Ovarian development, spawning season, size at maturity and fecundity of *Acentrogobius viridipunctatus* (Valenciennes, 1837) in the Vietnamese Mekong Delta

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ABSTRACT

This present study provides an overview of the reproductive traits, including ovarian development, spawning season, size at maturity (L_m), and fecundity of *Acentrogobius viridipunctatus* – a high economic value fish in the Vietnamese Mekong Delta (VMD). A total of 432 females were collected using trawl nets from January to December 2020 at four sites, including Long Huu-Tra Vinh (LHTV), Trung Binh-Soc Trang (TBST), Dien Hai-Bac Lieu (DHBL), and Tan Thuan-Ca Mau (TTCM). The ovarian diameter was seen to increase from 2.09 (stage I) to 6.57 mm (stage V). Histological analysis showed that the goby was a multi-spawner due to the different oocyte stages found in stages IV and V of ovaries. It can release eggs all year-round due to the monthly appearance of ovarian stages IV and V during the study period, with the main peak in the 2–4 months of the wet season noted with the highest value of gonadosomatic index at that time. The L_m increased from 6.6 cm at TBST to 9.4 cm at TTCM. This species displayed relatively high fecundity, ranging from 5,481 to 130,683 eggs/female. The average fecundity of this fish was 27,698 ± 7,983 eggs/female at LHTV, 46,592 ± 7,264 eggs/female at TBST, 23,271 ± 4,985 eggs/female at DHBL, and 31,408 ± 2,515 eggs/female at TTCM. Egg diameter ranged from 0.45 ± 0.01 at DHBL to 0.50 ± 0.01 at TBST. For sustainable exploitation, local governments should ask the fishers to avoid catching fish during the main spawning period, and the fish length at first capture should be >L_m.

INTRODUCTION

The Vietnamese Mekong Delta (VMD) is the third-largest delta globally (*Coleman & Huh, 2012*). The VMD is the downstream area of the Mekong River system and is adjacent to the sea. With more than 700 km of coastline and an interlaced river system, the VMD is the

preferable place for exploiting and cultivating fish ranging from freshwater to saltwater (*Le et al., 2006*). In addition, the fish communities that live here are diverse, with 77 families and 322 recorded fish species (*Tran et al., 2013*). Of these, there are ~80 fish species with high economic value (*Thai, 2015*). However, the overexploitation of economically valuable fish species has led to a rapid decrease in their population. There has also been the failure of appropriate measures to conserve and protect endangered fish species (*Thai et al., 2012*). As such, it is necessary to study their biological characteristics in able to develop a suitable strategy for improved fish resources.

Reproduction is necessary for a species to survive and develop (*Mai et al., 1979*). This is a complex process that involves gonadal development and fish behavior. Changes in the histological characteristics of gonads through different stages aid a better understanding of fish's reproductive cycle and patterns (*Pham & Tran, 2004*). In most fish, the gonads are composed of two tubular chambers (*Mai et al., 1979*). In addition, reproductive biology also plays a vital role in conserving fish species. The length at first maturity (L_m) helps recommend appropriate fishing length and is an essential indicator in managing fish stocks (*Fontoura, Braun & Milani, 2009*; *Teichert et al., 2014*). Furthermore, the gonadosomatic index (GSI) is helpful in determining the fish spawning season (*Plaza et al., 2007*; *Dinh & Le, 2017*). These reproductive biology features are closely related to the production and capture of aquatic species (*Miller, 1984*; *Komolafe & Arawomo, 2007*).

Goby is one of the fish groups with high economic and nutritional value in the VMD (*Nguyen, 2000*), living in both freshwater to saltwater environments (*Tran et al., 2013*). *Acentrogobius viridipunctatus* is a typical fish of this fish group and has prominent features such as a dark curved line under its eyes and many bright blue spots on its head and body (*Tran et al., 2013*). This fish is relatively small, with a maximum length recorded in estuarine Israel of 16.5 cm (*Bauchot, Ridet & Diagne, 1989*). With their modest size, their food source is small animals such as other fish, shrimp, and organic humus (*Le & Tong, 2011*). However, there is scarce data on this species in the VMD, particularly the reproductive biology of females. Also, along with several other fish species, this species is at risk of habitat loss and overfishing (*Thai et al., 2012*). Research on the reproductive characteristics of the fish, *e.g.*, ovarian development, spawning season and pattern, and length at first maturity, plays an essential role in the raising and artificial reproduction of this fish and may provide insights for a sustainable exploitation strategy.

MATERIALS AND METHODS

As *A. viridipunctatus* is distributed mainly in brackish water, the sampling was carried out in estuarine areas where they occur dominantly, such as Long Huu-Tra Vinh (LHTV), Trung Binh-Tran De (TBST), Dien Hai-Bac Lieu (DHBL) and Tan Thuan-Ca Mau (TTCM) (Fig. 1). Here the dry season was from January to May (with no rain), and the wet season was from June to December (with heavy rain) (*Le et al., 2006*). Fish were collected as previously described in *Dinh et al. (2022a)*. Specifically, fish were collected once a month using trawl nets and were anesthetized with MS222 (25 mg MS22 was diluted with 5 liters of water taken from the sampling site) for ~5 min before fixing in 5% formalin and shifting to the laboratory for analysis. The use of fish was approved by the Scientific Committee of
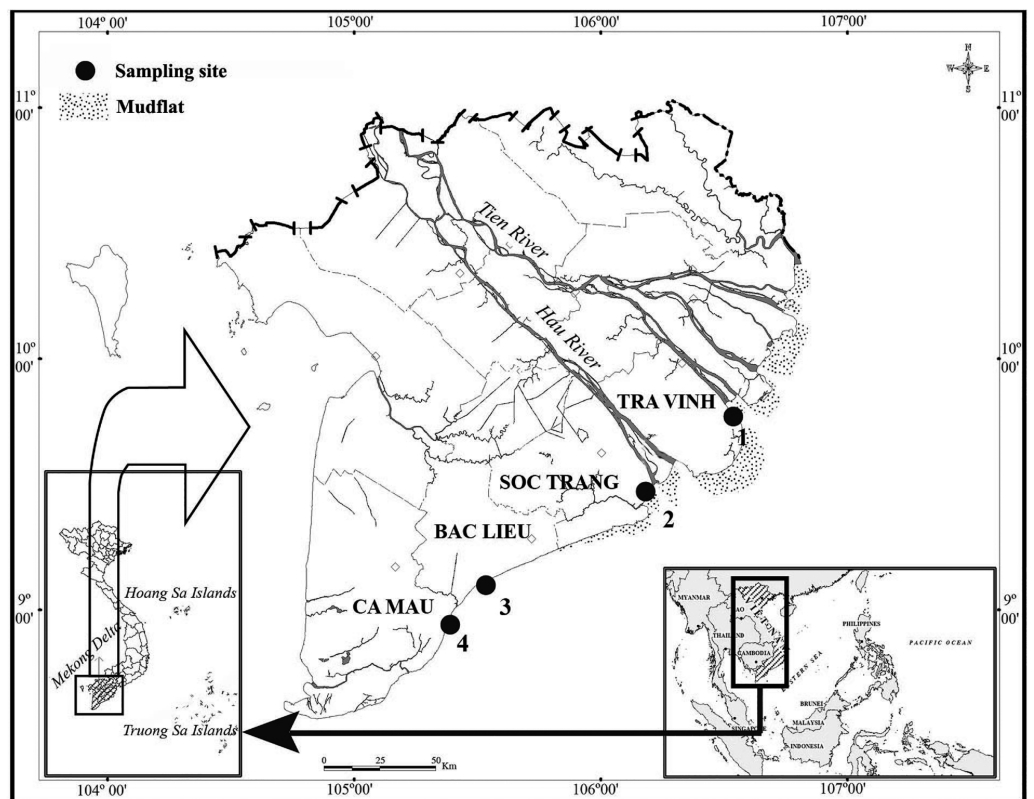
Figure 1 Map of sampling sites. 1: Long Huu-Tra Vinh; 2: Trung Binh-Soc Trang; 3: Dien Hai-Bac Lieu; 4: Tan Thuan-Ca Mau; *Dinh (2018)*.

the School of Education, Can Tho University, under the Animal Welfare Assessment number Q2020-01/KSP. According to *Dinh et al. (2016)*, after sampling, the fish samples were sexed using the genital spines between males (pointed triangles) and females (oval shapes). The temperature and salinity of these sites were also recorded using a temperature meter (HI98128) and a Refractometer (SLI-10), respectively.

