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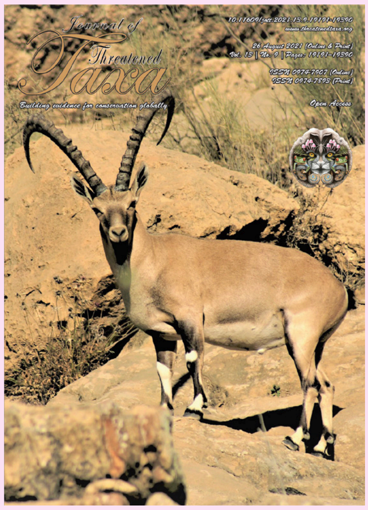

COMMUNICATION

ON THE REPRODUCTIVE BIOLOGY OF THE INVASIVE ARMOURED SAILFIN CATFISH *PTERYGOPLICTHYS PARDALIS* (CASTELNAU, 1855) (SILURIFORMES: LORICARIIDAE) FROM THE NATURAL DRAINAGES IN THIRUVANANTHAPURAM, INDIA

Smrithy Raj, Suvarna S. Devi, Amrutha Joy & A. Biju Kumar

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COMMUNICATION

On the reproductive biology of the invasive Armoured Sailfin Catfish *Pterygoplicthys pardalis* (Castelnau, 1855) (Siluriformes: Loricariidae) from the natural drainages in Thiruvananthapuram, India

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Abstract: The present paper deals with the breeding biology of the invasive fish *Pterygoplicthys pardalis* from the natural drainages of Thiruvananthapuram, Kerala. The specimens were collected from Amayizhanchan Thodu, a natural drainage running through the heart of the city. A total of 145 males and 142 females were collected from January to December 2018. The sex ratio was determined monthly as the percentage of males to females (M: F). Monthly mean gonadosomatic index (GSI) values were compared using R stat, and GSI was plotted monthly to identify the spawning seasons. The gonads were examined and different stages of maturity were noted using standard methods. The length at first maturity was also found out. The fish exhibit courtship behaviour and the eggs are deposited in burrows and also along the crevices in the granite walls; the burrows are guarded by the male fish till the young ones are hatched out. The sex ratio showed an average mean value of 1.04: 1 and showed no significant departure. The size at first maturity was 23.9 cm standard length. The ova diameter studies show the presence of ripe ovaries throughout the year, with peaks during March and April and between August and September and in December, indicating the fish is a batch spawner. The absolute fecundity ranges from 923 to 14,777 eggs, and the relative fecundity ranges 0.0142–0.0015. Regression analysis showed a significant relationship (P <0.001) between absolute fecundity and the total length, the total body weight, and ovary weight. The strong breeding behaviour, the presence of accessory respiratory organs, the absence of natural enemies and parental care makes *Pterygoplichthys pardalis* a successful invader in the natural drainage. More biological studies are needed for the successful eradication of the species from the invaded ecosystem.

Keywords: Breeding biology, breeding behaviour, fecundity, invasive alien species, invasion biology.

