

**Research Article** 

# Distribution in the estuary and salinity tolerance of armored catfish (Loricariidae) in Central Vietnam

Efim D. Pavlov<sup>1</sup>, Tran Duc Dien<sup>2</sup>, Ekaterina V. Ganzha<sup>1</sup>

1 Institute of Ecology and Evolution, Russian Academy of Sciences-IEE RAS, Moscow, Russia

2 Coastal Branch of Joint Vietnam-Russia Tropical Science and Technology Research Center, Nha Trang, Vietnam

Corresponding authors: Efim D. Pavlov (p-a-v@nxt.ru); Tran Duc Dien (mrtran\_cnvb@yahoo.com)



Academic editor: Charles Martin Received: 6 December 2022 Accepted: 23 March 2023 Published: 13 September 2023

**Citation:** Pavlov ED, Dien TD, Ganzha EV (2023) Distribution in the estuary and salinity tolerance of armored catfish (Loricariidae) in Central Vietnam. Aquatic Invasions 18(3): 401–414, https://doi. org/10.3391/ai.2023.18.3.104066

The authors contributed equally to this work.

**Copyright:** © Efim D. Pavlov et al. This is an open access article distributed under terms of the Creative Commons Attribution License (Attribution 4.0 International – CC BY 4.0).

#### Abstract

In the last decade, invasive suckermouth armored catfish Pterygoplichthys spp. spread among many river systems of Vietnam. Extended distribution of armored catfish might be associated with using brackish water in estuaries for fish spread from one river system to another. The first goal of our study was to assess the occurrence of armored catfish in the estuary of the Da Rang River (Phu Yen Province, Vietnam) and their distribution depending on the horizontal salinity gradient (4–25 PSU). Fish were mainly caught by stationary bottom traps in water salinity from 4 PSU to 18 PSU. The second goal of our study was to experimentally evaluate the ability of armored catfish to move and breathe in seawater (33 PSU). Fish moved in horizontal and vertical planes after transfer into seawater during the first 15 minutes. Fish moved around less by the 13<sup>th</sup>-15<sup>th</sup> minutes in seawater. Armored catfish moved around more in seawater than in freshwater. The exposure to seawater for 6 minutes led to deterioration of fish breathing. The results of our field and experimental studies established that armored catfish are found and able to move in brackish waters but avoid high salinity water. These facts provide support for the hypothesis of armored catfish invasion through the estuaries and coastlines.

**Key words:** invasive fish, fish spreading, brackish water, water salinity, locomotor activity, breathing

#### Introduction

The invasive suckermouth armored catfish genus *Pterygoplichthys* (Siluriformes: Loricariidae) is widely spread and successfully occupies inland waters of the central Indo-Pacific region (Orfinger and Goodding 2018). In the waterbodies of South Vietnam, this invasive genus was registered in 2003–2004 (Welcomme and Vidthayanom 2003; Serov 2004), and by 2010 it spread to Central Vietnam (Zworykin and Budaev 2013) and Northern Vietnam (Levin et al. 2008). Currently, armored catfish are found in the basins of many big rivers and some reservoirs in South and Central Vietnam (Stolbunov and Dien 2019; Stolbunov et al. 2020).

The hypothesis of armored catfish spreading through the estuaries to the rivers was discussed in Southeastern Mexico (Capps et al. 2011), India (Kumar et al.



2018), and Philippines (Brion et al. 2013). According to the estuary hypothesis, the spread of the genus could occur through the common estuary of several rivers or streams. Also, dispersal routes of fish could run along the coastlines between adjacent coastal rivers. However, occurrence of armored catfish in the estuary zones has not been frequently reported (Stevens et al. 2006; Capps et al. 2011), and specifically have not been previously reported in the estuaries of Vietnam (Lai et al. 2020).

Salinity tolerance is one of several important physiological features that determine invasion success and the pattern of dispersal of introduced aquatic organisms between freshwater bodies through brackish waters (Cognetti and Maltagliati 2000; Schofield et al. 2009; Capps et al. 2011). Loricariidae is considered a strictly freshwater family of fish in their native range throughout the Neotropics (Myers 1949). Nonetheless, Loftin (1965) found that *Hypostomus aspidolepis* could survive immediate immersion in 30 PSU and 40 PSU if acclimated. Fish genus Pterygo*plichthys* tolerates water salinity up to 15–16 PSU (Capps et al. 2011), while most freshwater fish species have salinity tolerance of no more than 5.0-8.0 PSU (Khlebovich 1974; Karpevich 1976). Our previous study showed that armored catfish Pterygoplichthys spp. from the Dinh River (Central Vietnam) tolerated water salinity up to 15 PSU for two days (Dien et al. 2022). Perhaps, armored catfish occur in natural brackish waters due to their high salinity tolerance. This could facilitate invasion of these fish into new rivers through the estuary. The hypothesis of estuary spreading of armored catfish explains their rapid (<10 years) invasion into the waterbodies of Vietnam.

The detection of armored catfish in estuaries and observation of their movements could support the estuary hypothesis. For subsequent spreading, fish need to move through the brackish waters with different salinity. To increase chances of successfully spreading and decrease mortality risk, fish need to be able to breathe in high salinity water.