After measuring total length (TL, in cm) and weight (W, in g), the fish was dissected to remove the ovary for morphological and histological analysis in the laboratory (Animal Laboratory, Department of Biology, School of Education, Can Tho University). The ovary was visually classified into five stages (immature in stage I to maturing in stage II and III, mature in stage IV, ripe in stage V) of development using the criteria description for *Parapocryptes serperaster*, documented by *Dinh et al. (2016)*. The ovary was then weighed to the nearest 0.1 mg using a precision balance and measured in diameter to the nearest 0.01 mm using a Motic Image-Pro Plus v.2.0 integrated with the stereomicroscope. Thereafter, 25 ovaries (five samples for each stage) were selected to examine histologically using the staining process of *Ho, Nguyen & Dinh (2021)*, including fixing, dehydrating, paraffin wax embedding impregnating, 6-μm thick cutting and staining with Hematoxylin and Eosin-Y, for gamete developmental determination.

The length at first maturity (L_m) was the length in which 50% of fish reached sexual maturity and calculated using the formula P = $1/(1 + e^{-r(TL - Lm)})$ (*King, 2013*), where P is the percentage of adult fish (%); and TL is the total length of the fish (cm).

The spawning season was determined based on ovarian frequency composition and the gonadosomatic index (GSI) (*Plaza et al., 2007*; *Dinh & Le, 2017*). Whereas, the GSI was calculated as GW * 100/W (GW, 0.1 mg) (*Lloret & Rätz, 2000*).

A total of 60 ovarian stages IV onwards (15 samples per site) were used to assess batch fecundity. The ovaries were soaked in water to prevent oocyte rupture, and a pen was used to remove the membranes and separate oocytes. The oocytes were observed and counted under a Motic stereomicroscope to determine the exact number and diameter of eggs (*Nguyen et al., 2021*). According to *Bagenal (1967)*, batch fecundity was from the formula: F = $(n \times G)/g$ (F: batch fecundity; n: the number of oocytes in sub-sample; g: the weight of sub-sample; and G: the ovarian weight). The oocyte diameter per ovarian stage, according to *Dinh et al. (2022c)*, was determined by randomly measuring 30 oocytes in each ovary using Motic Image-Pro Plus v.2.0.

One-way ANOVA with Tukey *Post-hoc* test determined the monthly variation of the GSI and spatial change of salinity and temperature at a 5% significance level. According to the method of *Morey et al. (2003)*, the t-test was used to verify if the L_m varied significantly between sites at a *p*-value < 0.05. SPSS v.21 software was used for statistical processing. The total length, weight, and batch fecundity values were transformed into the log10 before being used to qualify the relationship between fecundity with length and weight *via* linear regression.

RESULT

Environmental characteristics at the study sites

According to the study site, measurement results over 12 months showed a significant difference in salinity, with salinity reaching the highest value at DHBL (32.3 ± 1.1‰) followed by TTCM (28.7 ± 1.1‰) and lowest at LHTV (17.8 ± 2.3‰) and at TBST (19.8 ± 2.4‰) (One-way ANOVA, F = 15.73, df = 3, *p* < 0.001) (Table 1). However, the temperature did not differ over the study sites and ranged from 30.1 ± 0.5 °C in TTCM to 31.0 ± 0.4 °C in LHTV (F = 0.99, df = 3, *p* = 0.40) (Table 1).

Ovary development and spawning pattern

A total of 960 individuals (528 males and 432 females) were sampled at these sites from January to December 2020. Of them, 432 females were used in this study. Hereafter the number of males was used in another study (Table 2). Analytical results showed that the ovary of *A. viridipunctatus* was long, tubular, and composed of two chambers located close to the spine in the abdominal cavity. Connective membranes fixed the outside of the fish ovary. However, the ovary showed different morphological and histological features through each stage. Specifically,

Stage I: At this stage, the ovary was observed to be milky white, thin in size, and had an average diameter in 21 ovaries of 2.09 mm (Fig. 2A). The ovarian histological structure consisted of germ cells (GC) and oogonia (O). In addition to the GC and O, several

Table 1 Measured temperature and salinity at the four sampling sites.

Month	Long Huu-Tra Vinh		Trung Binh-Soc Trang		Dien Hai-Bac Lieu		Tan Thuan-Ca Mau	
	Temperature (°C)	Salinity (‰)	Temperature (°C)	Salinity (‰)	Temperature (°C)	Salinity (‰)	Temperature (°C)	Salinity (‰)
01/2020	28.3	23.0	29.2	20.0	28.0	25.0	26.5	24.0
02/2020	29.3	20.0	27.9	25.0	27.5	32.0	28.0	30.0
03/2020	28.8	27.0	26.3	29.0	27.1	28.0	28.3	31.0
04/2020	31.0	18.0	30.8	19.0	30.1	32.0	30.1	28.7
05/2020	31.0	28.0	30.1	31.0	31.7	34.0	32.0	32.0
06/2020	31.8	14.0	32.3	19.0	30.3	37.0	30.6	33.0
07/2020	32.0	10.0	33.3	13.0	31.2	33.0	30.7	27.0
08/2020	32.1	12.0	31.9	10.0	31.0	32.0	30.9	25.0
09/2020	32.2	9.0	33.1	10.0	30.9	31.0	30.7	24.0
10/2020	32.5	29.0	30.7	32.0	32.3	35.0	32.6	33.0
11/2020	31.9	9.0	32.8	10.0	30.6	31.0	30.4	24.0
12/2020	31.5	14.0	32.0	19.0	30.0	37.0	30.3	33.0
Mean	31.0	17.8	30.9	19.8	30.1	32.3	30.1	28.7
SE	0.4	2.3	0.7	2.4	0.5	1.0	0.5	1.1

Table 2 Total length (TL) and weight (W) of fish at sampling sites for 12 months.

Month	Long Huu – Tra Vinh			Trung Binh – Soc Trang			Dien Hai – Bac Lieu			TTCM		
	Total	W_{min}–W_{max}	TL_{min}–TL_{max}	Total	W_{min}–W_{max}	TL_{min}–TL_{max}	Total	W_{min}–W_{max}	TL_{min}–TL_{max}	Total	W_{min}–W_{max}	TL_{min}–TL_{max}
01/20	5	4.59–13.63	7.3–10.1	6	5.24–14.48	6.6–10.7	3	4.36–19.45	6.5–10.5	14	6.19–13.68	7.2–10.7
02/20	14	4.28–7.00	6.8–8.2	5	5.56–13.63	7.1–10.1	9	6.00–23.91	7.6–11.5	11	9.49–23.41	9.6–12.6
03/20	7	4.26–13.70	7.0–9.5	4	5.56–13.09	7.1–9.9	16	5.38–19.80	6.8–11.5	3	5.13–22.88	7.6–12.4
04/20	18	6.53–23.13	8.2–12.2	8	9.23–15.99	8.6–11.6	11	4.24–10.04	6.3–8.9	12	6.33–28.13	8.0–13.5
05/20	4	11.92–17.17	9.6–10.8	13	2.47–34.25	5.9–13.1	14	5.02–13.29	6.8–9.9	10	8.36–23.29	8.5–12.5
06/20	12	7.28–16.09	8.5–11.2	2	10.12–25.36	9.7–13.6	7	10.86–18.7	10.1–11.4	10	6.79–20.85	7.8–11.9
07/20	11	9.40–17.62	8.5–11.8	6	9.65–14.82	9.7–10.6	5	5.09–18.81	7.2–11.2	13	5.88–16.16	7.2–11.0
08/20	13	4.30–19.23	7.0–11.5	5	6.33–24.88	7.6–13.6	3	6.17–16.63	7.4–12.6	12	6.88–12.02	7.5–10.5
09/20	6	4.75–11.70	6.5–9.0	16	4.50–21.75	6.5–11.4	3	6.93–16.03	8.6–11.9	12	5.72–25.20	7.2–11.6
10/20	7	8.56–13.46	8.2–10.0	8	5.40–8.29	8.6–10.4	15	7.20–22.10	8.7–12.6	10	4.99–11.81	7.4–10.1
11/20	7	7.92–14.83	8.4–10.5	11	5.01–8.24	7.4–9.6	3	6.54–15.40	8.3–11.6	13	6.73–15.84	8.2–12.0
12/20	5	4.19–7.18	7.0–8.8	9	4.9–16.12	7.2–11.2	3	6.54–15.12	8.3–11.2	18	5.90–25.78	7.6–12.8
Total	109	4.19–23.13	6.5–12.2	93	2.47–34.25	5.9–13.6	92	4.24–23.91	6.3–12.6	138	4.99–28.13	7.2–13.5