Malayalam: കേരളത്തിൽ തിരുവനന്തപുരം നഗരത്തിലെ സ്വാഭാവിക നീർച്ചാലുകളിൽ ജീവിക്കുന്ന ആമസോൺ സക്കർ മത്സ്യത്തിന്റെ (പ്റ്ററിഗോപ്ലിക്ത്തിസ് പർഡാലിസ്)പ്രജനന ജീവശാസ്ത്രത്തെക്കുറിച്ചാണ് ഈ പ്രബന്ധത്തിൽ വിവരിക്കുന്നത്. നഗരത്തിന്റെ ഹൃദയഭാഗത്തുകൂടി ഒഴുകുന്ന പ്രകൃതിദത്ത ഡ്രെയിനേജ് ആയ അമയിഴഞ്ചാൻ തോട്ടിൽ നിന്നാണ് സാമ്പിളുകൾ ശേഖരിച്ചത്. 2018 ജനുവരി മുതൽ ഡിസംബർ വരെ മൊത്തം 145 ആൺ മത്സ്യങ്ങളെയും 142 പെൺ മത്സ്യങ്ങളെയും ശേഖരിച്ചു. ലിംഗ അനുപാതം പ്രതിമാസം ആൺ മത്സ്യങ്ങളുടെ ശതമാനം പെൺ മത്സ്യങ്ങളുടേതുമായി താരതമ്യം ചെയ്തു. പ്രതിമാസ ശരാശരി ഗോണാഡോസോമാറ്റിക് സൂചിക (GSI) മൂല്യങ്ങൾ R സ്റ്റാറ്റ് ഉപയോഗിച്ച് താരതമ്യം ചെയ്തു. മുട്ടയിടുന്ന സീസണുകൾ തിരിച്ചറിയാൻ GSI പ്രതിമാസം ഗ്രാഫുകളിൽ രേഖപ്പെടുത്തി. ജനനഗ്രന്ഥികൾ (ഗോണാഡുകൾ) പരിശോധിക്കുകയും ശാസ്ത്രീയ രീതികൾ ഉപയോഗിച്ച് വളർച്ചയുടെ വിവിധ ഘട്ടങ്ങൾ പരിശോധിക്കുകയും ചെയ്തു, ആദ്യമായി പ്രായപൂർത്തിയാകുന്ന ദൈർഘ്യവും കണ്ടെത്തി. മത്സ്യങ്ങൾ കോർട്ട്ഷിപ്പ് സ്വഭാവം പ്രദർശിപ്പിക്കുകയും മുട്ടകൾ മാളങ്ങളിലും കരിങ്കൽ ഭിത്തികളിലെ വിള്ളലുകളിലും നിക്ഷേപിക്കുകയും ചെയ്യുന്നു; കുഞ്ഞുങ്ങൾ വിരിയിക്കുന്നതുവരെ ആൺമത്സ്യം കാവൽ നിന്ന് മുട്ടകളെ സംരക്ഷിക്കുന്നു. ലിംഗാനുപാതത്തിന്റെ ശരാശരി മൂല്യം 1.04:1 ആണ്. ആദ്യമായി പ്രായപൂർത്തിയാകുന്ന വലുപ്പം 23.9 സെന്റിമീറ്റർ സ്റ്റാൻഡേർഡ് നീളമായിരുന്നു. അണ്ഡത്തിന്റെ വ്യാസത്തെപ്പറ്റിയുള്ള പഠനങ്ങൾ വർഷം മുഴുവനും (മാർച്ച്, ഏപ്രിൽ മാസങ്ങളിലും ഓഗസ്റ്റ് മുതൽ സെപ്റ്റംബർ വരെയും ഡിസംബറിലും) പൂർണവളർച്ചയെത്തിയ അണ്ഡത്തിന്റെ സാന്നിധ്യം കാണിക്കുന്നു. ഇത് മത്സ്യം വർഷം മുഴുവനും മുട്ടയിടാനുള്ള (ബാച്ച് സ്പോൺ) സാധ്യത സൂചിപ്പിക്കുന്നു. മുട്ടയുടെ കേവലമായ എണ്ണം 923 മുതൽ 14,777 മുട്ടകൾ വരെയാണ്; ആപേക്ഷികമായ മൂല്യം 0.0142-0.0015 വരെയാണ്. മൊത്തം മുട്ടകളുടെ എണ്ണവും, മൊത്തം ദൈർഘ്യവും, മൊത്തം ശരീരഭാരവും അണ്ഡാശയ ഭാരവും തമ്മിൽ സാരവത്തായ ബന്ധം (പി <0.001) റിഗ്രഷൻ വിശകലനം ചൂണ്ടിക്കാണിക്കുന്നുണ്ട്. ശക്തമായ പ്രജനനരീതികളും, സഹായക ശ്വസനാവയവങ്ങളുടെ സാന്നിധ്യവും, സ്വാഭാവിക ശത്രുക്കളുടെ അഭാവവും രക്ഷാകർതൃ പരിചരണവും ആണ് ആമസോൺ സക്കർ മത്സ്യത്തെ പ്രകൃതിദത്ത നീർച്ചാലുകളിൽ വിജയകരമായി ആധിപത്യം സ്ഥാപിക്കുന്നതിന് സഹായിച്ചിരിക്കുന്നതെന്ന് അനുമാനിക്കാം. അധിനിവേശ ആവാസവ്യവസ്ഥയിൽ നിന്ന് ഇവയെ വിജയകരമായി തുടച്ചുനീക്കുന്നതിന് കൂടുതൽ ജീവശാസ്ത്രപരമായ പഠനങ്ങൾ ആവശ്യമാണ്.

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INTRODUCTION

Reproductive biology is one of the key features considered for the invasion biology studies of fish (Feiner et al. 2012; Gutkowsky & Fox 2012; Zahorska et al. 2013; Horkova & Kovac 2015). The suckermouth armoured catfishes of the family Loricariidae, especially the genus *Pterygoplichthys* T.N. Gill, native to inland water bodies of South America, is an emerging invasive fish globally (Orfinger & Goodding 2018), also, one of the most popular and commonly traded aquarium fish in India (Knight 2010). The sailfin armoured catfish species recorded from the natural water bodies of India include *P. anisitsi, P. disjunctivus, P. multiradiatus, P. pardalis,* and possible hybrids (Sinha et al. 2010; Singh 2014; Kumar et al. 2015; Soundararajan et al. 2015; Sandilyan et al. 2016; Hussan et al. 2018).

The high invasiveness shown by this species is primarily because of its unique biological features, which include the ability to survive in water with varying levels of flow regimes (Nico & Martin 2001; Welcomme & Vidthayanom 2003; Chavez et al. 2006; Nico et al. 2012), temperature and dissolved oxygen using accessory respiration with diverticula of the gastrointestinal tract (Armbruster 1998; da Cruz et al. 2013), and pH (Mendoza et al. 2009; Parente et al. 2017), a broad range of diet (German et al. 2010) and the absence of natural predators (Nico & Martin 2001; Gibbs et al. 2008), which enable them to survive in the invaded ecosystems. They are salt-tolerant and survive well in brackish water regions (Mendoza et al. 2009; Kumar et al. 2018). They also have efficient reproductive strategies, including parental care and deter predation by possessing bony plates that cover their body (Hoover et al. 2004; Liang et al. 2005; Wei et al. 2017). One such extreme measure of parental care is exhibited by the male members of the genus *Pterygoplichthys.* It is the burrows they dig out on the river banks, and the female will lay eggs that are guarded by males (Nico et al. 2009; Alamdin & Jumawan 2016). Thus, site selection for spawning, nest building and caring of eggs and the young ones are a complex set of reproductive behaviour among loricariids (Covain & Fisch-Muller 2007).

The reproductive traits of *Pterygoplichthys* spp such as size at maturity, spawning, sex-ratio, ova diameter, fecundity and reproductive plasticity are studied to have a better knowledge on the reproductive dynamics (Jumawan and Herrera, 2014; Gibbs et al., 2017) which is a prerequisite for the management measures of an invasive species (Hoover et al., 2005; Kopp et al. 2009). The reproductive patterns of *P. disjunctivus* from Florida were extensively analysed by Gibbs et al. (2017) for 10 years. In contrast, studies on the particular aspects of reproduction have been published by Rueda-Jasso et al. (2013), Jumawa & Herrera (2014) on *P. disjunctivus* from Mexico and Philippines, Samat et al. (2016) on *P. pardalis* from Malaysian waters, and Cook-Hildreth et al. (2016) from Texas waters. Wei et al. (2017) studied the maturity of *Pterygoplichthys* spp., a hybrid from China and reported self-sustaining populations in the drainages of the area.