The study aimed to estimate the distribution of armored catfish genus *Pterygoplichthys* in the estuary of the Da Rang River and experimentally assess the effects of high salinity water on fish's movements and gill respiration.

# Materials and methods

The study was conducted from February to March 2022. The object of the study was the suckermouth armored catfish genus *Pterygoplichthys* (Siluriformes: Loricariidae). Two species of armored catfish identified in South Vietnam: *P. disjunctivus* and *P. pardalis* (Zworykin and Budaev 2013; Stolbunov and Dien 2019; Stolbunov et al. 2020). For species identification, we used systematic keys (Armbruster and Page 2006) based on the number of dorsal-fin rays and on body color patterns. Ninety percent of the fish in the study were identified as *P. disjunctivus*, and 10% of fish had traits of *P. pardalis*. It is possible that these fish were hybrids (Godwin et al. 2016) however we will call them *P. pardalis* and *P. disjunctivus* for simplicity.

#### The field study

The Da Rang River was chosen for the field study. This river is one of the largest river systems in Central Vietnam; its length is 374 km, and the basin area is 13900 km<sup>2</sup>. The river forms a wide and extended estuary in Phu Yen Province and flows into the Eastern Sea within Tuy Hoa City. The annual tide in the estuary varies from 0.5 m to 1.7 m. The Da Rang River discharge during the flood reaches 2100 m<sup>3</sup>/s, and the average annual river discharge is 275 m<sup>3</sup>/s (Hiep et al. 2022).



We caught armored catfish in the estuary of the Da Rang River to study their distribution in the brackish water. The capture was conducted within 3.2–4.2 km of the Eastern Sea coastline (13°03'53"N, 109°18'33"E). Within this zone, four sampling localities were selected with different levels of water salinity near the sea (Figure 1). The maximum depth in this area was 3.0–5.5 m. The water was sampled in each sampling locality from 0.2–0.5 m near the bottom with alpha water sampler 3-1120-G42 (Wildco, USA) during the maximum tide (1.3–1.4 m). We measured water salinity by optical refractometer RHS-10ATC (Kelilong Electron, China).



**Figure 1.** Scheme of sampling localities (black circles) in the estuary of the Da Rang River and the water salinity near the bottom. No 1 – nearest place to the Eastern Sea (3.2 km), No 2 – first median place (3.6 km), No 3 – second median place (3.9 km), No 4 – the distant place from the Eastern Sea (4.2 km) (QGIS 3.26.2). Arrow ( $\rightarrow$ ) indicates the water flow direction.

To estimate the water salinity stratification, the water salinity was measured from the bottom to the water surface on the sampling locality No 1 closest (3.2 km) to the sea (13°03'55"N, 109°18'40"E) (Figure 2). A sharp difference in water salinity from 27–13 PSU to 4–5 PSU was registered at the depth of 3–4 m. Such spatial pattern of seawater and freshwater characterized the estuary as highly stratified (Geyer 2010; Valle-Levinson 2010). The water salinity was irregular, i.e. on the left shore it was higher than on the right one. The water with a salinity of 18–25 PSU had a higher temperature (24.3 °C) than water with a salinity <5 PSU (23.1 °C). We used the test kits of Sera Aqua-Test Box (Sera North America, Inc.) for checking some water parameters on the surface and near the bottom in the sampling locality No 1 (see Suppl. material 1). Bottom saline water compared with surface water had higher pH and iron values and minimal concentration of nitrate and ammonium/ammonia ratio.

Given that armored catfish is benthivore, we placed sectional net traps on the bottom (Covain and Fisch-Muller 2007; Nico 2010). The net mesh size was





**Figure 2.** Scheme of water salinity (PSU) stratification in the estuary of the Da Rang River 3.2 km from the sea. The water salinity higher than 5 PSU was shaded. Counting points (hollow circle).

10 mm. Traps had a length of 9 m and consisted of rectangular metal frames (0.3  $\times$  0.2 m) located 0.3 m from each other. The left and right sides of the traps had 15 conical-shaped holes with an opening diameter of 100 mm. The traps were connected in a single line and placed at the bottom across the river in four sampling localities (Figure 1). Fifty traps were mounted on sampling locality No 1 and forty traps were mounted on the other three places. We used higher numbers of traps in the first locality because local fishermen considered armored catfish there absent. The traps were set up in the evening (17:00–18:00, GMT+7) and placed in each sample locality for entire night. The traps were checked the next morning (9:00-10:00, GMT+7). The fish were caught during a dark period because armored catfish are nocturnal (MacCormack et al. 2003; Nico 2010). Daily periods of fish catching in different sampling localities in the Da Rang River were on 23-24 February (No 1), 25-26 February (No 2), 26-27 February (No 3) and 27-28 February (No 4). During fish catching, the tides' height varied from 0.4 m to 1.5 m; there were no heavy rains; the wind direction was from the sea and did not change. Fish standard body length (SL, cm) and body weight (g) were measured.