primary oocytes (PO) were scattered throughout the ovary. The GC was small, had large nuclei, and was located in clusters, whereas O was more significant in size, acquired a dark purple colour with Hematoxylin, and divided. The PO grew from O and had a central nucleus. At this stage, yolk sacs did not appear (Fig. 2F).
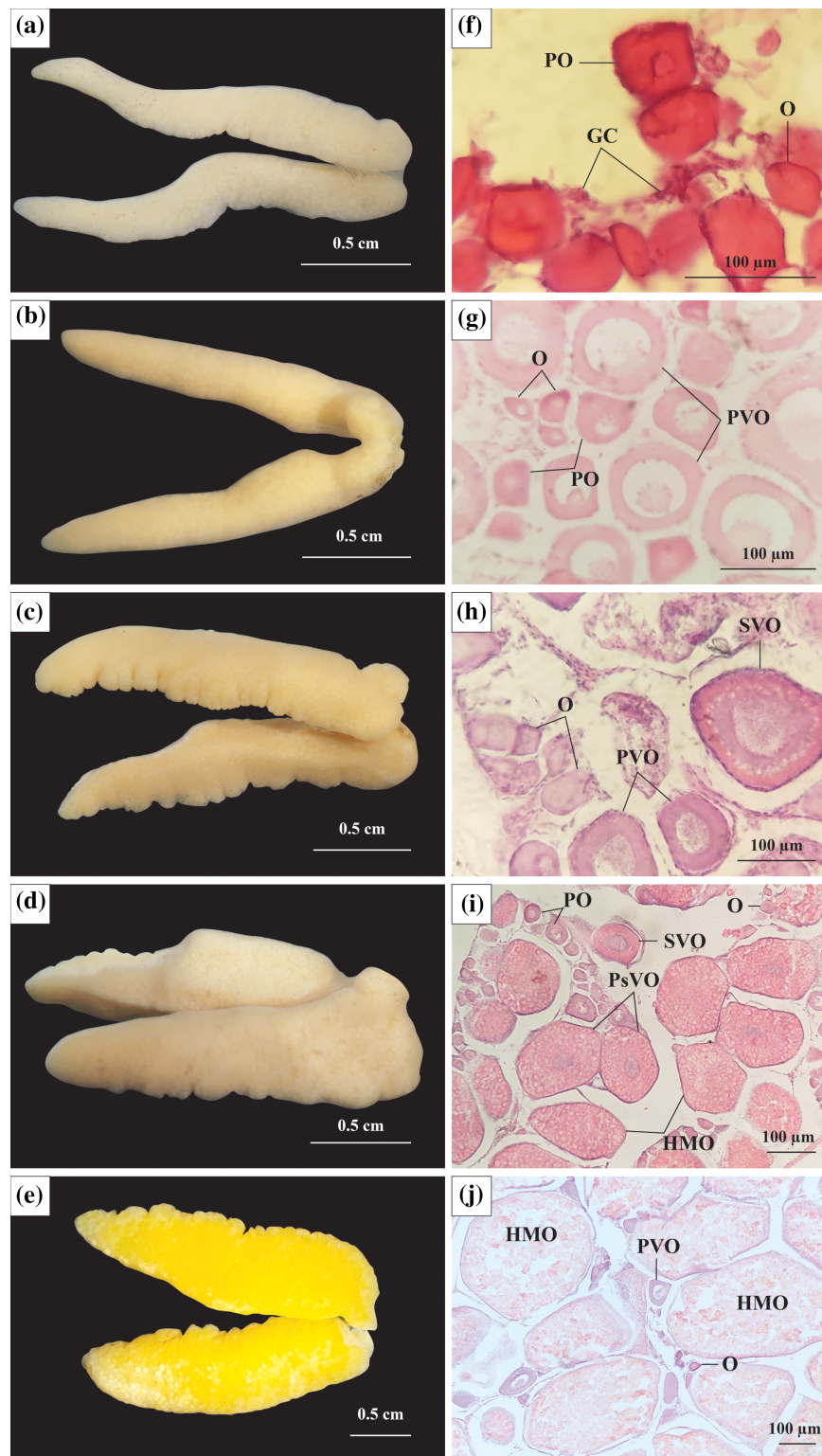
Figure 2 The morphology and histological in the ovary of *Acentrogobius viridipunctatus*. (A–E) Stage I–V of the ovary; (F–J): histology of the ovary in stages I–V; GC, germ cells; O, oogonia; PO, primary oocyte; PVO, primary vitellogenic oocytes; SVO, secondary vitellogenic oocytes; PsVO, post vitellogenic oocytes; HMO, hydrated oocytes; sampled from Tan Thuan-Ca Mau.

Stage II: At this stage, the ovary increased to 2.47 mm (45 ovaries) in diameter and was light yellow in colour (Fig. 2B). The PO appeared predominantly, and the number of O gradually decreased, leaving only a few clusters distributed interspersed with GC. Some PO was generated before dividing into primary vitellogenic oocytes (PVO). The nuclear rings were found in PO and PVO (pale with Hemtaoxyline cells) (Fig. 2G).

Stage III: The ovary diameter at this stage was 3.19 mm (83 ovaries), and the surface was rough and wavy (Fig. 2C). In the ovary, most of the PO had developed to PVO. The PVO further developed into secondary vitellogenic oocytes (SVO) with numerous yolk sacs containing yolk granules. At this stage, the oocyte developed strongly in cytoplasmic size. The ratio between nuclear volume and cell volume during this period was seen to be significantly reduced, yet the O still appeared during this period in small numbers. At the end of this phase, half of the cytoplasm located near the periphery was filled with yolk sacs (Fig. 2H).

Stage IV: Ovary occupied 1/2 volume of the abdominal cavity and reached 4.05 mm (250 ovaries) in diameter (Fig. 2D). The ovary had the addition of post vitellogenic oocytes (PsVO) and hydrated oocytes (HMO). The size of the oocyte was almost at its maximum, containing high nutrients. During this stage, the PsVO and HMO occupied most of the area in the ovary. The HMO was spherical pale pink, occupying most of the volume. The oocyte's nucleus began to contract, the nuclear membrane gradually disappeared, and most nuclei moved to the nucleus' centre (Fig. 2I).

Stage V: The size of the ovary reached its largest size with a diameter of 6.57 mm (33 ovaries) and bright yellow in colour (Fig. 2E). Each egg can be observed from the outside when pressed firmly against the fish belly. The histological structure at this stage was mainly HMO. In addition, in the cross-sectional smear of the ovary, GC regions were also observed. This was the basis for the continued development of the ovary (Fig. 2J).