The negative impacts caused by *Pterygoplichthys* spp. in the invaded ecosystems, include siltation problems, bank erosion in rivers and streams, competition with native species for food and space, consumption of the eggs of native and threatened species, displacement of vegetation and disturbance to the breeding grounds of native fish and economic losses to the fishermen including damage to the fishing gears, are reported earlier by many researchers (see Bunkley-Williams et al. 1994; Hoover et al. 2004; Chavez et al. 2006; Wakida-Kusunoki et al. 2007; Hossain et al. 2008; Cook-Hildreth 2009; Krishnakumar et al. 2009; Mendoza et al. 2009; Nico et al. 2009; Capps & Flecker 2013).

The reproductive parameters of oviparous fish outside their natural range will supplement the evidence to comprehend their establishment in the invaded ecosystems (Samat et al. 2016). Such studies, especially on reproductive biology, are required to better understand the natural history and reproductive plasticity, which are necessary tools for effectively managing this emergent invasive species. Despite the increasing numbers of publications regarding the invasion range extension of *Pterygoplichthys* spp. in various biogeographic regions, a knowledge gap on the reproductive biology of this invasive fish persists in India. Hence to address this significant gap, we investigated the reproductive biology of invasive loricariid fish *Pterygoplicthys pardalis* with a description of their breeding behaviour in natural streams.

MATERIALS AND METHODS

Collection Site and Sampling

The fishes for the study were collected from Amayizhanchan Thodu, (8.484711 – 8.566293^{0}N; 76.933348 – 76.949982^{0}E), a natural drainage of 3.4 km^2 in Thiruvananthapuram city, Kerala (Figure 1; Image 1). A total of 145 males and 142 females were collected fortnightly from January to December 2018 using a cast net, 5-m long and 3.8-cm mesh size. The fishes were

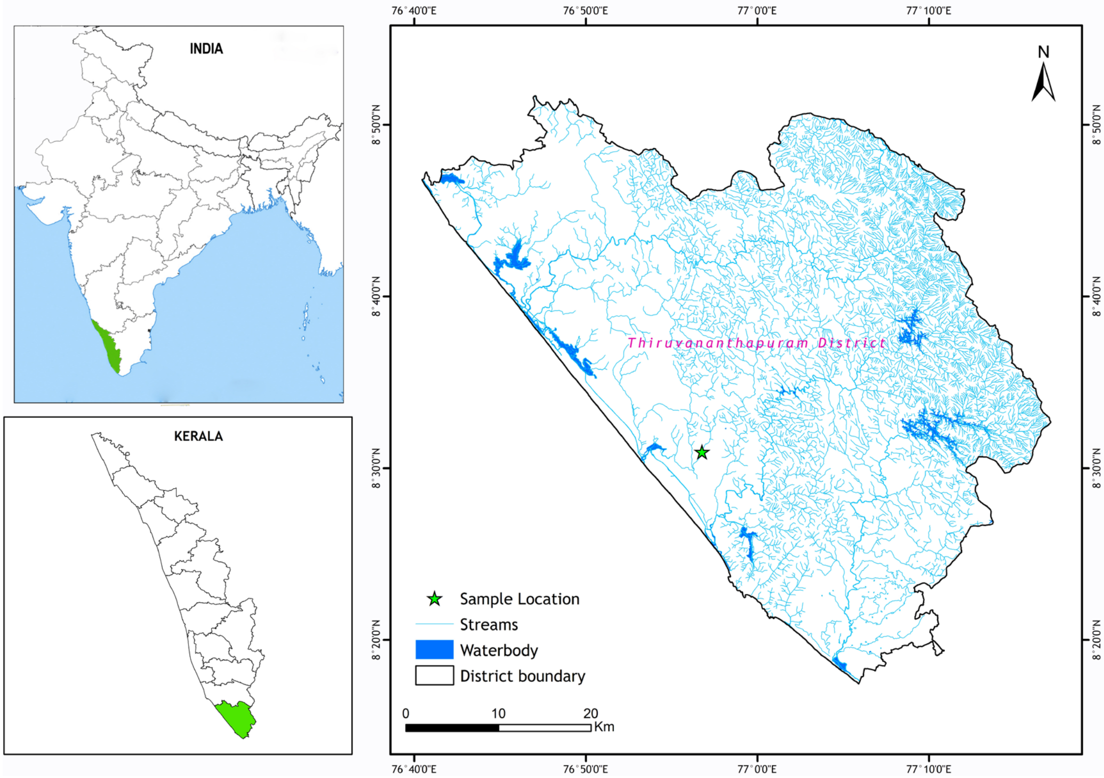

Figure 1. Sampling location at Amayinzhanthodu, Thiruvananthapuram, Kerala, India.

Image 1. *Pterygoplicthys pardalis*: A—lateral view | B—catch in cast net | C—specimens caught in single cast netting. © Biju Kumar

dissected, but before that, they were kept for four hours in a deep freezer. The sex ratio was determined monthly as the percentage of males to females (M: F). The burrow structure was captured with a digital camera, and its width, diameter, depth, the maximum height of entrance, shape of the tunnel, condition of the burrow, and occupancy of burrows were noted (Image 2).

Measurements of total length (TL, cm), standard length (SL), total weight (TW, g) and gonad weight (GW, g) were taken for male and female specimens. Total length and standard length were measured to the nearest 0.1 cm, and weight to the nearest gram 0.1 g using a digital balance. Gonadosomatic index (GSI) was calculated as GSI = [(gonad weight)/(total weight of fish)] × 100 (Vazzoler, 1996). Monthly mean GSI values were compared using R stat, and GSI was plotted monthly to identify the spawning seasons.