#### The experimental study

The study was conducted at the Coastal Branch of Joint Vietnam-Russia Tropical Science and Technology Research Center (Nha Trang, Vietnam) in March 2022. For experimental work, freshwater armored catfish were caught in the Dinh River in Khanh Hoa Province (12°29'45"N, 109°07'41"E). Fish from the Dinh River was used because we did not have enough fish for experiments from the Da Rang River. The Dinh River length is 50 km, and the basin area is 985 km<sup>2</sup>. The captured fish were transferred to the laboratory in tanks.

The study was performed on 60 fish with a standard body length  $14 \pm 0.5$  (9–24) cm and a body weight  $45 \pm 5.5$  (9–155) g (here and after before the brackets are the mean value and its error; in the brackets are min and max). In the laboratory, fish were separated equally between two 100L maintenance tanks with a water volume of 35L and water temperature 25–26 °C. The tap freshwater (0 PSU)

in a laboratory was conditioned by settling and aeration for 2 weeks in two 2000L basins. The water in the maintenance tanks with fish was aerated and changed once per day. The level of dissolved oxygen in the water was 7.0–7.2 mg/L (Pro Dissolved oxygen meter MW600, Milwaukee, USA). Illumination in the maintenance tanks was natural (through the laboratory windows) and varied during the twenty-four hours from 0.1 Lx to 100 Lx (Lux meter Lutron LX-1102). Fish were fed with Pro's choice tablets – Bottom feeders (Fwusow Industry, Taiwan) for demersal fish twice per day (at 10:00 and 16:00, GMT+7). The fish started feeding on the tablets three days after the transfer. The cumulative fish mortality did not exceed 5% during the first week of maintenance.

Two series of experiments were conducted. In the first series of experiments, we estimated armored catfish locomotor activity and survival rate under exposure with seawater (33 PSU). In the second series of experiments, we estimated locomotor activity and gill respiration under exposure with freshwater (0 PSU) and seawater (33 PSU). Seawater was used from Nha Trang Bay (12°13'03"N, 109°12'47"E) and was conditioned by settling and aeration for 2 weeks. The level of dissolved oxygen in seawater was 7.0–7.2 mg/L. In both series of experiments, the test chambers were two 15L glass aquaria. We used LED backlight placed outside the aquarium (~100 Lx).

The first series of experiments were carried out on 26 fish with a standard body length  $12 \pm 0.4$  (9–18) cm and a body weight  $24 \pm 2.7$  (9–67) g. Each aquarium was filled with 6L of seawater, and the water level was 0.1 m. Two remotely controlled video cameras SjCam A10 (China) were positioned under each aquaria at a distance of 0.2 m from their bottom. Camera recording was controlled by iPad 2018 (Apple Co., USA). At the beginning of the trial, one fish at a time was randomly transferred into each aquarium from the maintenance tanks (see Suppl. material 2). The duration of each individual trial with video recording was 15 minutes. In total, 26 trials with 6.5 hours of video were assessed. The water in the test chamber was changed after each trial to remove fish metabolites and to control salinity value. After each trial, fish were transferred into a recovery tank with freshwater and aeration. Survival rate after the seawater exposure was measured during twenty-four hours. Time duration of fish locomotor activity during each minute of the trial was estimated on video recordings. Movements in horizontal and vertical (movements towards water surface) direction were scored when a fish moved for over 2 seconds.

The second series of experiments were performed on 26 fish with a standard body length  $17 \pm 0.7$  (12–24) cm and a body weight  $62 \pm 8.7$  (18–155) g. For the second series, one aquarium was filled with 6L of freshwater, and the second was filled with 6L of seawater. For video records, we used two GoPro Hero 8 (GoPro Inc., USA) cameras positioned under each aquarium. One fish at a time from the maintenance tanks was randomly transferred to the aquaria with freshwater and one fish was transferred to seawater. Fish assessed under freshwater and seawater exposure did not differ in body length and weight (Student's *t*-test: p = 0.62, p = 0.57, respectively). Water in the test aquaria was changed after each trial, and the aquaria with freshwater and seawater were switched. The duration of the trial was six minutes; timing was based on the first series of experimental results to prevent fish mortality in seawater tests. In total, 13 fish were used in freshwater trials and 13 fish were used in seawater trials; 2.5 hours of video were assessed. After trials, fish were transferred into a recovery tank with freshwater and aeration. We used video records to register the duration of fish locomotor activity and their rises to the water surface in the tests. The gill ventilation rate  $(f_{,,})$  (MacCormack et al. 2003) was calculated by counting the opening and closing of the gill slits for each fish at 30 s intervals. One opening and closing of the gill slits was considered as a single completed breath (one breath per minute, 1 breath/min). In all tests, each fish was used once.

Statistical data analysis (Minitab 18.1) was conducted with Student's t-test and Spearman rank correlation coefficient ( $r_s$ ). The Shapiro-Wilk test was used to assess normality distribution of samples. The individual locomotor activity during each minute of the test was assessed by normalized values using the formula:

$$f_{a} = t_{i}^{*} 100/7$$

Where:  $f_a$  – frequency (%) of locomotor activity of fish during each minute of the test,  $t_i$  – duration of fish moving during  $i^{ab}$  minute of the test, T – total time of fish moving during the test.

The distribution of the locomotor activity of fish depending on the time of the test was compared with the  $\chi^2$  criterion.