Spawning season

A graph of the maturation stages of *A. viridipunctatus* showed the number of individuals obtained per month and the stage of ovary development in this species (Fig. 3). Each color in each column represents the ratio of a different stage, and the number in each box represented the number of individuals at that stage. Analysis of the frequency of occurrence of the ovary at the four study sites for the 12 months revealed that this fish belongs to the group of fish that reproduce many times during the spawning season. Ovarian stages IV to V were found at all months (Fig. 3). To know the exact breeding season of fish, it was necessary to rely on the change in the value of GSI between months. At LHTV, the GSI of this fish had reached its highest value from August to September (One-way ANOVA, df = 11, F = 2.22, p = 0.02, Fig. 4A). Meanwhile, in TBST, the highest value of GSI only appeared in the months of June and July (F = 3.28, df = 11, p = 0.001, Fig. 4B). Similarly, at DHBL, GSI displayed high values in August and September (F = 1.98, df = 11, p = 0.04, Fig. 4C). Finally, at TTCM, the period from May to July was the time with the highest GSI value (F = 2.99, df = 11, p = 0.001, Fig. 4D). Although the change in GSI was seen across study sites, this species generally has a highly concentrated spawning season in the wet season.
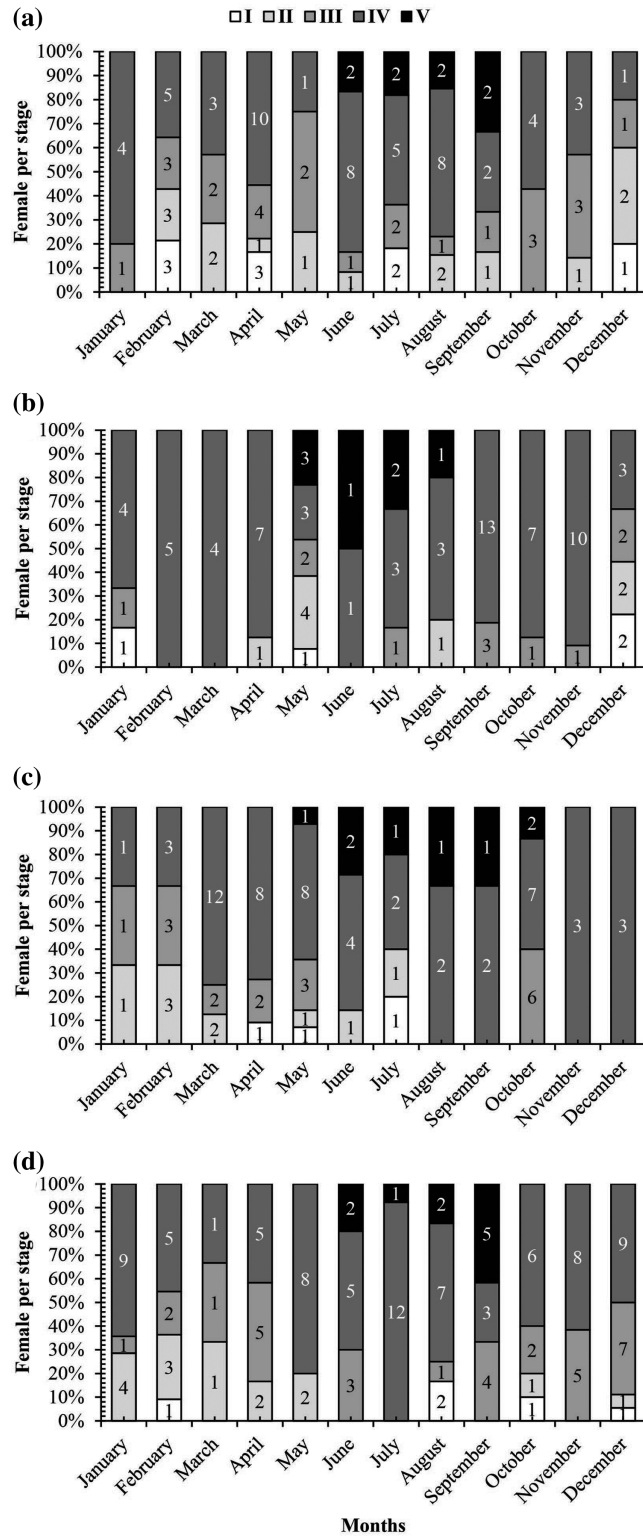
Figure 3 The ovarian frequency composition of *Acentrogobius viridipunctatus*. (A) Long Huu-Tra Vinh, (B) Trung Binh-Soc Trang, (C) Dien Hai-Bac Lieu, (D) Tan Thuan-Ca Mau; number in each column: number of individuals.
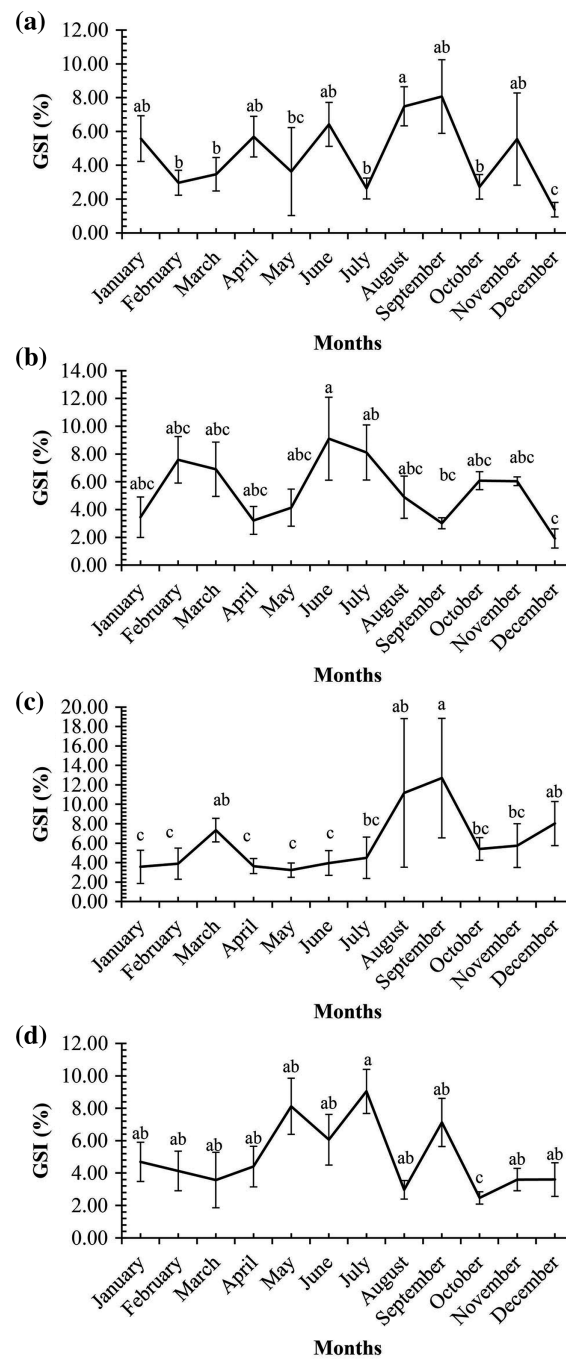
Figure 4 Gonadosomatic index of *Acentrogobius viridipunctatus*. (A) Long Huu-Tra Vinh, (B) Trung Binh-Soc Trang, (C) Dien Hai-Bac Lieu, (D) Tan Thuan-Ca Mau; different letters denote the variation in the monthly GSI values at each site.

Length at first maturity and fecundity

The length at first maturity of *A. viridipunctatus* varied between the four study sites, reaching the highest value at TTCM (9.4 ± 0.4 SE), followed by DHBL (8.6 ± 0.2 SE) and LHTV (7.5 ± 0.3 SE), and the lowest value at TBST (6.6 ± 0.2 SE) (Fig. 5). Specifically, L_m at
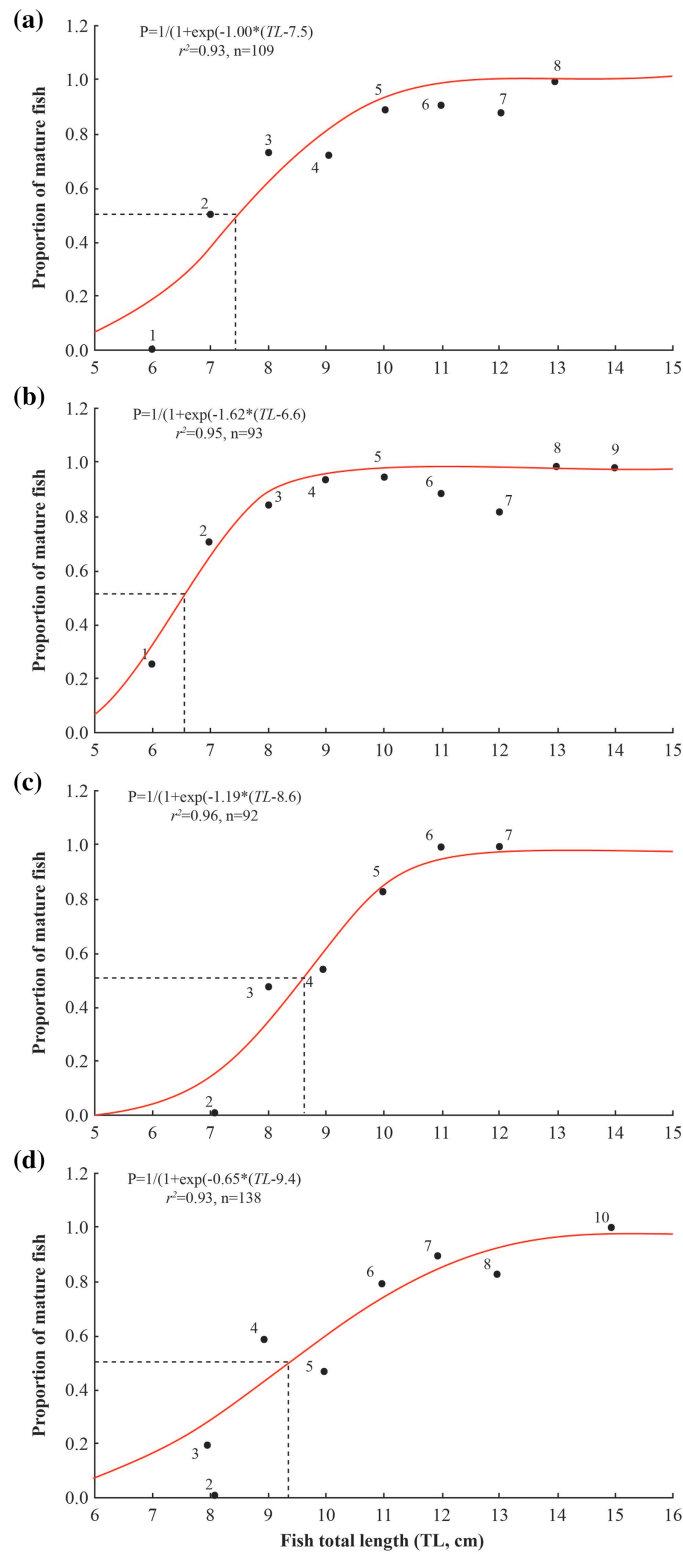
Figure 5 Size at first maturity of *Acentrogobius viridipunctatus*. (A) Long Huu-Tra Vinh, (B) Trung Binh-Soc Trang, (C) Dien Hai-Bac Lieu, (D) Tan Thuan-Ca Mau.
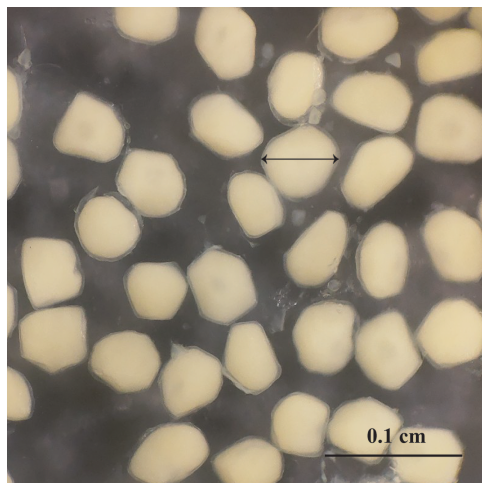
Figure 6 The outer appearance of the egg in *Acentrogobius viridipunctatus*. Sampled from Tan Thuan-Ca Mau.

