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Reproductive Biology of Moonlight Gourami (*Trichopodus microlepis*) in U Minh Thuong and U Minh Ha National Parks

Nguyen Thi Ngoc Tran^{1*}, Nguyen Thi Kim Ngan², and Duong Thuy Yen¹

¹College of Aquaculture and Fisheries, Can Tho University, Viet Nam

²Master Program in Aquaculture, Course 27, Can Tho University, Viet Nam

*Correspondence: Nguyen Thi Ngoc Tran (email: ntntran@ctu.edu.vn)

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ABSTRACT

This study aimed to provide information about the reproductive biology of moonlight gourami (*Trichopodus microlepis*) distributed in U Minh National Parks. Fish samples were collected monthly from January to October 2019 at two locations: U Minh Ha (UMH, Ca Mau, n=462) and U Minh Thuong (UMT, Kien Giang, n=635). Results showed that the maturation-stage ratios of females were similar between the two fish populations. Mature females (with stage IV-ovaries) and the increase in their gonadal somatic index (GSI) were found from June to October, indicating that this period is their spawning season. GSI reached the highest values in August with 8.04% in UMH and 6.44% in UMT. Female fish of the UMH population showed higher average fecundity (7,483±3,008 eggs/female) and larger egg diameter (762±55 μm) than those of UMT (6,898±2,952 eggs/female and 754±43 μm, respectively), but these differences were not significant ($p>0.05$).

1. INTRODUCTION

Moonlight gourami (*Trichopodus microlepis*), belonging to the genus *Trichopodus* of the family Osphronemidae, is native to Southeast Asia, widely distributed in both the Mekong and Chao Phraya basins (Kottelat, 2001). In the Mekong Delta of Viet Nam, there are three species of genus *Trichopodus* including *T. pectoralis* (snakeskin gourami), *T. trichopterus* (three-spot gourami), and *T. microlepis* (moonlight gourami) (Tran et al., 2013). While snakeskin gourami has become one of the most popular cultured species, the other two are mainly caught from the wild and are well-known as ornamental fish (Rainboth, 1996). However, moonlight gourami is also a significant food fish. This species can reach a maximum length of 16 cm, not very small compared to snakeskin gourami with

a maximum length of 25cm (Low & Lim, 2012). As such, moonlight gourami has large unexplored potential to become a cultured species.

Understanding the reproductive biology of a fish species is crucial to develop its farming. However, there is limited information on the biological characteristics of moonlight gourami. Previous studies have only emphasized key morphological parameters for species classification (Low & Lim, 2012), or provided only a general overview of its habitat and distribution (Kottelat, 2001; Rainboth, 1996; Tran et al., 2013). Low and Lim (2012) reported the spawning behavior of *Trichopodus* species in the wild. They observed that before spawning, males excrete mucous-coated bubbles into floating vegetation to form a nest at the water surface. Furthermore, after spawning, males protect

the nest until the fry hatch (Low & Lim, 2012). However, little is known about fecundity, egg size, and other reproductive characteristics are crucial for the captive production of moonlight gourami.

U Minh National Parks comprising U Minh Ha (UMH) and U Minh Thuong (UMT), which are located in the two provinces of Ca Mau and Kien Giang, are typical Melaleuca forest ecosystems (Vietnam Forestry, 2019). In these areas, moonlight gourami is an abundant wild fish (Duong et al., 2021). Although both National Parks have a similar ecosystem, the water exchange and canal systems around the two parks are different. As a result, water quality (including the bio-physico-chemical aspects) can vary, which may result in differences in the reproductive parameters of moonlight gourami, as well as other fish species. Therefore, this study investigated selected reproductive characteristics of

moonlight gourami in the U Minh National Parks in order to provide fundamental information for further research for the artificial breeding of the species.

2. MATERIALS AND METHOD

2.1. Fish sampling

Moonlight gourami specimens (Figure 1) were collected monthly from January to October 2019 using traditional fishing gear in the surrounding areas of two National Parks - U Minh Thuong (Kien Giang) and U Minh Ha (Ca Mau). A total of 1,097 individuals were collected during the sampling period (462 individuals from Ca Mau and 635 individuals from Kien Giang). Fish samples were preserved freshly in ice and transported to the laboratory of the College of Aquaculture and Fisheries, Can Tho University for further analyses.