# Results

## Armored catfish distribution in the estuary

The number of fish caught in traps was different between sampling localities and increased with a distance away from the sea (Table 1). Only one live fish was caught in the nearest sampling locality to the sea (No 1). The fish escaped when we tried to get it out of the trap, so we were unable to measure its body length and weight. We registered live and dead captured fish in approximately equal proportions in the median sampling localities (No 2 and No 3). Two fish were alive and two fish were dead in locality No 2. Four fish were alive and five fish were dead in locality No 3. All ten fish were alive at a distant place from the sea (No 4). The body length and weight of all captured armored catfish varied. But the fish from different locations (No 2, No 3, No 4) did not significantly differ in body length and weight (Student's t-test: p > 0.37). The living and dead fish also did not significantly differ in body length and weight (Student's t-test: p = 0.48 and p = 0.52, respectively).

Sampling localities	Maximum registered water salinity, PSU	SL, cm	W, g	Number of alive/dead fish
No 1	25	_	_	1/-
No 2	18	18 ± 2.0 (16–24)	90 ± 31.2 (47–182)	2/2
No 3	14	16 ± 1.8 (13–30)	71 ± 30.1 (28–310)	4/5
No 4	4	18 ± 1.8 (11–28)	109 ± 32.9 (23–300)	10/-

Table 1. Standard body length and body weight of armored catfish *Pterygoplichthys* spp. from the Da Rang River estuary.

Note. Before the brackets are the mean value and its error; in the brackets are min and max.

Along with armored catfish were also captured: Malabar glassy perchlet *Ambassis dussumieri*, *Ambassis* sp., silver barb *Barbonymus gonionotus*, *Glossogobius* spp., marble goby *Oxyeleotris marmorata*, Nile tilapia *Oreochromis niloticus*, long whiskers catfish *Mystus gulio*, and bartail flathead *Platycephalus indicus*.

#### Fish locomotor activity and survival rate

Armored catfish lay down motionless at the bottom of the maintenance tank with freshwater during daylight. Less frequently, fish were located vertically pressed against the side walls.

In the first series of the experiment, all tested fish were transferred to seawater. After transfer, fish usually moved no longer than first minute of the trial;



then they lay at the bottom. During the test, fish alternated periods of locomotor activity and inactivity. On average, 20% of the test time fish moved along the perimeter of the aquaria: both near the bottom and rising to the surface water. Fish movements were often abrupt and with brief stops (2–3 sec). Fish locomotor activity in seawater significantly decreased by  $13^{th}-15^{th}$  minutes of the trial (Student's *t*-test: 13–15 minutes versus 1–3 minutes, p = 0.009; 13–15 minutes versus 7–9 minutes, p = 0.005) (Figure 3). Thus, by the 15<sup>th</sup> minute of the test, more than half (>50%) of the fish (14 fish) were motionless for more than one minute. Six of these fish did not move for at least three minutes. After transferring the tested fish back to the freshwater, 50% of them died within 24 hours. Fish that survived had a standard body length of  $12 \pm 0.8$  (9–18) cm and a body weight of  $26 \pm 5.0$  (9–67) g. The dead fish's body length and weight were  $12 \pm 0.4$  (9–14) cm and  $22 \pm 2.0$  (9–33) g. The surviving fish did not differ in body length and weight compared to the dead fish, Student's *t*-test: p = 0.51 and p = 0.44, respectively.



**Figure 3.** Locomotor activity (% of the interval timing) of armored catfish *Pterygoplichthys* spp. under seawater exposure for 15 min (first series of the experiment). Different letters (a, b) indicate significant differences between intervals timing (Student's *t*-test: p < 0.05). Standard error (|).

In the second series of the experiment, 77% of armored catfish after transfer into freshwater aquaria did not move during the test (for six minutes). Twenty-three percent of fish moved on average for  $3.8 \pm 0.69$  (3.0-5.2) minutes during the trial. Fish movement in the freshwater was smooth. Under seawater exposure, 69% of fish moved on average for  $2.0 \pm 0.33$  (1.1-4.1) minutes during the trial. The fish movements were abrupt with frequent changes in speed and direction. The timing of the locomotor activity of fish in seawater differed at the first 3 minutes and the last 3 minutes of the trial ( $\chi^2$  criterion: p = 0.01, n = 9). In the seawater in contrast to freshwater, the posterior end of the fish was raised contrary to the anterior end. Sometimes air bubbles were released from the oral cavity and the gill slits. Both processes were observed in 62% of fish at  $4.1 \pm 0.42$  (2.3-5.4) minutes of the trial and were often accompanied by fish movement towards the water surface to capture air. In seawater, 69% of fish had spasms from the fourth minute of the trial. We registered on average  $11 \pm 2.9$  (1-28) body spasms per fish.