TBST was significantly lower than that at LVTH (t = 4.50, df = 16, $p < 0.001$), DHBL (t = 10, df = 2, $p = 0.01$), and TTCM (t = 14, df = 45, $p < 0.001$). The L_m at LHTV was significantly lower than that at DHBL (t = 3.67, $p = 0.002$) and TTCM (t = 6.33, df = 29, $p < 0.001$). Similarly, the L_m of DHBL was significantly lower than that of TTCM (t = 4.00, df = 46, $p < 0.001$).

Observation results under the stereomicroscope showed that the HMO (*e.g.*, egg) of *A. viridipunctatus* was spherical (Fig. 6). The mean egg diameter varied between the four study sites. The fecundity *of A. viridipunctatus* was quite high (5,481–130,683 eggs/female) and varied from site to site. Similarly, this fish also exhibited the highest value of *F* at TBST (46,592 ± 7,264 eggs/female, *n* = 15) and the lowest at DHBL (23,271 ± 4,985 eggs/female, *n* = 15). The F value of the two remaining sites was 27,698 ± 7,983 eggs/female (*n* = 15) at LHTV and 31,408 ± 2,515 eggs/female (*n* = 15) at TTCM. In this fish, fecundity was seen to be closely related to total fish length ($r^2_{LHTV} = 0.68$; $r^2_{TBST} = 0.67$; $r^2_{DHBL} = 0.75$; $r^2_{TTCM} = 0.75$; Fig. 7) and body weight ($r^2_{LHTV} = 0.68$; $r^2_{TBST} = 0.62$; $r^2_{DHBL} = 0.72$; $r^2_{TTCM} = 0.64$; Fig. 8). This correlation was expressed through the regression equation LogF = a × LogW + b showing that the larger the fish, the more eggs were released.

DISCUSSION

The detection of stage IV and V oocytes in this fish during all months of the study showed that they were capable of spawning all year-round. In addition, the histological development in the adult stage of the gonads (stages IV and V) showed the appearance of various types of oocytes at the immature stage, such as O, PO, PVO, and SVO. Thereby it was seen that after the mature oocytes were released, the lower grade oocytes continued to develop and continue the cycle. It suggests that the reproductive form of this fish was spawning many times during the spawning season. This was considered a familiar property of the most economical fish species in the VMD. Some species belonging to this group include *Eleotris melanosoma* (*Vo & Tran, 2014*), *Oxyeleotris urophthalmus* (*Vo,*
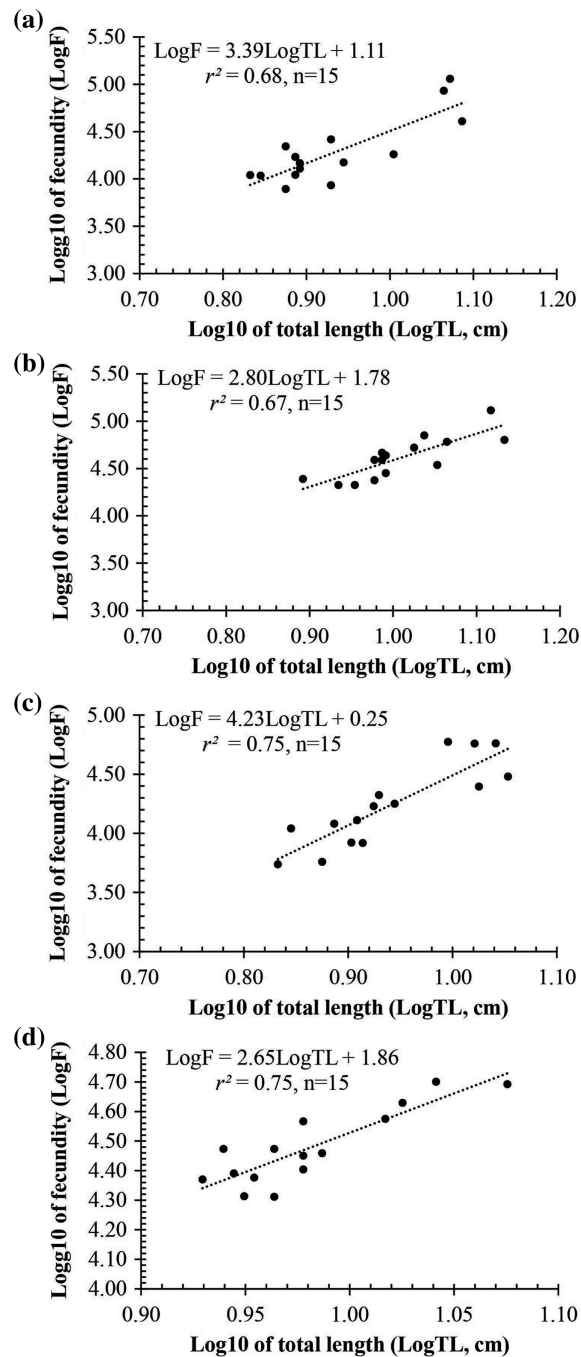
Figure 7 **The relationship between fecundity with the total length of *Acentrogobius viridipunctatus* at study sites.** (A) Long Huu-Tra Vinh, (B) Trung Binh-Soc Trang, (C) Dien Hai-Bac Lieu, (D) Tan Thuan-Ca Mau.

