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NI Made Suartini
a) Doctoral Program, Faculty of Animal Science, Universitas Udayana, Jln. P.B. Sudirman, Denpasar, Bali-Indonesia
b) Biology Program, Faculty of Mathematics and Natural Sciences, Universitas Udayana. Jl. Raya Kampus Unud, Bukit Jimbaran, Kuta Selatan, Badung 80361, Bali, Indonesia

I GST Nym GDE Bidura
Faculty of Animal Science, Universitas Udayana, Jln. P.B. Sudirman, Denpasar, Bali-Indonesia

Komang Budaarsa
Faculty of Animal Science, Universitas Udayana, Jln. P.B. Sudirman, Denpasar, Bali-Indonesia

NI Wayan SITI
Faculty of Animal Science, Universitas Udayana, Jln. P.B. Sudirman, Denpasar, Bali-Indonesia

Corresponding Author:
NI Made Suartini
a) Doctoral Program, Faculty of Animal Science, Universitas Udayana, Jln. P.B. Sudirman, Denpasar, Bali-Indonesia
b) Biology Program, Faculty of Mathematics and Natural Sciences, Universitas Udayana. Jl. Raya Kampus Unud, Bukit Jimbaran, Kuta Selatan, Badung 80361, Bali, Indonesia

Shell morphology of golden apple snails (*Pomacea canaliculata*) from several rice fields in Bali, Indonesia

NI Made Suartini, I GST Nym GDE Bidura, Komang Budaarsa and NI Wayan SITI

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Abstract
Golden apple snail (*Pomacea canaliculata*) has wide adaptability and rice fields are ones of the habitats for the snail. Golden apple snail has phenotypic variations. This study conducted to determine the morphology of golden apple snail from several rice fields in Bali, Indonesia. Samples were taken at twelve locations in three districts. Three rice fields were taken in each location. Samples were taken in quadrate plot of 1x1m at four points each rice field. The morphological of the golden apple snail samples were observed. The results showed that the golden apple snail shells found in all locations has a variety of colors. The color was yellowish to dark brown. The shell also has a shell band and the shell band vary in their appearance. The color of the shell band was more visible on the darker shell. Range of shell heights found in all locations was 8.7-61.5 mm. Most of the golden apple snails found in all location was adult snails.

Keywords: Mollusk, phenotypic variations, shell colour, shell band

Introduction
Golden apple snails includes phylum Mollusca, family Ampulariidae and one of its habitats is rice fields. Environmental factors affect the survival of golden apple snails. Temperature is a factor that greatly influences the growth and feeding rate of the snails. The comfortable temperature range for the snails is 28 - 31°C (Memon *et al*., 2011) [7]. Seuffert and Martín (2013) [12] stated that at 15°C and 20°C there was no death in golden apple snails but the growth rate was very low and at 25°C; 30°C; and 35°C, the growth is very fast. Teo (2004) stated that golden apple snails are less resistant to high temperatures, where at temperatures above 33°C, the mortality rate is high. Oosterom *et al*., (2016) [8] stated that golden apple snails showed plasticity in food. The snails are generalist, which can eat vascular plants, detritus and periphyton. Even animal remains (Acarii, Ostracoda and Chironomidae) can be found in the stomach contents analyzed. Rudianto *et al*. (2014) [11] stated that the type of mud substrate affects the density of golden apple snails.

The study of the apple snail shells showed that there were four species of the snails that had spread to Indonesia. The four species are: *Pomacea canaliculata*; *P. insularum*; *P. scalaris*; and *P. paludosa*. Species of *P. canaliculata* have a wider adaptability to various types of inland water habitats compared to the other three species so that they have the widest distribution (Isnaningsih dan Marwoto, 2011) [3]. The snails have a variety of shell shapes which indicate phenotypic plasticity. The snail population develops alternative phenotypes to adapt to fluctuating environmental factors (Madjos and Olive, 2016) [6].

The difference in shell shape between some golden apple snails occurs because of the very large differences in environmental conditions. Shell specimens from outside Java were relatively large compared to specimens collected from areas in Java. The community's habit of herding ducks in the fields was thought to be a factor affecting the size of the golden apple snail. This is because the ducks eat the snails, including the small ones before they develop into larger ones (Isnaningsih dan Marwoto, 2011) [3]. Golden apple snail shell has a variety of colors. The color of the shell was influenced by the presence or absence of dark pigment in the eye (Yusa, 2004). There are also variations in the color, intensity, number and width of the shell band among individuals of the same population. These variations indicate an ontogenetic and ecophenotypic component. The intensity of the shell band color increases after hatching and during growth. (Estebenet *et al*., 2006) [2]. This research was conducted to determine the morphology of golden snails in several rice fields in Bali.