The fishes were dissected to remove their gonads. Stages of maturity were determined following (Mazzoni & Caramaschi 1997) and ova diameter observations. The number of fishes in various stages include: immature (16), maturing (33), ripe (45), regressing spent (18), and recovering spent (30). Ovaries were excised, weighed to the nearest gram, and preserved in 10 per cent formalin for later assessment. Spawning type was designated according to the stage of the ripe and spent ovaries. Testis was characterized using a visual-based macro scale of maturity based on testicular size, colour, and swelling (Lowerre-Barbieri et al. 2011).

The length at first maturity (L_{50}) was defined as the total length, where 50% of all individuals are predicted to be adults (Vazzoler 1996). For ova diameter studies, the diameter of maturing and mature eggs (Vitellogenic oocytes) was measured to the nearest 0.01 mm with digital calipers and from the photos taken using Leica stereo zoom dissecting microscope.

To evaluate the absolute fecundity (AF), ripe ovaries which are in late-maturing stages to ripe ones (mid to late vitellogenic phase- Patiño & Sullivan 2002) were taken for the studies, and 100 mg sub-samples of eggs from the anterior, middle, and posterior regions were weighed and counted under a binocular stereo microscope. The counted eggs were averaged and extrapolated for the entire ovaries using the formula: F= nG/g where F is the fecundity, n is the number of eggs in the sub-sample, G, ovary weight and g the subsample weight. The number and size of eggs were determined under a binocular dissecting microscope. The gonadal cycle also has been estimated based on macroscopic observation, and five stages have been described (Araújo et al. 1998; Duarte & Araújo 2002). To evaluate the relative fecundity (RF), the absolute fecundity is divided by the total weight of the fish (Bagenal 1978). The relationship between relative fecundity (RF) and the variables total body length (TL), the total fish weight (FW), and ovary weight (OW) were estimated.

RESULTS

Breeding behaviour

Pterygoplichthys pardalis that invaded the natural drainages in Thiruvananthapuram city (Images 1A–C) excavate burrows for breeding <https://www.youtube.com/watch?v=h5VZ-SVw7Wc>. Our observations reveal that the male fish excavate burrows (Image 2A, B) before spawning for laying eggs.

The burrows (older/used) above the water level are small and triangular to circular, measuring 10–20 cm in diameter. In contrast, those below water are larger without definite shape, measuring 30–50 cm in width. The horizontal burrows are 120–140 cm deep, with the slope extending downwards into the bank. Courtship behaviour was exhibited in the form of circular movements near the burrows (Images 2C–E), by rubbing their bodies with the flashing of water, and in a few cases, multiple males take part in the process, and the eggs are guarded till the young ones emerge from the nest (Image 2F).

It was also observed that this species also selects crevices in the granite walls of the stream to deposit eggs, which may be one of the reasons for their higher rate of survival in the drainages of Thiruvananthapuram city in Kerala.

Sex ratio

A total of 145 males and 142 females of *P. pardalis* collected from the study site showed sex ratio (M: F) of an average mean value of 1.04: 1. The ratio was tested by chi-square analysis for differences from hypothetical ratio 1: 1, which showed no significant departure.

Stages of the reproductive cycle

Females (N= 142) ranging from SL 17.2 cm (TL 24.4 mm) to 45.6 cm (TL 58.4 mm) were considered for ascertaining reproductive stages. Mature ovaries exhibited asymmetry, whereas immature ones were symmetrical. It was observed that the larger the size of the individual, the greater would be the occupancy of the ripe ovary in the abdominal cavity and vice versa. Based on macroscopic and microscopic examination (Table 1) and ova diameter studies, five developmental stages of

Table 1. Macroscopic and microscopic characteristics of ovarian maturity in *Pterygoplichthys pardalis*.

Stages of maturity	Macroscopic and microscopic features
1. Immature	Tiny ovaries, ranging 11.08–32.5mm, SL mostly less than 33.4 cm occupying only a tiny percentage of the body cavity. Ovary thin transparent to light pink, no visible oocytes
2. Maturing	Size of the ovary ranges between 33–59.36 mm with tiny granules to less yolky oocytes with SL around 35 cm, colour opaque to pale yellow
3. Ripe	Highly vascularised, thin-walled large asymmetrical ovaries occupying mostly half of the body cavity, brightly orange coloured fully yolked oocytes, size (1.88–2.81 mm)
4. Regressing spent	Large flaccid thick-walled ovaries usually with very few or no vitellogenic oocytes, vascularisation still visible but less, thick brush-like fimbriae projects from the ovarian wall into the lumen.
5. Recovering spent	Ovaries purple to dark pink with thick inner ovarian walls with slight vascularisation with small oocytes of different diameters, absence of ripe oocytes