Tran & Mai, 2014), *Glossogobius giuris* (*Hossain, 2014*; *Dinh et al., 2022b*), *Butis butis* (*Dinh & Le, 2017*), *Stigmatogobius pleurostigma* (*Dinh & Tran, 2018*), *Periophthalmodon schlosseri* (*Tran et al., 2019*), *Glossogobius sparsipapillus* (*Nguyen et al., 2019*),
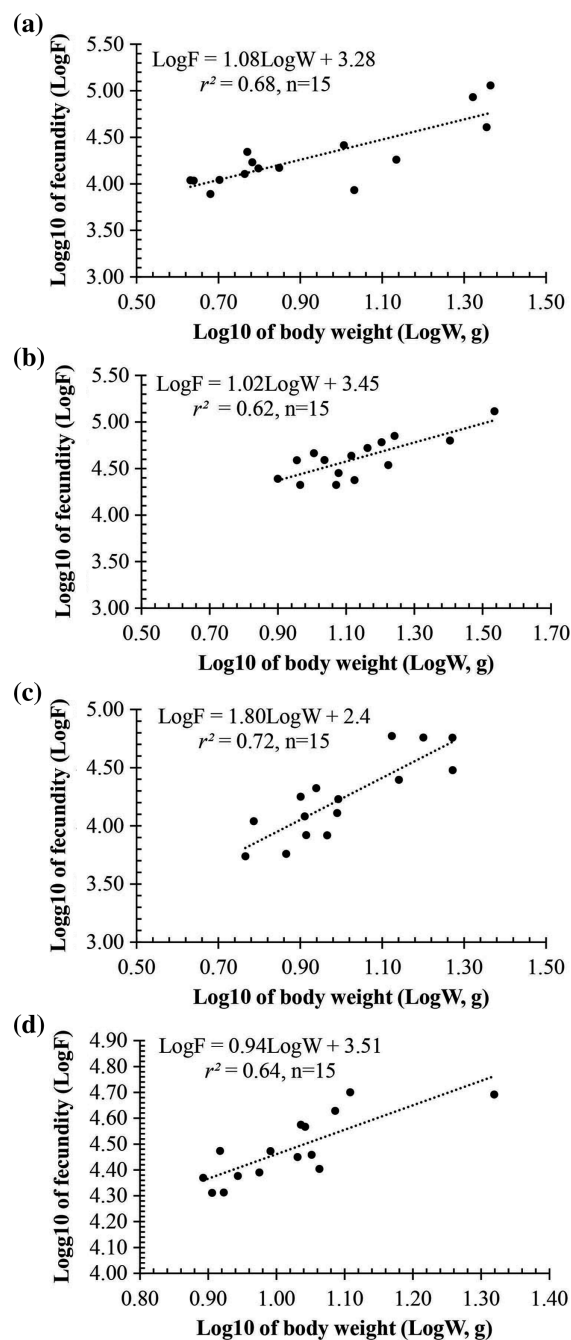
Figure 8 The relationship between fecundity with the body weight of *Acentrogobius viridipunctatus* at study sites. (A) Long Huu-Tra Vinh, (B) Trung Binh-Soc Trang, (C) Dien Hai-Bac Lieu, (D) Tan Thuan-Ca Mau.

Periophthalmodon septemradiatus (*Dinh et al., 2020*), and *Glossogobius aureus* (*Dinh et al., 2021b*).

Although *A. viridipunctatus* was capable of spawning year-round, the spawning season focuses on specific months. The primary spawning season of this fish showed different durations in the four study sites. The spawning season lasted for two months at TBST

Table 3 Spawning season, the length at first maturity, and fecundity in some species.

Species	Spawning season	L_m	Fecundity	References
Gobius paganellus	Mid-April to mid-June	–	–	Miller (1961)
Padogobius martensi	May and June	–	–	Cinquetti & Rinaldi (1987)
Acentrogobius plaumi	–	–	3,600–9,700	Baeck, Kim & Huh (2004)
Afurcagobius tamarensis	October to December	–	–	Cheshire et al. (2013)
Acentrogobius sp.	–	–	8,250	Soekiswo, Widyorini & Solichin (2014)
Butis butis	Year-round	6.82	46,017–78,500	Dinh & Le (2017)
Stigmatogobius pleurostigma	March to November	4.14	3,100–5,650	Dinh & Tran (2018)
Periophthalmodon schlosseri	Year-round	19.3–19.7	41,822–53,402	Tran et al. (2019)
Periophthalmodon septemradiatus	Year-round	6.05–7.23	5,916–11,451	Dinh et al. (2020)
Glossogobius sparsipapillus	July to September	6.50–6.78	17,918–28,700	Nguyen et al. (2021)
Glossogobius aureus	August to October	7.77–12.21	1,044–27,349	Dinh et al. (2021b)
Glossogobius giuris	April and in September	4.82–6.14	5,118–100,003	Dinh et al. (2022c)
Acentrogobius viridipunctatus	June to October in LHTV	6.6 ± 0.2 in LHTV	27,698 ± 7,983 in LHTV	This study
	June and July in TBST	7.5 ± 0.3 in TBST	46,592 ± 7,264 in TBST	
	August and September in DHBL	8.6 ± 0.2 in DHBL	23,271 ± 4,985 in DHBL	
	May to September in TTCM	9.4 ± 0.4 in TTCM	31,408 ± 2,515 in TTCM	

(June and July) and DHBL (August and September), whereas the spawning season at the other two sites was longer, with five months at LHTV (from June to October) and TTCM (from May to September). Thereby it was seen that in each different location, the fish had a spawning season adapted to the conditions of that area. In general, this fish species had a wet season spawning season in all study sites. Studies have found that some fish species exhibit a spawning season that starts early from the end of the dry season and lasts until the end of the wet season, such as *Glossogobius giuris* (Hossain, 2014; Dinh et al., 2022b), *Eleotris melanosoma* (Vo & Tran, 2014), and *Stigmatogobius pleurostigma* (Dinh & Tran, 2018). While in fish like *Pseudapocryptes elongatus* (Tran, 2008), *Oxyeleotris urophthalmus* (Vo, Tran & Mai, 2014), *Eleotris melanosoma* (Vo & Tran, 2014), *Butis koilomatodon* (Dinh et al., 2021a), and *Periophthalmodon schlosseri* (Tran et al., 2019) studies have shown a spawning season that lasts most months during the wet season (Table 3). *Glossogobius sparsipapillus* (Nguyen et al., 2021) and *Glossogobius aureus* (Dinh et al., 2021b) were two species found with a short spawning season of 2–3 months in the wet season like the results seen at TBST and DHBL. Not only do species in the VMD have a spawning season that occurs during the wet season, but some other fish species in the Gobiinae subfamily distributed worldwide also have a similar spawning season. *Gobius paganellus* distributed on the Isle of Man has a spawning season from mid-April to mid-June (Miller, 1961). *Glossogobius giuris* distributed in Payara River, Bangladesh has a

spawning season from April to June (*Qambrani et al., 2016*). The same species, but distributed in Patuakhali, Bangladesh has a later spawning season in December (*Roy et al., 2014*). The spawning season of *Afurcagobius tamarensis* distributed in Murray Mouth and Coorong (Australia) takes place from October to December every year (*Cheshire et al., 2013*), whereas, *Padogobius martensi* in Italy has a spawning season believed to take place in May and June (*Cinquetti & Rinaldi, 1987*).

The length at first maturity in *A. viridipunctatus* varied by study area. This may be due to environmental effects and changes in each study site that have affected the L_m of fish (*Dinh et al., 2021a*). In this fish, L_m was found in areas of high salinity, such as DHBL. However, the temperatures at the study sites have similar values. That suggests that the temperature was not the factor that changed the L_m of fish. According to the salinity data collected at these four sites during the study months, the salinity changes were similar to the changes of L_m. Specifically, in areas with high salinity, such as TTCM (28.7 ± 1.1‰) and DHBL (32.3 ± 1.0‰), L_m was significantly more prominent than in low salinity areas in TBST (19.8 ± 2.4‰) and LHTV (17.8 ± 2.3‰). This similarity showed that females in *A. viridipunctatus* had L_m that changes proportionally with salinity, revealing that this goby was observed to mature early at the site with higher salinity. Changes in L_m with salinity have been demonstrated in several other fish species. The L_m of *G. giuris* was adjusted to salinity variation as it matured earlier in saline areas (4.8 cm) than at the sampling sites with year-round freshwater conditions (6.1 cm) (*Dinh et al., 2022c*). In *Glossogobius aureus*, L_m tended to decrease from freshwater (12.5 ± 1.5 cm) to saltwater (10.5 ± 0.3 cm) (*Dinh et al., 2021b*). Similarly, in another species of mudskipper, *P. septemradiatus*, L_m increased from saltwater (8.2 cm) to freshwater (9.2 cm) (*Dinh et al., 2020*).