Figure 1. Moonlight gourami collected in UMT National Park

2.2. Biological and reproductive analyses

Each gender of each individual was first identified based on body color and shape of the dorsal fin. Moonlight gourami males are more colorful with an orange colored abdomen compared to females with the normal silvery body. Moreover males typically have dorsal fin lengths longer than females.

Thereafter, the fish samples were measured for body weight with (or total weight, W) and without internal organs (W_o), and the weight of reproductive organ (W_g). The maturity stages of fish were classified based on the method of Nikolsky (1963). Ovaries that reached maturity stage IV were preserved in 2.5% formalin for fecundity estimation and egg diameter measurement.

Reproductive parameters which were evaluated included:

(i) Gonadal somatic index (GSI), an index to predict the reproductive cycle of fish, was analyzed by the equation $GSI=(GW/W)*100$, in which: GW is

gonadal weight, and W is the total weight of fish (Biswas, 1993).

(ii) Absolute fecundity of each female was identified by the counting method (Biswas, 1993). Three representative samples were randomly taken from each ovary solution (containing eggs and formalin). The weight and egg number of each sample were recorded. The average number of eggs per gram from three counting samples (n) was then calculated and multiplied by the total weight of sample solution (A) to estimate the absolute fecundity (F), as $F=n*A$.

(iii) Relative fecundity was computed by the equation: $RF=F/W$, in which: F is the absolute fecundity (eggs), and W is the total weight of female fish (g).

(iv) Egg diameter was measured using a stereo microscope Nikon SMZ745T under 1X magnification by NIS-Elements Basic Research software. A group of 15-20 eggs from each stage IV-

ovary were randomly chosen for diameter measurements.

2.3. Data analysis

Reproductive parameters including GSI, fecundity, and egg diameter were compared between the two populations by using t-tests. The relationships between reproductive parameters (absolute fecundity and egg diameter) and female weight were investigated by linear regression analysis. These data were analyzed using SPSS 22 and Excel 2013.

3. RESULTS

3.1. Maturity stages of moonlight gourami in U Minh National Parks

Maturity stages of moonlight gourami were identified only in females. The gonad of male fish is very small and tends to decomposed during the preservation process, leading to the inability to observe their maturity stages. Ovary maturation before spawning was classified into four stages based on Nikolsky’s scales (1963). Stage I-II is recognized by a small and transparent ovary of pale

yellow color. From stage III onwards, the ovary becomes more yellowish with the appearance of blood vessels and uneven oocytes. Ovaries at stage IV turn to yellow with prominent blood vessels and a uniform size of oocytes (Figure 2). When a female releases eggs, its ovary is at stage V, which cannot be observed. After spawning, the ovary is at stage VI, characterized by a very soft ovary with some oocytes and fluid.

The percentage of the sampled four maturity stages of moonlight gourami females in both two National Parks during 10 months (January to October) is presented in Figure 3. Similar trends occur in both areas with the appearance of stages I-II and III from January to May. Stage IV is seen from June onwards, reaching the highest percentage in August (54.55% from UMH and 31.25% from UMT) and decreases from September. Ovaries at stage V-VI are found in September and October. These results indicates that the spawning period of moonlight gourami can be assumed to occur from June to October with a peak in August.