# Gill respiration features of fish

After the fish were transferred into freshwater, the gill ventilation rate  $(f_v)$  remained stable: 206 ± 3.4 (108–294) breaths/min (Figure 4). This parameter correlated with fish body length and weight:  $r_s = -0.71$  with p = 0.006 and  $r_s = -0.69$  with p = 0.009, respectively. During the first minute after the fish were transferred into seawater, the gill ventilation rate was similar to fish transferred to freshwater (Student's *t*-test: p > 0.05). Then the gill ventilation rate of the fish in seawater began to decline and by the 6<sup>th</sup> minute of the test and reached a minimal value: 15 ± 4.6 (0–90) breaths/min. There was no correlation between the gill ventilation rate and the body length and weight of the fish in seawater (p > 0.05). In the last three minutes of the trial, four fish did not have gill slit movements for more than a minute. A day after the tests were completed, one fish died.



**Figure 4.** Dynamics of the gill ventilation rate ( $f_v$ ) of armored catfish *Pterygoplichthys* spp. under freshwater (hollow square, n = 13) and seawater (black square, n = 13) exposure during six minutes of testing. The characters indicate: \* – significant differences between the data of three initial timing intervals (the first minute of the test) according to Student's *t*-test: \* – p < 0.001, \*\* – p < 0.01, \*\*\* – p < 0.05. Standard error (|).

#### Discussion

Our results confirmed the occurrence of armored catfish in the brackish waters of the Da Rang River estuary in Central Vietnam. The fact that fish were captured in stationary traps is the evidence of their active movements in the estuary. However, this result does not reflect the distance of movements or movement directions of armored catfish in estuaries. To date, armored catfish were found only in brackish waters (up to 8 PSU) in Southeastern Mexico Rivers (Capps et al. 2011). In the brackish water of the Mekong Delta (South Vietnam), armored catfish (*P. disjunctivus* × *pardalis*) were not previously detected (Lai et al. 2020). Lai et al. (2020) reported collecting fish using bottom trawl net (4 m wide, 7 m long, and 2 cm mesh size) in the morning. We hypothesize that the absence of



captured armored catfish in the brackish waters of the Mekong River was due to different fishing techniques or a small population of fish. The armored catfish population in the estuary could depend on the appropriate food availability. River mouths are highly productive environments. Estuaries could noticeably differ among themselves in food supply and many physical, chemical, and biological factors (Mateus et al. 2008) which could affect armored catfish numbers in brackish water.

In the Da Rang River estuary, we often caught armored catfish in places located 3.5 km or more from the sea with water salinity up to 18 PSU. That was consistent with our findings that armored catfish could survive in water salinity up to 12.5–15.0 PSU for two days (Dien et al. 2022). According to Capps et al. (2011), some armored catfish survived in water salinity up to 16.0 PSU for a few hours. Survival in high water salinity could explain by long-term fish adaptation to environmental factors. For example, exposure to high water salinity over generations may lead to adaptations that increase the salinity tolerance of freshwater macroinvertebrates (Kay et al. 2001; Kefford et al. 2004). It is currently unknown whether armored catfish in the Da Rang River has similar adaptation. However, we hypothesize that armored catfish could avoid the high salinity water (>25 PSU) because only one fish was caught in the nearest place to the sea (sample locality No 1).

Water salinity is one of the dynamic factors in estuaries. The water salinity depends on the magnitude and phase of the sea tides, the direction and strength of the wind, as well as the intensity and speed of the water flow in the river (Hartog 1974; Mateus et al. 2008; Geyer 2010). During the period of our study, in the estuary of the Da Rang River the seawater was mostly located in the bottom layer and poorly mixed with freshwater. However, the water salinity in the bottom layer could change noticeably, depending on the diurnal regimes in the estuary. We hypothesize, that diurnal variation of water salinity in the bottom layer relates to fish survival rate in the stationary traps. Thus, fish that entered the traps in sample localities No 2 and No 3 earlier had higher mortality risk. We propose that fish were getting inside the traps during the lower water salinity, but they could not leave the traps during the increased salinity. No dead captured fish were observed at the distant place from the sea (No 4), probably due to low water salinity (4 PSU).

Fish moved for an average of 20% of 15 minutes of the trial (first series of the experiment). Generally, for the first 12 minutes their locomotor activity did not decrease. This feature could allow armored catfish to leave the increased salinity area in nature. But seawater treatment for 15 minutes negatively affected fish survival in the future: 50% of the fish died in the next twenty-four hours.

Most fish (77%), transferred from a freshwater maintenance tank into freshwater aquaria did not move in the aquaria for six minutes (second experiment series). Their gill ventilation rate did not change noticeably during the test period. Most fish (69%), transferred from the freshwater maintenance tank into the seawater aquaria, moved during the trial both horizontally and to the water surface. Thus, the seawater treatment increased the locomotor activity of armored catfish and could stimulate them to leave the area with the high salinity water. Based on this, we conclude that the armored catfish genus *Pterygoplichthys* can avoid increased salinity water by horizontal movements and rising to the water surface. Hypothetically, armored catfish movements towards the water surface positively affect their survival in the strong stratification of water salinity in the Da Rang River estuary. More studies needed to prove this.