Fecundity is a characteristic factor for the development of each fish species. A high batch fecundity (*F*) was recorded in this fish with 5,481–130,683 eggs/female. This fish can easily repopulate with a high *F* with reduced numbers (*McDowall, 1997*). A species of the genus *Acentrogobius* was *Acentrogobius plaumi* in Korea, which showed relatively low fertility and ranged from 3,600 to 9,700 eggs (*Baeck, Kim & Huh, 2004*). Similarly, *Acentrogobius* sp. distribution in Indonesia also showed a low fecundity with 8,250 eggs (*Soekiswo, Widyorini & Solichin, 2014*) (Table 3). The results showed this fish species has a significantly higher reproductive capacity than other fish of the same genus. In addition, compared with some other fish species distributed in the VMD area, this fish has a significantly higher number of eggs. Some species with low fecundity can be mentioned as *Stigmatogobius pleurostigma* (3,100–5,650 egg/female) (*Dinh & Tran, 2018*), *Parapocryptes serperaster* (6,000–11,700 egg/female) (*Dinh et al., 2016*), *Trypauchen vagina* (4,000–12,750 egg/female) (*Dinh, 2018*), and *Periophthalmodon septemradiatus* (969–19,536 egg/female) (*Dinh et al., 2020*). Higher fecundity was observed in species such as *Pseudapocryptes elongatus* (2,100–29,400 egg/female) (*Tran, 2008*), *Boleophthalmus boddarti* (9,800–33,800 egg/female) (*Dinh, Nguyen & Nguyen, 2015*), and *Butis koilomatodon* 3,085–32,087 egg/female) (*Dinh et al., 2021a*). Moreover, *Glossogobius*

sparsipapillus (8,568–95,191 egg/female) (*Nguyen et al., 2021*) and *Glossogobius giuris* (5,118–100,003 egg/female) (*Dinh et al., 2022c*) were two species of goby fish in the family Gobiidae shown to have a high fecundity and roughly equivalent to *A. viridipunctatus* (Table 3). In particular, *Glossogobius giuris* distributed in Kissorgonj, Bangladesh displayed fecundity up to 14,987–716,400 eggs/female (*Hossain, 2014*). These results showed that fish fecundity depends not only on species but also on their living environment.

CONCLUSION

The results show the fish was a multi-spawner releasing eggs all year-round with a peak in the wet season. Fish size at first maturity varied by study site. It displayed a high fecundity that increased relatively with fish total length and body weight. In order to ensure sustainable exploitation, the fish should not be caught during the main spawning period, whilst the length at first capture should be $>L_m$.

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Competing Interests

The authors declare that they have no competing interests.

Author Contributions

- Quang Minh Dinh conceived and designed the experiments, performed the experiments, analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the article, and approved the final draft.
- Ton Huu Duc Nguyen conceived and designed the experiments, performed the experiments, analyzed the data, prepared figures and/or tables, authored or reviewed drafts of the article, and approved the final draft.
- Ngon Trong Truong conceived and designed the experiments, prepared figures and/or tables, and approved the final draft.
- Diep Xuan Doan conceived and designed the experiments, prepared figures and/or tables, and approved the final draft.
- Tien Thi Kieu Nguyen conceived and designed the experiments, prepared figures and/or tables, and approved the final draft.

Animal Ethics

The following information was supplied relating to ethical approvals (*i.e.*, approving body and any reference numbers):

This research was conducted with approval from The Scientific Committee of the School of Education, Can Tho University (No. Q2020-01/KSP) after animal welfare assessment.

Field Study Permissions

The following information was supplied relating to field study approvals (*i.e.*, approving body and any reference numbers):

In terms of the field trip, our research has no field permit requirement as we collect fish from the public-owned where local fishers collect fish daily. Moreover, we collect fish just once a month at each site, and the number of fish collection is within allowable limits of the animal assessment.

Data Availability

The raw measurements are available in the Supplemental File.

Supplemental Information

Supplemental information for this article can be found online at http://dx.doi.org/10.7717/peerj.14077#supplemental-information.

REFERENCES

Baeck GW, Kim JW, Huh S-H. 2004. Maturation and spawning of striped goby (*Acentrogobius pflaumi*) (Teleostei; Gobiidae) collected in the Gwangyang Bay, Korea. *Korean Journal of Fisheries and Aquatic Sciences* **37(3)**:226–231 DOI 10.5657/kfas.2004.37.3.226.

Bagenal TB. 1967. A short review of fish fecundity. In: Gerking SD, ed. *The Biological Basis of Freshwater Fish Production*. New York: John Wiley, 89–111.

Bauchot ML, Ridet JM, Diagne MD. 1989. Encephalization in Gobioidei (Teleostei). *Japanese Journal of Ichthyology* **36(1)**:63–74 DOI 10.11369/jji1950.36.63.

Cheshire K, Fredberg J, Ye Q, Short D, Earl J. 2013. Aspects of reproductive biology of five key fish species in the Murray Mouth and Coorong. Report number: SARDI Publication No. F2009/000014-3. SARDI Research Report Series No. 699. Adelaide: South Australian Research and Development Institute (Aquatic Sciences). *Available at https://www.researchgate.net/profile/Jason-Earl-2/publication/283658722_Aspects_of_reproductive_biology_of_five_key_fish_species_in_the_Murray_Mouth_and_Coorong/links/5642c18008aec448fa628f2a/Aspects-of-reproductive-biology-of-five-key-fish-species-in-the-Murray-Mouth-and-Coorong.pdf*.

Cinquetti R, Rinaldi L. 1987. Changes in the gonadal histology of *Padogobius martensi* (Pisces: Gobiidae) during the reproductive cycle. *Italian Journal of Zoology* **54(3)**:233–241 DOI 10.1080/11250008709355589.

Coleman M, Huh OK. 2012. *Major deltas of the world: a perspective from space*. Baton Rouge, LA, USA: Coastal Studies Institute, Louisiana State University.

Dinh QM. 2018. Aspects of reproductive biology of the red goby *Trypauchen vagina* (Gobiidae) from the Mekong Delta. *Journal of Applied Ichthyology* **34(1)**:103–110 DOI 10.1111/jai.13521.

Dinh QM, Lam TTH, Nguyen THD, Nguyen TM, Nguyen TTK, Nguyen NT. 2021a. First reference on reproductive biology of *Butis koilomatodon* in Mekong Delta, Vietnam. *BMC Zoology* **6(1)**:1–14 DOI 10.1186/s40850-021-00072-y.

Dinh QM, Le TTM. 2017. Reproductive traits of the duckbill sleeper *Butis butis* (Hamilton, 1822). *Zoological Science* **24(5)**:452–458 DOI 10.2108/zs170013.

Dinh QM, Nguyen TTG, Nguyen TKT. 2015. Reproductive biology of the mudskipper *Boleophthalmus boddarti* in Soc Trang. *Tap Chi Sinh Hoc* **37(3)**:362–369 DOI 10.15625/0866-7160/v37n3.6720.

Dinh QM, Nguyen THD, Nguyen-Ngoc L, Nguyen TTK. 2022a. Temporal variation in length-weight relationship, growth and condition factor of *Acentrogobius viridipunctatus* in the Mekong Delta, Viet Nam. *Regional Studies in Marine Science* **55(6)**:102545 DOI 10.1016/j.rsma.2022.102545.

Dinh QM, Qin JG, Dittmann S, Tran DD. 2016. Reproductive biology of the burrow dwelling goby *Parapocryptes serperaster*. *Ichthyological Research* **63(3)**:324–332 DOI 10.1007/s10228-015-0502-7.

Dinh QM, Tran NTT. 2018. Reproductive biological traits of the goby *Stigmatogobius pleurostigma* (Bleeker, 1849) from the Mekong Delta, Vietnam. *Indian Journal of Fisheries* **65(1)**:20–25 DOI 10.21077/ijf.2018.65.1.68188-04.

Dinh QM, Tran LT, Ngo NC, Pham TB, Nguyen TTK. 2020. Reproductive biology of the unique mudskipper *Periophthalmodon septemradiatus* living from estuary to upstream of the Hau River. *Acta Zoologica* **101(2)**:206–217 DOI 10.1111/azo.12286.

Dinh QM, Truong NT, Lam TTH, Nguyen THD, Tran NS, Nguyen TTK. 2021b. Evidencing some reproductive aspects of a commercial gobiid species *Glossogobius aureus* Akihito & Meguro, 1975 in Hau River, Vietnam. *Egyptian Journal of Aquatic Research* **47**:393–400 DOI 10.1016/j.ejar.2021.09.006.