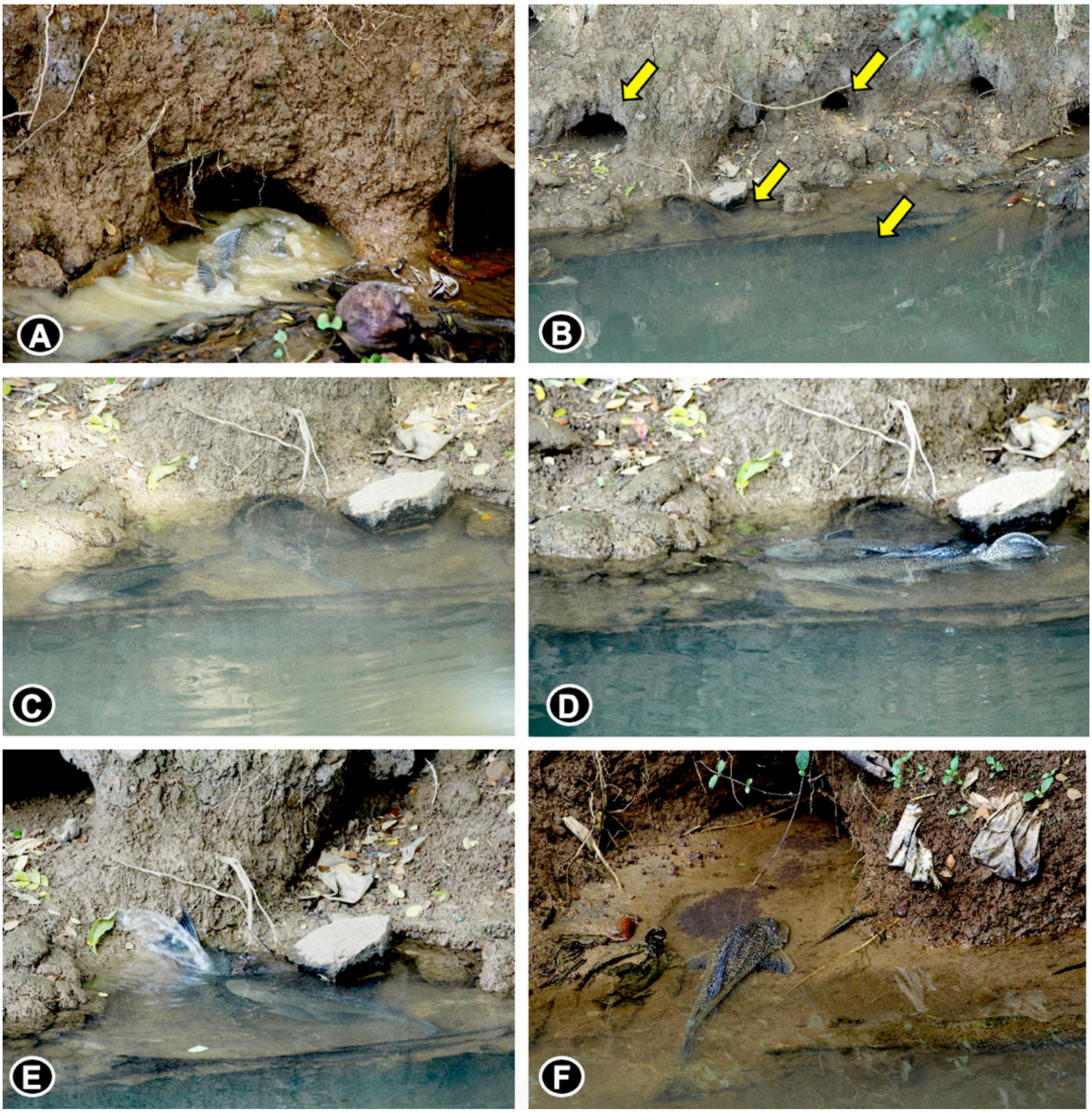

Image 2. Breeding behaviour of *Pterygoplicthys pardalis*: A—*P. pardalis* making burrows in canal margins | B—old and new burrows above and below the water level | C, D, E—courtship behaviour of *P. pardalis* around the burrow opening | F—*P. pardalis* guarding the burrow. <https://www.youtube.com/watch?v=h5VZ-SVw7Wc> © Biju Kumar

gonads (immature, maturing, ripe, regressing spent, and recovering spent) were identified in female fish (Table 1, Image 3 A–D, 4 A–F). In males, three maturity stages were identified: immature, maturing and mature, depending upon the colour and size of testes (Image 5 A–F).

The macroscopic and ova diameter studies showed *P. pardalis* has an intermittent spawning period with ripe ovaries throughout the year, maximum during March and April. Immature stages were noticed from May onwards, followed by maturing ones from June to November with a peak in July. The ripe ovaries were present in all months except February, with a maximum during March and April. Accordingly, the spent ovaries (regressive and recovering ones) and immature stages were noticed in the subsequent months, which mean the fish spawns during the rains (as summer rains occur during May). The second set of a large number of ripe ovaries was visible in August and December, with regressing and recovering spent in September and January, respectively showing the extended spawning season for the fish.

Length at first maturity

The minimum length to attain sexual maturity with vitellogenic oocytes was noticed in females at a standard length of 23.9 cm. The length at which 50% of the fish gets matured is at 36.56 cm (Figure 3).

Fecundity

To understand the absolute fecundity (AF), the ovary (left lobe) of ripe fishes were dissected, and the ripe ova were fully counted and extrapolated. The absolute fecundity of fish ranges from 923 (TL 393 mm; SL 294 mm) to 14,777 eggs (TL 516 mm; SL 414mm), and the relative fecundity ranges from 0.0142 (TL 459 mm) to 0.0015 (TL 393 mm). In mature fish, both the ovaries (left and right lobe) showed a clear asymmetry inside the abdominal cavity (Image 4A).

Gonado-Somatic index and ova diameter

The GSI of females showed three peaks, with the first one in March–April, the second in August–September and a third in December, which indicates an extended batch spawning nature of the fish (Figure 4). Similar to GSI, three peaks could also be observed with regard to the mean ova diameter, confirming an extended spawning season for *P. pardalis* (Figure 5). The maximum ova diameter obtained in the present study was 3.75 mm. While comparing ova diameter frequencies corresponding to different maturity stages, maximum oocyte diameter (mean) was noticed in the ripe stage (Figure 6).