Materials and Methods

Study area

Golden apple snail samples were taken at twelve locations including three districts (Badung, Gianyar and Karangasem districts) from June to August 2020. In each district, the snail was taken at two locations. The sampling locations are presented in Figure 1. The altitude and name of the subak area (irrigation system for rice field on Bali Island) each location is presented in Table 1.

Fig 1: Golden apple snail sampling location

Table 1: The altitude and subak name at each location

Location			Altitude (meter above sea level)	Subak name
No	Villages	Districts		
1	Pererenan	Badung	40	Sepang
2	Cemagi	Badung	32	Cemagi Let
3	Pangsan	Badung	505	Bergiding
4	Petang	Badung	508	Penglumbaran
5	Tulikup	Gianyar	22	Tulikup
6	Ketewel	Gianyar	20	Subhur
7	Sebatu	Gianyar	739	Jasan
8	Taro	Gianyar	801	Pisang kelod
9	Bugbug	Karangasem	60	Mupahang
10	Tumbu	Karangasem	62	Cangah
11	Duda	Karangasem	500	Padang tunggal
12	Duda	Karangasem	546	Padang tunggal

Sampling technique

Golden apple snails were taken at four points in each rice field with a 1x1m quadrate plot on rice plants less than one month old. Small to large sized snails in the quadrate are taken, both those on the surface of the water, in the waters or attached to aquatic plants. Environmental factors at each sampling location were measured including: water temperature, water pH, Dissolved Oxygen (DO) and water depth.

The golden apple snails were cleaned, put in a sample container, taken to the laboratory to be counted, their morphological and morphometric characters were observed. Morphological character data were taken by observing shell morphology, including shell shape and shell band patterns. Morphometric data were taken by measuring the parts of the shell using a caliper. The characters measured were shell height and width, aperture height and width, number of shell rotations, and body whorl height.

Data analysis

Morphological data from all sites were described. Morphometric data were grouped according to Wada *et al.* (2004) [15], that is, a shell height <25 mm is classified as juvenile and a shell height of 25 mm or more is an adult. The data is presented in a table.

Result

Environmental factors that were measured at the time of sampling were within the range of environmental factors needed for golden apple snail life. The range of measured environmental factors is presented in Table 2.

Table 2: Range of environmental factors at each location

Location		Water temperature (ºC)	Water pH	DO (mg/L)	Water depth (cm)
1	Pererenan	25.0-26.0	7.0-7.5	5.9-6.2	6.0-7.5
2	Cemagi	24.0-26.0	6.5-7.0	5.3-6.4	6.0-8.0
3	Pangsan	24.0-25.0	6.5-7.0	8.3-8.9	6.0-10.0
4	Petang	22.0-22.5	6.5-7.0	9.0-10.1	6.5-11
5	Tulikup	24.5-25.0	6.5-7.0	7.0-7.8	4.0-7.8
6	Ketewel	24.5-26.0	6.5-7.0	7.2-8.0	4.5-11.0
7	Sebatu	20.0-20.5	6.5-7.0	6.7-7.6	3.0-10.0
8	Taro	20.5-21.5	6.0-7.0	6.5-9.8	7.5-10.5
9	Bugbug	25.0-26.0	6.5-7.5	4.0-5.7	7.0-8.5
10	Tumbu	26.0-27.0	6.5-7.5	3.9-4.1	3.5-5.0
11	Duda	22.0-23.0	6.5-7.0	8.5-9.0	9.5-12.0
12	Duda	22.0-23.0	6.5-7.0	7.9-8.7	4.5-9.0

There are variations in the color of the golden apple snail shells found in this study. The color variations of the shell are yellowish to dark brown. Another character is the presence of shell band. The shell band is darker than the shell color. The shell is rounded (globose), sipre is rather high, body whorl enlarged with the shoulders rounded, the sutures clear and deep with an angle of <90º, shell turns to the right (dextral), umbilicus is clear, open and deep, aperture is rounded, inner pallial lip is colorless, operculum shell dark brown to blackish. At each location, the three golden snail shell morphology can be found. Figure 2 presented golden apple snalis in rice fields and in Figure 3 presented a morphology of the golden snail shells were found.

Fig 2: Golden apple snails in rice field

Fig 3: The morphology of the golden apple snail shells

Goden apple snail morphometry varies at each location. The smallest shell size was found at the Bugbug location (altitude about 60 m above tsea level) with shell height 8.7 mm, shell width 6.2 mm, shell aperture height 4.3 mm, shell aperture width 0.37 mm and body whorl height 7.8 mm. The largest shell size was found at the Duda location (altitude about 546 m above sea level) with shell height 61.5 mm, shell width 55.2 mm, shell aperture height 45.3 mm, shell aperture width 34.6 mm and body whorl height 54,6 mm. Grouping juvenile and adult snails based on shell height is presented in Table 3.