Dinh QM, Truong NT, Sy Tran N, Nguyen THD. 2022b. Testicular development and reproductive references of *Glossogobius giuris* in Mekong Delta, Vietnam. *The Egyptian Journal of Aquatic Research* **48(1)**:61–66 DOI 10.1016/j.ejar.2021.09.005.

Dinh QM, Truong NT, Tran NS, Nguyen THD. 2022c. Ovarian and spawning reference, size at first maturity and fecundity of *Glossogobius giuris* caught along Vietnamese Mekong Delta. *Saudi Journal of Biological Sciences* **29(3)**:1911–1917 DOI 10.1016/j.sjbs.2021.10.030.

Fontoura NF, Braun AS, Milani PCC. 2009. Estimating size at first maturity (L50) from gonadossomatic index (GSI) data. *Neotropical Ichthyology* **7(2)**:217–222 DOI 10.1590/S1679-62252009000200013.

Ho NK, Nguyen TM, Dinh QM. 2021. Reproductive traits of the goby *Glossogobius sparsipapillus* Akihito & Meguro, 1976 in Tra Vinh province, Vietnam. *Journal of Environmental Biology* **42(SI)**:879–886 DOI 10.22438/jeb/42/3(SI)/JEB-23.

Hossain MS. 2014. Reproductive characteristics of Bele, *Glossogobius giuris* from Mithamoin Haor, Kissorgonj, Bangladesh. *World Journal of Fish and Marine Sciences* **6(6)**:537–543 DOI 10.5829/idosi.wjfms.2014.06.06.86210.

King M. 2013. *Fisheries biology, assessment and management*. Hoboken: John Wiley & Sons.

Komolafe OO, Arawomo GAO. 2007. Reproductive strategy of *Oreochromis niloticus* (Pisces: Cichlidae) in Opa reservoir, Ile-Ife, Nigeria. *Revista de Biología Tropical* **55(2)**:595–602 DOI 10.15517/rbt.v55i2.6034.

Le T, Nguyen MT, Nguyen VP, Nguyen DC, Pham XH, Nguyen TS, Hoang VC, Hoang PL, Le H, Dao NC. 2006. Provinces and City in the Mekong Delta. In: Le T, ed. *Geography of Provinces and Cities in Vietnam*. Vol. VI. Ha Noi: Education Publishing House, 49–94.

Le TNT, Tong TN. 2011. Characteristics of the growth and nutrification of *Acentrogobius viridipunctatus* (Valenciennes, 1837) at lagoon system, Thua Thien Hue Province. *Hue University Journal of Science* **67**:153–163 DOI 10.26459/jard.v67i4.3191.

Lloret J, Rätz H-J. 2000. Condition of cod (*Gadus morhua*) off Greenland during 1982–1998. *Fisheries Research* **48(1)**:79–86 DOI 10.1016/s0165-7836(00)00111-9.

Mai DY, Vu TT, Bui L, Tran MT. 1979. *Ichthyologist*. Hanoi: Publishing House University and Professional High School.

McDowall RM. 1997. The evolution of diadromy in fishes (revisited) and its place in phylogenetic analysis. *Reviews in Fish Biology and Fisheries* **7(4)**:443–462 DOI 10.1023/A:1018404331601.

Miller PJ. 1961. Age, growth, and reproduction of the rock goby, *Gobius paganellus* L., in the Isle of Man. *Journal of the Marine Biological Association of the United Kingdom* **41(3)**:737–769 DOI 10.1017/S0025315400016283.

Miller PJ. 1984. The topology of gobioid fishes. In: Potts GW, Wootton RJ, eds. *Fish Reproduction: Strategies and Tactics*. Orlando, London, United Kingdom: Academic Press, 119–153.

Morey G, Moranta J, Massutí E, Grau A, Linde M, Riera F, Morales-Nin B. 2003. Weight—length relationships of littoral to lower slope fishes from the western Mediterranean. *Fisheries Research* **62(1)**:89–96 DOI 10.1016/S0165-7836(02)00250-3.

Nguyen NT. 2000. *Fauna of Vietnam – Gobioidei.* Vol. 5. Ha Noi: Science and Technics Publishing House.

Nguyen HDT, Nguyen TTH, Tran CC, Dang HT, Nguyen TNY, Dinh QM. 2019. Morphological and histological characteristics of testis of the goby *Glossogobius sparsipapillus* living from coastal Estuaries from Bac Lieu to Ca Mau. *VNU Journal of Science: Natural Sciences and Technology* **35(4)**:81–87 DOI 10.25073/2588-1140/vnunst.4958.

Nguyen HDT, Nguyen TTH, Tran CC, Nguyen TNY, Dinh QM. 2021. Ovarian development, spawning characteristics, size at first mature and fecundity of *Glossogobius sparsipapillus* (Gobiiformes: Gobiidae) living along estuarine and coastal regions in the Mekong Delta, Vietnam. *Acta Zoologica Bulgarica* **73(2)**:253–260.

Pham TL, Tran DD. 2004. *Research methods in fish biology*. Can Tho: Can Tho University.

Plaza G, Sakaji H, Honda H, Hirota Y, Nashida K. 2007. Spawning pattern and type of fecundity in relation to ovarian allometry in the round herring *Etrumeus teres*. *Marine Biology* **152(5)**:1051–1064 DOI 10.1007/s00227-007-0756-3.

Qambrani GR, Soomro AN, Palh ZA, Baloch WA, Tabasum S, Lashari KH, Qureshi MA. 2016. Reproductive biology of *Glossogobius giuris* (Hamilton), in Manchar Lake Sindh, Pakistan. *Journal of Aquaculture Research & Development* **7**:392–394 DOI 10.4172/2155-9546.1000392.

Roy A, Hossain MS, Rahman ML, Salam MA, Ali MM. 2014. Fecundity and gonadosomatic index of *Glossogobius giuris* (Hamilton, 1822) from the Payra River, Patuakhali, Bangladesh. *Journal of Fisheries* **2(2)**:141–147 DOI 10.17017/j.fish.81.

Soekiswo YA, Widyorini N, Solichin A. 2014. Aspek biologi ikan mendo (*Acentrogobius* sp.) di waduk malahayu kabupaten brebes. *Management of Aquatic Resources Journal (MAQUARES)* **3(4)**:154–160 DOI 10.14710/MARJ.V3I4.7050.

Teichert N, Valade P, Fostier A, Lagarde R, Gaudin P. 2014. Reproductive biology of an amphidromous goby, *Sicyopterus lagocephalus*, in La Réunion Island. *Hydrobiologia* **726(1)**:123–141 DOI 10.1007/s10750-013-1756-6.

Thai NT. 2015. Research of the biodiversity of the Mekong Delta fish family and their change by impacts of climate change and social-economic development. PhD thesis, Vietnam Academy of Science and Technology, Hanoi, Vietnam.

Thai NT, Hoang DD, Nguyen HN, Phan DD, Nguyen BC, Nguyen VS, Trinh TL, Nguyen LP, Thai TMT, Huynh VNQ, Tran HA, Nguyen CT, Le VT, Le TNN. 2012. Assessment of impacts of climate change and sea-level rise on coastal habitat communities and recommendation of Adaptation resolution. Ben Tre Province: The National Target Program of Adaptation to Climate Change.

Tran DD. 2008. Some aspects of biology and population dynamics of the goby *Pseudapocryptes elongatus* (Cuvier, 1816) in the Mekong Delta. PhD thesis, Malaysia, Universiti Malaysia Terengganu.

Tran DD, Shibukawa K, Nguyen TP, Ha PH, Tran XL, Mai VH, Utsugi K. 2013. *Fishes of Mekong Delta, Vietnam*. Can Tho: Can Tho University Publisher.

Tran LT, Son MS, Vo CNM, Hoang HD, Dinh QM. 2019. Reproductive biology of *Periophthalmodon schlosseri* (Pallas, 1770) along the coastline in Soc Trang and Bac Lieu. *Tap Chi Sinh Hoc* **41(2se)**:229–240 DOI 10.15625/0866-7160/v41n2se.14153.

Vo TT, Tran DD. 2014. Some of characteristics reproductive biology of fish *Eleotris melanosoma* distributed along the Hau River. *Can Tho University Journal of Science* **1(SI)**:115–122.

Vo TT, Tran DD, Mai VV. 2014. Some reproductive biology of *Oxyeleotris urophthalmus* (Bleeker, 1851) distributed along Hau River. In: *The 2nd Conference on Marine Biology and Sustainable Development*, Hai Phong: Science and Technology Publishing House, 515–522.