Figure 2. Ovaries at different stages of moonlight gourami

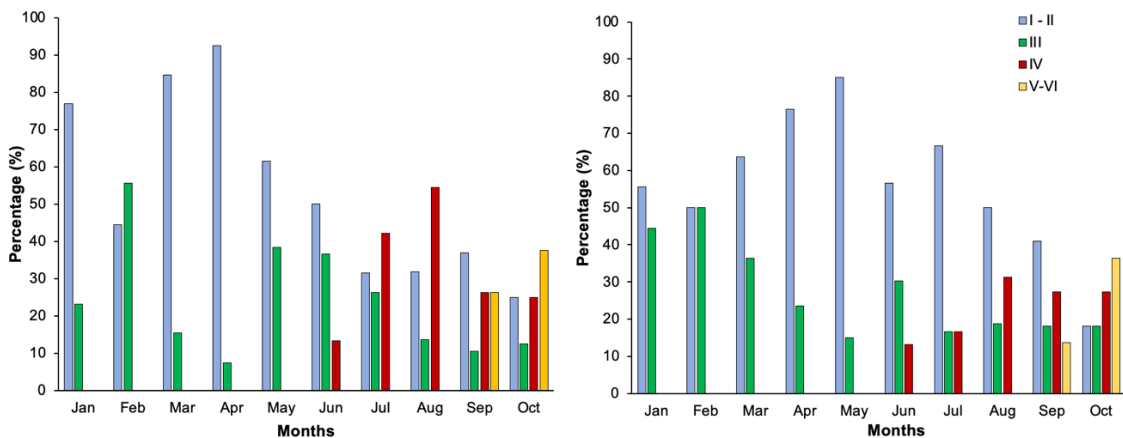


Figure 3. Percentage of maturity stages of female moonlight gourami by months in (the U Minh Ha (left) and U Minh Thuong (right) National Parks

3.2. Gonadal somatic index (GSI) (%) of moonlight gourami females in U Minh National Parks

The lowest values of GSI were in stage II (0.71% of UMH and 0.46% of UMT) and the highest in stage IV (7.53% in UMH and 6.73% in UMT). GSI at almost all stages of the UMH population was higher than that of the UMT population (Table 1).

Both the UMH and UMT moonlight gourami populations showed a similar trend of monthly variations in GSI (Figure 4). However, the monthly GSI of the UMH population was lower than that of UMT. GSI values were lower for the period from January to May, reaching the peaks in August with 8.04% in UMH and 6.44% in UMT. GSI values decreased in September, but were seen to increase slightly again in October.

Table 1. Gonadal somatic index (%) by maturity stages of moonlight gourami females

Population	Parameters	I - II	III	IV	V-VI
U Minh Ha	Range	0.06 – 1.70	1.71 – 4.25	4.56 – 11.46	0.94 – 3.49
	Mean ± SD.	0.71±0.47	2.58±0.72	7.53±1.81	1.81±1.03
U Minh Thuong	Range	0.03 – 1.91	1.95 – 6.14	4.12 – 9.52	1.63 – 3.49
	Mean ± SD	0.46±0.45	3.42±1.19	6.73±1.48	2.40±0.82

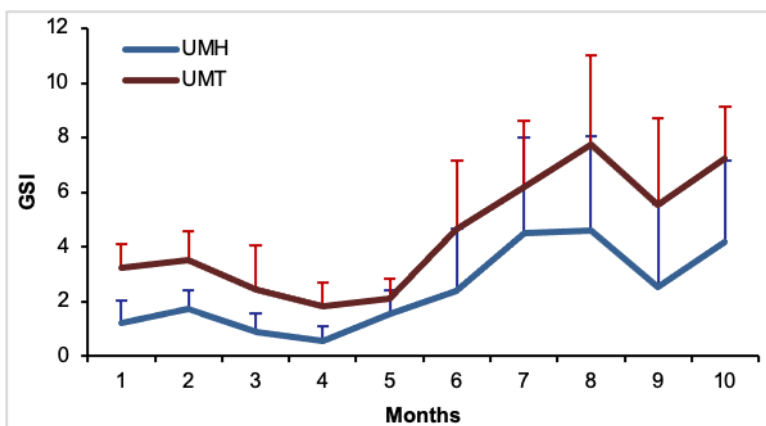


Figure 4. Variation in GSI in moonlight gourami females in U Minh National by month

3.3. Fecundity and egg diameter of moonlight gourami in U Minh

Absolute fecundity and relative fecundity of moonlight gourami in two populations were compared (Table 2). Fecundity parameters of fish were insignificantly different between the two populations ($p>0.05$). Absolute fecundity averaged $7,483\pm3,008$ eggs/female in UMH and $6,898\pm2,952$

eggs/female in UMT. The average relative fecundity of the two populations was similar to each other with $186,610\pm59,110$ eggs/kg of female fish in UMH and $189,430\pm58,718$ /kg female in UMT. Moreover, the egg diameter of moonlight gourami ranged from 639 to 931 μm in UMH, similar to that of the fish from UMT, ranging from 658 to 863 μm ($p>0.05$).