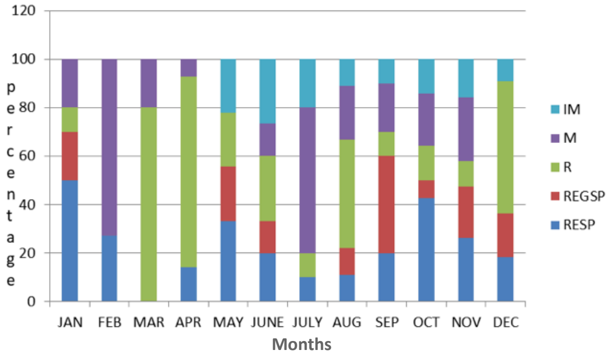

Figure 2. The maturity stages of female *Pterygoplichthys pardalis* in various months under study: IM—Immature | M—Maturing | R—Ripe | REGSP—Regressing spent | RESP—Recovering spent.

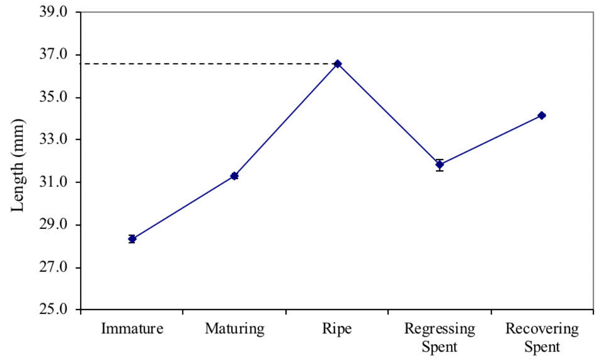

Figure 3. *P. pardalis*: Length at first maturity of female in the present study.

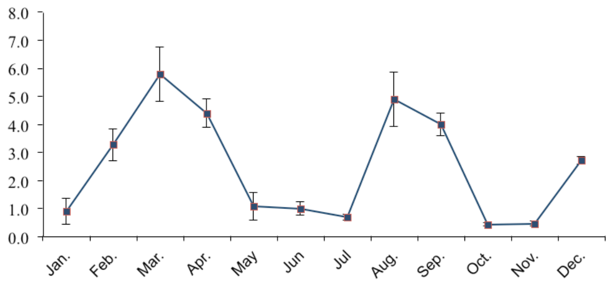

Figure 4. Monthly variations of GSI in *P. pardalis* (female) from the present study. Error bars represent SD.

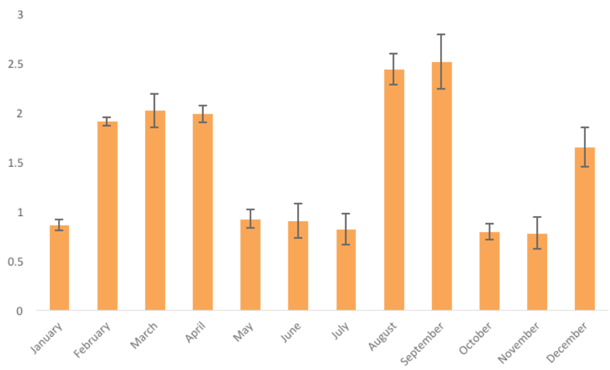

Figure 5. Mean ova diameter (mm) of *P. pardalis* in different months during the present study. Error bars represent SD.

Image 3. Maturity stages of female *Pterygoplichthys pardalis*: A—immature ovary in situ | B—immature ovary | C—maturing ovary in situ | D—maturing ovary. © Smrithy Raj

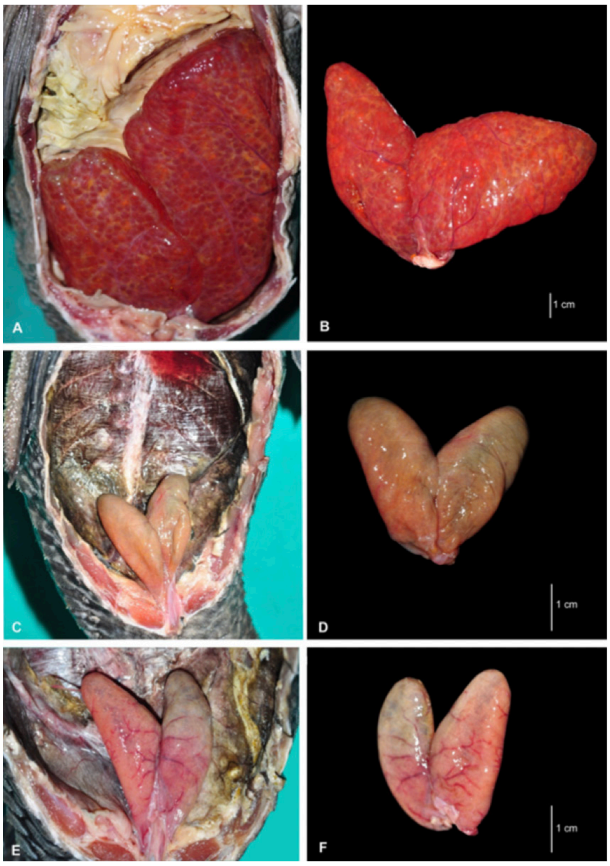

Image 4. Maturity stages of female *Pterygoplichthys pardalis*: A—ripe ovary in situ | B—ripe ovary | C—spent/regressing ovary in situ | D—spent/regressing ovary | E—recovering spent ovary in situ | F—recovering spent ovary. © Smrithy Raj

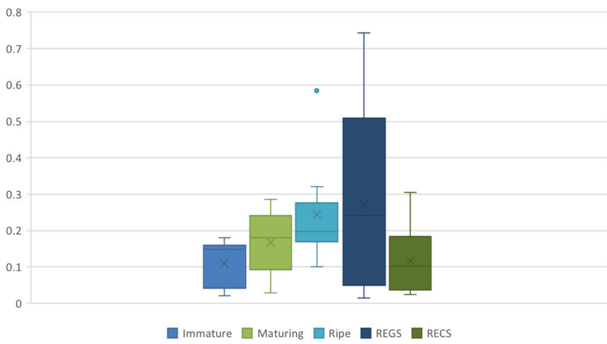

Figure 6. Ova diameter (mm) and maturity stages of *P. pardalis* in the present study. Error bars represent SD.

