in both species and highest in the anterior area for *S. woodiana* and in the posterior area for *P. exilis* (Figure 6C-D). In contrast, the a* value of *P. exilis* was higher than that of *S. woodiana* in posterior, central, and ventral parts of the shell, excepting the dorsal and anterior positions.



Figure 5. Color distribution in the inner shell of *Pilsbryochonca exilis*; A) body color; B) illumination color; C) illumination color of *Sinanodonta woodiana*.

Table 3

Inner shell color variation of Sinanodonta woodiana and Pilsbryochonca exilis

| Color | | P. exi | lis | | S. woodiana | | | |
|-----------|----|---------------|-----------------|----|--------------|-----------------|---------------|--|
| variables | n | mean ± SD | range | n | mean ± SD | range | <i>t-lest</i> | |
| ΥI | 70 | 11.05±12.33 | (-25.37)-35.81 | 75 | 21.45±27.39 | (-22.37)-79.48 | * | |
| h* | 70 | 119.92±56.76 | 22.25-357.76 | 75 | 137.87±67.45 | 82.5-330.71 | * | |
| с* | 70 | 5.9±3.6 | 0.23-14.93 | 75 | 12.09±12.58 | 0.85-98.36 | * | |
| L* | 70 | 69.46±3.65 | 57.16-77.71 | 75 | 75.86±4 | 68.09-84.27 | * | |
| a* | 16 | 1.19 ± 0.84 | 0.03-2.67 | 16 | 0.65±0.51 | 0.1-1.57 | * | |
| -a* | 54 | (-1.08)±0.89 | (-0.01)-(-4.72) | 59 | (-1.55)±1.15 | (-5.69)-(-0.02) | n.s. | |
| b* | 62 | 5.89±3.74 | 0.12-14.7 | 56 | 13.59±8.86 | 0.37-36.79 | * | |
| -b* | 8 | (-3.81)±3.33 | (-0.05)-(-8.7) | 19 | (-2.54)±1.81 | (-0.21)-(-6.17) | n.s. | |

Note: YI - yellowness index; h* - hue; c* - chromatic; L* - lightness; a* - red color saturation; -a* - green color saturation; b* - yellow color saturation; -b* - blue color saturation; * - significance at p<0.05; n.s. - not significant (p>0.05).



Figure 6. Color distribution in the inner shell surface at anterior (an), dorsal (do), posterior (po), central (ce), and ventral (ve) areas; A) yellowness index; B) chromatic; C) hue; D) lightness; E) the value in color saturation on the red and green axis; F) b in color saturation on the blue and yellow axis; * - significant difference at p<0.05 between *Pilsbryochonca exilis* and *Sinanodonta woodiana* inner shell color.

The potential effect of aliens on native species in a habitat. Globally, *S. woodiana* is an invasive freshwater mussel in many regions, i.e., the United States of America (Sousa et al 2014), Europe (Urbańska et al 2019), Russia, and Southeast Asia (Bolotov et al 2016). Commonly, *S. woodiana* plays a role as a fish parasite when it is in the glochidium larva stage (Lima et al 2006; Douda et al 2012; Kovitvadhi & Kovitvadhi 2012), being harmful to fish cultures. Unfortunately, this species has not yet been assessed for the potential prediction of invasiveness in Indonesia. Our research found that the body size parameters of *S. woodiana* were significantly larger than those of *P. exilis*. We argue that the shell size might affect the housing competition in the habitat. Hence, with a continuous increase in numbers, it is predicted that it might occupy the habitat of the native species. Accordingly, further studies are needed to compare population dynamics, reproduction, fish hosts, and others to obtain a larger perspective.

Moreover, the mussel's body size and weight is a directional measurement predictor of the food quantity needed (Coughlan et al 2021). Biologically, a large body size needs more feed and *S. woodiana* will intake more feed. Both species will compete for food to survive in their habitat. Whoever is more responsive will survive in the habitat.

S. woodiana is a highly tolerant mussel to organic pollution (Kolarević et al 2011; Sousa et al 2014). Consequently, we argue that *S. woodiana* is more resilient than the native species *P. exilis* in the habitat. If *S. woodiana* continues to increase in population, we predict that it might result in the exclusion of the native species from the habitat. To get a more comprehensive discussion, further studies are required. **The economic benefit and future conservation of freshwater mussels**. The potency of freshwater mussel utilization in Indonesia is diverse. They can reduce water pollution as a biofilters (Putra et al 2016; Yulianto et al 2019). They have a good nutritional quality (Nurjanah et al 2020) and can increase the diversity of food products for human consumption (Ghazali et al 2015; Nur'aini et al 2019; Mustaring et al 2020). These utilizations are only partially developed and have a lower contribution to the economy. Furthermore, the utilization needs a good management the sustainability and health of the ecosystem.