Table 2. Fecunity of moonlight gourami in UMH and UMT National Parks

Parameters		UMH (n=29)	UMT (n=31)
Weight (g)	Range	6.54 – 61.3	8.87 – 59.0
	Mean ± SD	35.5±11.5 ^b	27.8±10.7 ^a
Absolute fecundity (eggs/ female)	Range	2,768 – 13,994	1,819 – 12,572
	Mean ± SD	7,483±3,008 ^a	6,898±2,952 ^a
Relative fecundity (eggs/kg female fish)	Range	86,529 – 362,000	65,604 – 301,632
	Mean ± SD	186,610±59,110 ^a	189,430±58,718 ^a
Egg diameter (μm)	Range	639 – 931	658 – 863
	Mean ± SD	762±55 ^a	754±43 ^a

(*) Value in the same row with the different characters shows significant differences with $p<0.01$

The linear relationship between absolute fecundity and weight of female fish in the National Parks is illustrated in Figure 5. The regression equation of UMH population is $F_{UMH} = 190.12W_{UMH} - 116.58$, while UMT is $F_{UMT} = 180.54W_{UMT} + 496.37$. In both populations, a increased female weight was positively correlated with more eggs ($p > 0.05$).

However, there was also a negative linear relationship between female weight and egg

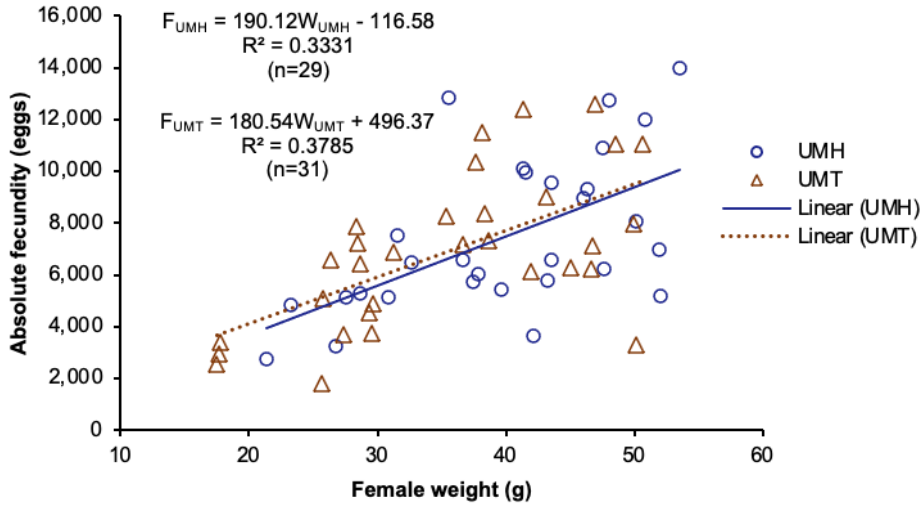


Figure 5. Relationship between absolute fecundity and female weight of moonlight gourami in UMH and UMT National Parks

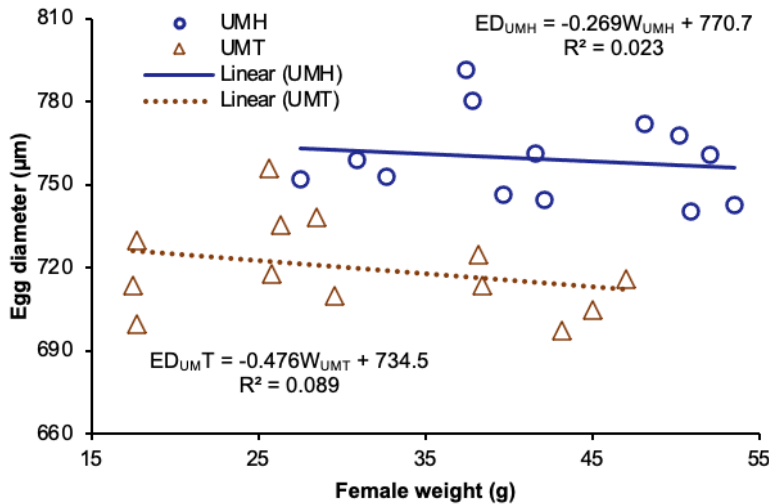


Figure 6. Relationship between egg diameter and female weight of moonlight gourami in UMH and UMT National Parks