Table 3: The number of juvenile and adult snails found in each location

Location		Juvenile		Adult		Total juvenile and adult
		Number of individuals	Shell height range (mm)	Number of individuals	Shell height range (mm)	
1	Pererenan	59	19.1-23.0	25	25.6-29.1	84
2	Cemagi	68	14.3-24.6	57	25.0-41.9	125
3	Pangsan	37	13.0-24.9	46	25.0-40.2	83
4	Petang	19	13.0-24.9	25	25.9-39.1	44
5	Tulikup	23	14.1-24.2	43	25.6-51.6	66
6	Ketewel	48	12.6-22.8	59	26.2-52.0	107
7	Sebatu	13	13.2-24.4	24	25.2-40.0	37
8	Taro	20	14.0-24.8	29	25.1-41.0	49
9	Bugbug	52	8.7-22.9	43	26.3-40.2	95
10	Tumbu	30	13.3-23.6	47	25.4-57.6	77
11	Duda	3	23.6-24.8	27	30.5-61.5	30
12	Duda	6	18.7-24.8	12	32.0-52.0	18

Discussion

The water temperature in all locations was smaller than 33ºC and water pH 6.0-7.5 (Table 2) so that it still supports the life of the golden snail. Teo (2004) [14] stated that golden snails less resistent to high temperatures where at 33ºC, the mortality rate of golden snails is high. Phoong *et al*. (2018) [9] found that golden apple snails were abundant at locations with a pH of 6.5-7.0 and did not find snails at locations with a pH>8.

Based on the morphological characters (Fiure 2 and Figure 3), samples collected at all locations included the species *Pomacea canaliculata*. Strengthening characters were deep suture lines, forming an angle of <90º, spire is hihg, and rounded shoulders of the body whorl. Morphological characters that are considered strong and can be used to distinguish between species in the genus Pomacea are the depth of sutures, the height or the low of spire (Isnaningsih, 2013) [4]. *P. canaliculata* has a higher spire than other species (Rama Rao *et al*., 2018) [10], deep sutures (Arfan *et al*., 2014) [1] and rounded body whorl (Joshi *et al*., 2017; Arfan *et al*., 2014) [5, 1].

Golden apple snail shells found in all locations have a variety of colors. The color was yellowish to dark brown. Golden apple snails shell from all location also has a shell band. Arfan *et al*. (2014) [1] stated that *P. canaliculata* has a yellowish to dark brown shell color and has a shell band. The variety of shell color found in the golden apple snail is due to the influence of color pigments as mentioned in Yusa (2004), namely the shell color of the golden apple snail is determined by the dark pigment in the eyes, skin and shell.

Golden apple snail shell bands also vary in their appearance. The color of the shell band is more visible on the darker shell. Estebenet *et al*. (2006) [2] found golden apple snails with indistinct shell bands and only visible when the body part was removed and observed with adequate lighting. Yusa (2004) stated that golden apple snails with yellow shells lack dark pigment in the eyes, skin and shell so that they appear to have no banding pattern. Estebenet and Martin (2003), state that the variation of golden apple snail shell in one population (interpopulation) is higher than in different populations (intrapopulation).

Based on the results of the study, golden apple snails which were included in the juvenile group showed different shell bands from adults. The younger the snail is, the less obvious the shell bands are. The shell band is related to age. Estebenet *et al*. (2006) [2] stated that the intensity of the color of the golden apple snail shell band increased during its growth.

Grouping golden apple snails into age groups, there are differences in the number of juvenile and adult groups in each location (Table 3). Most of the golden apple snails found was groups of adult with the range of shell height were 25.0-61.5 mm. Environmental factors, processing of rice fields and the activity of collecting golden snails for animal feed also affect the presence of golden apple snails. Based on shell height as a reference for grouping into age groups, the range of shell heights found was 8.7-61.5 mm. The size of the shell height found by Suarmustika *et al*. (2018) [13] were 15.3-33.6 mm, there is a difference in the shell height of the juvenile and adult age groups found in this study. Widiastuti *et al.* (2015) [16] found that golden snails were 9.1 - 40.5 mm in size with the largest shell sizes of 29-33 mm which indicated that golden apple snails were considered mature and ready to produce.

Conclusion

Environmental factors in all locations are in the range where golden snails can live. Golden apple snail shells in all locations have a yellowish to darkner color and shell band is more visible on the darker shell. The smallest golden apple snails shell was found at the Bugbug location with shell height 8.7 mm and the largest shell size was found at the Duda location with shell height 61.5 mm. Most of the golden apple snails found was adult.

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