Significant deceleration or cessation of gill respiration was observed during six minutes' exposure to seawater. Many fish retained the ability to move despite the breathing deterioration. Sixty-two percent of the fish moved to seawater surface to capture air. According to Armbruster (1998), loricariid catfishes have evolved several

adaptive modifications of the digestive tract (enlarged stomach and thinning its wall, U-shaped or ringlike diverticulum), that appear to function as accessory respiratory organs or hydrostatic organs. We propose that the aquatic surface respiration (da Cruz et al. 2013; Gibbs and Groff 2014; Gibbs et al. 2021) of tested armored catfish compensate for not being able to breathe in seawater. Seawater could affect the gas exchange processes in the gills epithelium, resulting in a lack of oxygen in the fish's blood despite sufficient water saturation with oxygen. For example, with hypoxia in *P. pardalis* (*=Liposarcus pardalis*) 70% of the oxygen in the arterial blood was formed by aquatic surface respiration (Val 1995). In addition, armored catfish had exceptional heart resistance to anoxia (Bailey et al. 1999; MacCormack et al. 2003). Facultative breathing in armored catfish is consistent with their ability to move in seawater. Armbruster (1998) suggested that some loricariid catfishes could use respiratory stomach for buoyancy control, which seems odd because they are generally considered to be benthivore. This feature possibly could help armored catfish to spread through surface water with low salinity in the estuary and near the coast.

Experimental results indicated that the locomotor activity of armored catfish in seawater varied between individual fish in duration and intensity. The success of the survival of armored catfish in water with high salinity is probably determined by individual and species characteristics of navigating in the salinity gradient, which was noted by other researchers (Capps et al. 2011), but not studied in detail.

Fish survival rate under seawater treatment for six minutes exceeded 90%, which means that armored catfish could tolerate water with high salinity for a limited period of time. This limited period of time might be enough to change direction of movement, leave the area with high water salinity and spread through the estuary to other influent rivers. But it might not be enough to move to another river system along the coastline. We propose that some environmental factors could facilitate armored catfish to spread along the coastline: the mouths of rivers are located close to each other where water salinity is lower (due to mixing of freshwater and seawater), fish move along a freshwater plume at the surface which is common in estuarine systems dominated by the river. For example, the water sulface salinity varied noticeably from 0.5 PSU to 27.0 PSU in Nha Trang Bay located between two estuaries of the adjacent rivers (Cai River and Be River) (Nezdolii et al. 2014). The success of armored catfish invasion through the coastline area could also be determined by the ability of fish perceive water salinity changes. That will be the subject of our future research.

# Conclusion

We discovered the armored catfish in the brackish waters of the Da Rang River. Our experiments demonstrated that the fish genus *Pterygoplichthys* could tolerate seawater for some period of time with preservation of locomotor activity, despite the deterioration of gill respiration. Our results show that armored catfish have physiological features that allow them to spread through estuary and coastline. Armored catfish could stay and move in the estuary. Fish encounter water with different salinity and could tolerate sudden water salinity increases (33 PSU) for a short period of time. Increased movement in seawater might allow them to leave the area with high salinity water.

We propose that the main obstacle to armored catfish invasion is water salinity above 15–18 PSU. However, a combination of favorable factors increases the chances of fish spreading not only through the estuaries but also through the coastlines. These factors are closely located adjacent river estuaries, plenty of freshwater in the estuary and the coastline area, a freshwater plume at the surface.



A wide range of physiological adaptations of armored catfish allows them not only to successfully spread to new ecosystems but also compete with aboriginal fish for resources. Risks of the potential impact of armored catfish invasion need to be considered when managing the ecosystem.

# Funding

Field work and experiments were supported by Coastal Branch of Joint Vietnam -Russia Tropical Science and Technology Research Center (Ecolan 3.2 "Taxonomic diversity, ecology and behavior of freshwater hydrobionts"); video data analysis was supported by Russian Science Foundation No. 19-14-00015.

## Author's contributions

All authors contributed to the study conception and design. Material preparation and data collection in field were performed by EP and DTD. EG and EP performed experimental work and analyzed data. The first draft of the manuscript was written by EP and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

## **Ethics and permits**

All experimental procedures with fish were carried out according to the guidelines and following the laws and ethics of Socialist Republic of Vietnam and approved by the ethics committee of the Institute of Ecology and Evolution, Russian Academy of Sciences.

#### Acknowledgments

Authors would like to thank the administration and staff of Coastal Branch of Joint Vietnam - Russia Tropical Science and Technology Research Center for their help in organizing field sample collection and for kindly allowing us to use their laboratories and experimental facilities. We are grateful to Prof. A.O. Kasumyan, Prof. J.W. Armbruster, and anonymous reviewers for valuable comments on the manuscript's text.