4. DISCUSSION

Results of this study found that moonlight gourami in both UMH and UMT National Parks have similar spawning seasons from June to October. A previous

diameter (Figure 6). Regression equation of UMH population is $ED_{UMH} = -0.269W_{UMH} + 770.7$, while UMT is $ED_{UMT} = -0.476W_{UMT} + 734.5$. Bigger moonlight gourami females produced smaller eggs. However, the slopes (-0.0269 and -0.476) and determination coefficient ($R^2_{UMH}=0.023$ and $R^2_{UMT}=0.089$) of the linear regressions were small, indicating that the relationship is weak.

study on the reproductive biology of three spot gourami (*Trichopodus trichopterus*) in Ca Mau province found a similar spawning season (Nguyen & Chung, 2017). *Trichogaster fasciata*, one species of the Osphronemidae family, is not distributed in

Viet Nam but mainly lives in South Asia (India, Bangladesh, Nepal, Pakistan) also has a spawning season in the rainy season from March to October (Gupta, 2015). Some ornamental gouramies in India such as pearl gourami (*T. leerii*), and chocolate gourami (*Sphaerichthys osphromenoides*) have a peak spawning season during the monsoon season from April to August/September on the Indian sub-continent (Jena et al., 2019). Therefore, species from the Osphronemidae family spawn in the rainy season.

The two populations have the same trend of maturation stage ratios and GSI variation by month, but the monthly values in UMT were seen to be higher than those in UMH. The variations in GSI between the two populations can be explained by differences in water conditions between the two areas, with pH playing a dominant role. UMH has acidic water with a pH <4, which results in low natural food availability and hinders the growth and reproduction of fish. On the other hand, the pH in UMT fluctuates between 6 to 7, which provides better conditions for fish to reach full maturity (Boyd & Pillai, 1985).

When compared to other Osphronemidae species, moonlight gourami has medium values of GSI. GSI of this species is higher than that of three spot gourami (*T. trichopterus*) with the highest GSI of 3.93% in July (Nguyen & Chung, 2017), but lower than 10% of banded gourami *Trichogaster fasciata* in the peak of spawning season (Mitra et al., 2007).

The fecundity and egg size of moonlight gourami were similar between the two populations. Yet, absolute fecundity varied within each population (a large range). However, both populations showed the

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- same increase in fecundity with female size (indicated by similar slopes of the linear regression between fecundity and female size). This positive relationship is also reported for different fish species such as white-spotted charr *Salvelinus leucomaenis* (Morita & Takashima, 1998), *Tilapia zilli* (Dadzie & Wangila, 1980), common carp *Cyprinus carpio* (Weber & Brown, 2012).
- In two moonlight gourami populations, egg sizes are also similar and slightly decrease with female size. Some tilapia species such as black chin tilapia (*Sarotherodon melanotheron*) and *Tilapia zilli* were also reported no correlation between female sizes and egg sizes (Peters, 1983). However, the opposite trend has been found in some other species. For example, in the common carp (Weber & Brown, 2012) and some species of fork-tailed catfishes (Pisces: Ariidae), larger females produce bigger eggs. Compared to other gourami species, the egg size of moonlight gourami (639.1 - 931.5 μm) is comparable to three spot gourami with 545–705 μm (Atomar & Nazma, 2017).

5. CONCLUSION

Moonlight gourami in U Minh Ha and U Minh Thuong National Parks have similar fecundity, egg size, and similar relationships between female size and these reproductive parameters. Both populations showed the same pattern of spawning season which is from June to October with a peak in August.

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