Regression analysis

Regression analysis showed a significant relationship (P <0.001) between absolute fecundity and the total length, the total body weight and ovary weight (Figures 7–9). As the total length, total body weight, and ovary weight of fish increases, the fecundity does not increase correspondingly due to lesser 'b' value (2.0482, 0.8214, 0.6944).

DISCUSSION

Although the suckermouth armoured catfish of the genus *Pterygoplichthys* is an emerging global invader, the details of its invasion biology are also being studied from its extended invasion ranges (Orfinger & Goodding 2018), the unique breeding behaviour of *Pterygoplichthys* spp. by excavating burrows in river banks have been documented from Florida (Nico et al. 2009), Mexico (Lienart et al. 2013) and the Philippines (Almadin & Jumawan 2016). Similar breeding behaviour was also observed in the present study. The females use the burrows dug by the males to deposit eggs and are guarded by the males till the young ones emerge from the nest; similar behaviour was also noted by Mazzoni et al. (2002), Power (2003), and Liang et al. (2005), which establish that the males of this fish exhibit parental care by building nests, protecting eggs and as well as the juveniles. Lienart et al. (2013) observed egg clutches frequently inside active nests, and such observation was

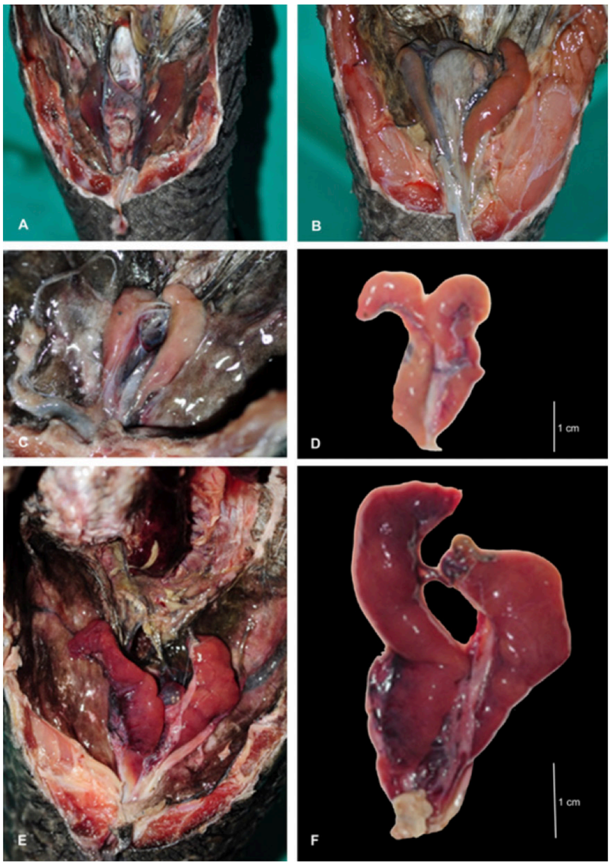

Image 5 A–F. Maturity stages of male *Pterygoplichthys pardalis*: A, B—immature testis | C, D—maturing testis | E, F—mature testis. © Smrithy Raj

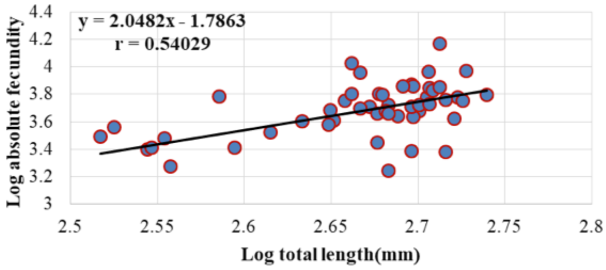

Figure 7. Logarithmic relationship between absolute fecundity and the total length of *P. pardalis* from the present study.

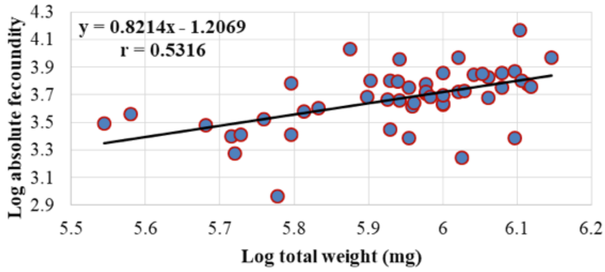

Figure 8. Logarithmic relationship between absolute fecundity and the total weight of *P. pardalis* from the present study.

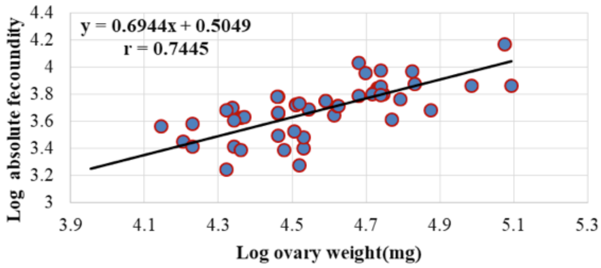

Figure 9. Logarithmic relationship between absolute fecundity and ovary weight of *P. pardalis* from the present study.

not observed in the present study. The present study observes that *Pterygoplichthys pardalis* use the crevices in the granite wall for breeding, which was the first record of breeding behaviour of the species in the Indian water bodies. The benefit of additional natural space also compounded the reproductive behaviour of the fish enhanced its invasive nature in the invaded ecosystem. The sex ratio (M: F) of *P. pardalis* showed an average mean value of 1.04: 1, indicating no bias, showing an equal representation of both sexes in the population.