#### References

- Armbruster JW (1998) Modifications of the digestive tract for holding air in loricariid and scoloplacid catfishes. Copeia 1998: 663–675. https://doi.org/10.2307/1447796
- Armbruster JW, Page LM (2006) Redescription of *Pterygoplichthys punctatus* and description of a new species of *Pterygoplichthys* (Siluriformes: Loricariidae). Neotropical Ichthyology 4: 401–409. https://doi.org/10.1590/S1679-62252006000400003
- Bailey J, Val AL, Almeida-Val VMF, Driedzic WR (1999) Anoxic cardiac performance in Amazonian and north-temperate zone teleosts. Canadian Journal of Zoology 77: 683–689. https://doi. org/10.1139/z99-019
- Brion MA, Guillermo Jr JG, Uy C, Chavez J, Carandang IV JS (2013) Salinity tolerance of introduced South American sailfin catfishes (Loricariidae: Pterygoplichtys Gill 1858). Philippine Journal of Science 142: 13–19.
- Capps KA, Nico LG, Mendoza-Carranza M, Arévalo-Frías W, Ropicki AJ, Heilpern SA, Rodiles-Hernández R (2011) Salinity tolerance of non-native suckermouth armoured catfish (Loricariidae: *Pterygoplichthys*) in south-eastern Mexico: implications for invasion and dispersal. Aquatic Conservation: Marine and Freshwater Ecosystems 21: 528–540. https://doi.org/10.1002/aqc.1210



- Cognetti G, Maltagliati F (2000) Biodiversity and adaptive mechanisms in brackish water fauna. Marine Pollution Bulletin 40: 7–14. https://doi.org/10.1016/S0025-326X(99)00173-3
- Covain R, Fisch-Muller S (2007) The genera of the Neotropical armored catfish subfamily Loricariinae (Siluriformes: Loricariidae): a practical key and synopsis. Zootaxa 1462: 1–40. https://doi. org/10.11646/zootaxa.1462.1.1
- da Cruz AL, da Silva HB, Lundstedt LM, Schwantes AR, Moraes G, Klein W, Fernandes MN (2013) Air-breathing behavior and physiological responses to hypoxia and air exposure in the air-breathing loricariid fish, *Pterygoplichthys anisitsi*. Fish Physiology and Biochemistry 39: 243–256. https://doi.org/10.1007/s10695-012-9695-0
- Dien TD, Ganzha EV, Pavlov ED, Samoilov KYu, Pavlov DS (2022) Survival and level of thyroid hormones and ions in armored catfish (Loricariidae) during increasing water salinity. Journal of Ichthyology 6: 1201–1207. https://doi.org/10.1134/S0032945222060054
- Geyer W (2010) Estuarine salinity structure and circulation. In: Valle-Levinson A (Ed.) Contemporary Issues in Estuarine Physics. Cambridge University Press, Cambridge, 12–26. https://doi.org/10.1017/CBO9780511676567.003
- Gibbs MA, Groff BW (2014) Patterns of aerial respiration by *Pterygoplichthys disjunctivus* (Loricariidae) in Volusia Blue Spring, Florida. Florida Academy of Sciences 77: 53–68. https://www.jstor. org/stable/24321983
- Gibbs MA, Thornton A, Pasko S, Crater A (2021) Patterns of air-breathing behavior in juvenile armored catfish, *Pterygoplichthys* sp. (Gill 1858). Environmental Biology of Fishes 104: 171–180. https://doi.org/10.1007/s10641-021-01068-w
- Godwin JC, Werneke DC, Steen DA, Armbruster JW (2016) Two significant records of exotic neotropical freshwater fish observed in southern Alabama. Southeastern Naturalist 15: 57–60. https://doi.org/10.1656/058.015.0401
- Hartog DC (1974) Brackish-water classification, its development and problems. Aquatic Ecology 8: 15–28. https://doi.org/10.1007/BF02254902
- Hiep NT, Tanaka H, Nguyen XT, Nguyen TV (2022) Shoreline change and impacts of coastal protection structures on Da Rang River mouth and adjacent coast, south central of Vietnam. Journal of Natural Disaster Science 58: 105–110.
- Karpevich AF (1976) Guidelines for the study of the endurance of fish and invertebrates with changes in the salinity of the environment and the methodology for its determination. VNIRO, Moscow, 55 pp. [In Russian]
- Kay WR, Halse SA, Scanlon MD, Smith MJ (2001) Distribution and environmental tolerances of aquatic macroinvertebrate families in the agricultural zone of southwestern Australia. Journal of the North American Benthological Society 20: 182–199. https://doi.org/10.2307/1468314
- Kefford BJ, Papas PJ, Metzeling L, Nugegoda D (2004) Do laboratory salinity tolerances of freshwater animals correspond with their field salinity? Environmental Pollution 129: 355–362. https:// doi.org/10.1016/j.envpol.2003.12.005

Khlebovich VV (1974) Critical salinity of biological processes. Nauka, Leningrad, 117 pp. [In Russian]

- Kumar AB, Schofield PJ, Raj S, Satheesh S (2018) Salinity tolerance of non-native suckermouth armoured catfish (Loricariidae: *Pterygoplichthys* sp.) from Kerala, India. Management of Biological Invasions 9: 49–57. https://doi.org/10.3391/mbi.2018.9.1.05
- Lai QT, Orfinger AB, Tran TT, Le NK (2020) Distribution of Suckermouth Armoured Catfishes (Siluriformes, Loricariidae) Across the Salinity Gradient of the Mekong Delta, Vietnam. Asian Fisheries Society 33: 300–306. https://doi.org/10.33997/j.afs.2020.33.3.011
- Levin BA, Phuong PH, Pavlov DS (2008) Discovery of the Amazon sailfin catfish *Pterygoplichthys pardalis* (Castelnau, 1855) (Teleostei: Loricariidae) in Vietnam. Journal of Applied Ichthyology 24: 715–717. https://doi.org/10.1111/j.1439-0426.2008.01185.x
- Loftin HG (1965) The geographical distribution of freshwater fishes in Panama. Unpubl. Ph.D. diss., Florida State University, 264 pp.
- MacCormack TJ, McKinley RS, Roubach R, Almeida-Val VMF, Val AL, Driedzic WR (2003) Changes in ventilation, metabolism, and behaviour, but not bradycardia, contribute to hypoxia



survival in two species of Amazonian armoured catfish. Canadian Journal of Zoology 81: 272–280. https://doi.org/10.1139/z03-003