In addition, some freshwater mussels may have a nacre layer in the inner shell. Species with this characteristic are: *Margaritifera margaritifera* (Hastie et al 2000), *Hyriopsis cumingii* (Bai et al 2017), *Lasmigona costata* and *Strophitus undulatus* (Fichtel & Smith 1995), *Anodonta anatine, A. cygnea, Unio pictorum*, and *U. tumidus* (Urbańska et al 2019). The nacre in the inner shell indicates they can produce pearls with economic potential. A previous study shows that *S. woodiana* has various nacre colors, increasing half pearl quality (Sahidin et al 2022), but *P. exilis* has not yet been assessed. In the present study, we found that the nacre colors of *P. exilis* were yellow and silvery. In contrast, in the previous study, the nacre colors of *S. woodiana* were gold, yellow, and silvery (Sahidin et al 2022). Based on our result, the color quality of the nacre surface of the inner shell (in yellowness index and brightness) of *S. woodiana* is better than that of *P. exilis*. The yellowness index value is used as an indicator of gold color expression on the nacre surface (Henderson & Manchanda 2005). Jewelry with a gold color is highly marketable in Indonesia (Sahidin et al 2021).

Brightness is another important determinant of freshwater pearl quality and economic value (Wang et al 2020). The term brightness refers to the perception of how much light reflects off something (Stenger et al 2019) and expresses the mirroring and shininess of pearls (Nagata et al 1997). In practice, luster is interpreted as the relative brightness of diffuse reflecting areas (Salyacheewin et al 2022). Our results indicate that *S. woodiana* has better luster and brightness, resulting in high-quality half-pearls. In addition, our study also analyzed the nacre microstructure *S. woodiana*, which has a thinner whole nacre layer, large nacre tablet area, and thick nacre tablet to produce the high-quality nacre in the pearls. Accordingly, we speculate that *S. woodiana* has a higher potential to produce better pearls for commercial culture than *P. exilis*.

For over 50 years, *S woodiana*, after accidentally being introduced in 1969 with tilapia and silver carp imported from Taiwan (Djajasasmita 1982), they have been widely distributed in the main islands of Indonesia (Sahidin et al 2021). They also spread across different elevations (Sahidin et al 2022). People do not harvest it from natural habitats because they assume it is not economically beneficial. Consequently, this species had a rapid growth in Indonesia, especially on Java Island (Sahidin et al 2021). We assume that *S. woodiana* has invasiveness properties and will eliminate native species in the future. It is dangerous to the Indonesian freshwater ecosystem because biological invasions are a significant component of global change, imposing a serious threat to the conservation of biodiversity (Sousa et al 2014). Recently, invasive species management and control is the biggest challenge faced by conservation biologists and there is increasing pressure on policymakers to regulate and mitigate this component of global change.

Accordingly, we recommend periodical harvesting of alien species from natural habitats as management actions. Thus, the population of this species could be restrained to reduce the negative impact, the ecosystem remaining stable. In addition, we also recommend producing freshwater pearls from this species to add economic benefit and maintain continuous regular harvesting (6 or 12 months). Thus, the species would not be overexploited from nature. In addition, prevention, relying on public awareness, legislation, and monitoring, tends to be the most cost-effective and ecologically viable measure to minimize the spread of *S. woodiana*. Integrated research, management, and implementation at the national and regional levels are required strategies to control this species in the Indonesian freshwater ecosystem.

Conclusions. *S. woodiana* had a higher growth rate compared to *P. exilis* indicated by the morphometric and biometric characteristics. This could indicate the potential threat of the invasiveness character of *S. woodiana*. Even though *S. woodiana* has potential economic benefits, its invasiveness in the native species' habitats should lead to some management measures, like periodic harvesting. Furthermore, our study presents information on biometrics and nacre color in both species, which may serve as a basis for future studies.

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Conflict of Interest. The authors declare that there is no conflict of interest.

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Mochamad Candra Wirawan Arief, Department of Fisheries, The Faculty of Fisheries and Marine Sciences, Universitas Padjadjaran, Jl. Bandung-Sumedang Km 21, Sumedang Regency, 45363 West Java, Indonesia, e-mail: mochamad.candra@unpad.ac.id

Asep Sahidin, Department of Fisheries, The Faculty of Fisheries and Marine Sciences, Universitas Padjadjaran, Jl. Bandung-Sumedang Km 21, Sumedang Regency, 45363 West Java, Indonesia, e-mail: asep.sahidin@unpad.ac.id

Izza Mahdiana Apriliani, Department of Fisheries, The Faculty of Fisheries and Marine Sciences, Universitas Padjadjaran, Jl. Bandung-Sumedang Km 21, Sumedang Regency, 45363 West Java, Indonesia, e-mail: izza.mahdiana@unpad.ac.id

Isni Nurruhwati, Department of Fisheries, The Faculty of Fisheries and Marine Sciences, Universitas Padjadjaran, Jl. Bandung-Sumedang Km 21, Sumedang Regency, 45363 West Java, Indonesia, e-mail: isni@unpad.ac.id Zahidah Zahidah, Department of Fisheries, The Faculty of Fisheries and Marine Sciences, Universitas Padjadjaran, Jl. Bandung-Sumedang Km 21, Sumedang Regency, West Java 45363, Indonesia, e-mail: zahidah@unpad.ac.id

Gunawan Muhammad, Graduate School of Bioresources, Mie University, Kurimamachiya-cho 1577, Mie Prefecture, 514-8507 Tsu, Japan, e-mail: gunawan.muhammad@hotmail.co.id

Akira Komaru, Graduate School of Bioresources, Mie University, Kurimamachiya-cho 1577, Mie Prefecture, 514-8507 Tsu, Japan, e-mail: komaru@bio.mie-u.ac.jp

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