The current study on the macroscopic and the ova diameter clearly showed different reproductive strategies, showing an extended spawning period with ripe ovaries throughout the year, mostly during March, April, August, and December. Spent ovaries (regressive and recovering ones) and immature stages were noticed in the subsequent months. Such reproductive plasticity was also reported for *P. disjunctivus* by Gibbs et al. (2017) from Volusia Blue Spring for a decade. The peak breeding season reported in *P. pardalis* by Wakida-Kusunoki, & Amador-del Angel (2011) was from June to September. GSI and ova diameter also showed the same results, which coincided with these months. Fish with ripe gonads were obtained throughout our study period except for February, which is consistent with the studies of Rueda-Jasso et al. (2013) for *P. disjunctivus* from Mexico. In the ripe ovaries itself, we came across oocytes with different diameters, consisting of several immature and maturing ova at the same time, which proves the fish as a determinate batch spawner. Similar to other congeners, *P. pardalis* also spawn in batches (Suzuki et al. 2000; Duarte & Araujo 2002). Studies on *P. disjunctivus* from the Philippines by Jumawan & Herrera (2014) also support this view. Batch spawning in *P. pardalis* and its congeners like *Loricariichthys platymetopon*, *Loricariichthys* sp., and *Loricaria* sp. (Suzuki et al. 2000), and *Hypostomus affinis* (Duarte & Araujo 2002) have been reported, where the mature ovaries are seen along with immature and maturing ones with pre-vitellogenic eggs. All Loricariids do not spawn in batches, as *Hypostomus ternetzi*, *Megalancistrus aculeatus*, and *Rhinelepis aspera* are total spawners (Suzuki et al. 2000).

Changes in climatic events could disrupt the reproductive process in fishes (Yoneda & Wright 2005; Pankhurst & Munday 2011). Stable water temperature in tropical rivers is considered best instead of fluctuating waters in subtropical rivers and a shift in temperature could confine the spawning period as well (Samat et al. 2016). According to Humphries et al. (1999), flooding is likely the dominant factor in the breeding behaviour of fish. *Pterygoplichthys* may be adapted to take advantage of flooding by initiating reproduction before or at the time of the flood, which allows fry to feed and grow within inundated floodplain habitats (Kramer 1978; Humphries et al. 1999; Lienart et al. 2013). Based on the results of this work, it may be presumed that *P. pardalis* inhabiting the natural drainages without noticeable temperature variations is a batch spawner and temperature may not be an important limiting factor for spawning of fish that live in a habitat with stable or less fluctuating water temperature. The population assessment of *P. pardalis* from natural drainages of Thiruvananthapuram indicated rapid growth, high-performance index and continuous recruitment, which resulted in their successful invasion (Raj et al. 2020). Maximum reproductive activity of *P. disjunctivus* and *L. multiradiatus* were reported from July to September (Liang et al. 2005; Rueda-Jasso et al. 2013).

The size at first maturity of *P. pardalis* in our study was 23.9 cm standard length (TL 33.5 cm). The minimum size at sexual maturity with highly vitellogenic ova in *P. disjunctivus* was reported as 26.7 cm SL onwards by Jumawan & Herrera (2014) from the Philippines. The case of precocious maturation was also reported in smaller females during the peak spawning time. A report of sexual maturity in *P. disjunctivus* by Gibbs et al. (2008) was of 300 mm SL. The length at which 50% of fish gets mature is 36.56 cm. Gonadal development of male and females in the present study also corroborates with the observations of researchers from other parts of the world.

Absolute fecundity of fish ranges from 923 to 14,777 in the present study, and the fecundity reported for *P. pardalis* from Malaysian waters ranged between 1,297 and 18,791 (Samat et al. 2016). A linear relationship is also exhibited between fecundity and TL, TW, and GW of the fish (Bagenal 1978; Mazzoni & Caramaschi 1995; Duarte & Aroujo 2002). The highest degree of correlation was exhibited in the present study between fecundity and the total length of the fish (r= 0.7445).

The maximum ova diameter obtained in the present study was 3.75 mm, which was in accordance with the ova diameter of 3.3 mm for *P. pardalis* by Samat et al. (2016), 3.8 mm and 3.6 mm obtained for *P. disjunctivus* by Gibbs et al. (2008) and Jumawan & Herrera (2014) respectively. The largest mean ova diameter was 2.327 for ripe ova, as the ova samples contain vitellogenic oocytes of different sizes from the anterior, middle and posterior regions.

A clear case of asymmetry in the ripe ovary with the left lobe larger than the right in the abdominal cavity was observed in the present study. Similar asymmetry in the mature ovary was also observed in *P. disjunctivus* (Gibbs et al. 2008). This asymmetry may be due to the large area occupied by the intestine and the bulk of fat in the abdominal cavity as reported by Rounsefell (1957) in three female salmonids species.

There are no predators for *P. pardalis* in the natural drainages where they have established. This, coupled with the availability of plenty of detritus in the polluted drainages as food, provide them with a competitive edge over indigenous species, better reproductive strategies, including the ability to breed in burrows along the stream banks and crevices in the granite walls, accessory respiratory organs, batch spawning abilities and parental care assisted *P. pardalis* to colonize the system successfully. The population assessment of *P. pardalis* from the same habitat indicated that rapid growth, high growth performance index, and continuous recruitment are the reasons for their successful invasion, and targeting the young individuals would help in controlling the population of the invasive fish (Raj et al. 2020). We recommend more research on the invasion biology of invasive *P. pardalis*, incorporating long-term studies to fully understand the long-term strategies for their establishment and plasticity in the reproductive behaviour.

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