- Mateus M, Mateus S, Baretta JW (2008) Basic concepts of estuarine ecology. In: Salomons W, Turner RK, Lacerda LD, Ramachandran S (Eds) Perspectives on Integrated Coastal Zone Management in South America. IST, 3–134.
- Myers GS (1949) Salt-tolerance of fresh-water fish groups in relation to zoogeographical problems. Bijdr Dierk 28: 315–322. https://doi.org/10.1163/26660644-02801038
- Nezdolii VK, Pavlov DS, Ngo Chi Then, Nguen Dui Tuan, Nguen Quoc Tanh (2014) Downstream migration of juvenile fish in the Cai River. In: Pavlov D, Zworykin D (Eds) Ecology of Inland Waters of Vietnam. KMK, Moscow, 298–319. [In Russian]
- Nico LG (2010) Nocturnal and diurnal activity of armored suckermouth catfish (Loricariidae: *Ptery-goplichthys*) associated with wintering Florida manatees (*Trichechus manatus latirostris*). Neotropical Ichthyology 8: 893–898. https://doi.org/10.1590/S1679-62252010005000014
- Orfinger AB, Goodding DD (2018) The global invasion of the suckermouth armored catfish genus *Pterygoplichthys* (Siluriformes: Loricariidae): annotated list of species, distributional summary, and assessment of impacts. Zoological Studies 57: e7. https://doi.org/10.6620/ZS.2018.57-07
- Schofield PJ, Loftus WF, Fontaine JA (2009) Salinity effects on behavioural response to hypoxia in the non-native Mayan cichlid *Cichlasoma urophthalmus* from Florida Everglades wetlands. Journal of Fish Biology 74: 1245–1258. https://doi.org/10.1111/j.1095-8649.2009.02192.x

Serov DV (2004) Harnischwelse in Südostasien. Die Aquarium- und Terrariumzeitschrift 2: 18–19.

- Stevens PW, Blewett DA, Casey JP (2006) Short-term effects of a low dissolved oxygen event on estuarine fish assemblages following the passage of Hurricane Charley. Estuaries and Coasts 29: 997–1003. https://doi.org/10.1007/BF02798661
- Stolbunov IA, Dien TD (2019) Mass alien fish species in the fish fauna of inland waters in Central Vietnam. Inland Water Biology 12: 477–480. https://doi.org/10.1134/S1995082919040163
- Stolbunov IA, Dien TD, Armbruster JW (2020) Suckermouth-armored catfish (Siluriformes: Loricariidae) of Central and Southern Vietnam. Inland Water Biology 13: 626–639. https://doi. org/10.1134/S1995082920040100
- Val AL (1995) Oxygen transfer in fish: morphological and molecular adjustments. Brazilian Journal of Medical and Biological Research 28: 1119–1127.
- Valle-Levinson A (2010) Contemporary Issues in Estuarine Physics. Cambridge University Press, Cambridge. https://doi.org/10.1017/CBO9780511676567
- Welcomme RL, Vidthayanom C (2003) The impacts of introductions and stocking of exotic species in the Mekong Basin and policies for their control. Phnom Penh, Cambodia, MRC Tech Paper 9. https://doi.org/10.52107/mrc.akbo77
- Zworykin DD, Budaev SV (2013) Non-indigenous armoured catfish in Vietnam: invasion and systematics. Ichthyological Research 60: 327–333. https://doi.org/10.1007/s10228-013-0356-9

# **Supplementary material 1**

Chemical parameters of water on the surface and near the bottom (five meters deep) in the Da Rang River estuary

Authors: Efim D. Pavlov, Tran Duc Dien, Ekaterina V. Ganzha Data type: table (docx. file)

Copyright notice: This dataset is made available under the Open Database License (http://opendatacommons.org/licenses/odbl/1.0/). The Open Database License (ODbL) is a license agreement intended to allow users to freely share, modify, and use this Dataset while maintaining this same freedom for others, provided that the original source and author(s) are credited.

Link: https://doi.org/10.3391/ai.2023.18.3.104066.suppl1



## **Supplementary material 2**

#### Scheme of test chambers

Authors: Efim D. Pavlov, Tran Duc Dien, Ekaterina V. Ganzha Data type: figure (docx. file)

Copyright notice: This dataset is made available under the Open Database License (http://opendatacommons.org/licenses/odbl/1.0/). The Open Database License (ODbL) is a license agreement intended to allow users to freely share, modify, and use this Dataset while maintaining this same freedom for others, provided that the original source and author(s) are credited.

Link: https://doi.org/10.3391/ai.2023.18.3.104066.suppl2