

Early Ontogeny of the Climbing Perch *Anabas testudineus* (Anabantidae) in Relation to the Buoyancy Dynamics

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Abstract—This article describes early development of the climbing perch *Anabas testudineus* in relation to its buoyancy dynamics. Main patterns of the ontogeny during the first 140 h of development are described. The climbing perch is characterized by positive buoyancy of eggs and early larvae not usually found in other freshwater fish. This allows the fish development close to the surface of the water and is enabled by a large oil globule in the yolk. The data on the spatial orientation of the larva body, their vertical distribution in the water column, the beginning of exogenous feeding and locomotion, and the fright reaction of the larvae at different ages are presented. The most significant changes in the behavior of the climbing perch larvae are associated with changing the shape of the yolk sac, beginning to function as a provisional hydrostatic organ from about the 80s hour of development.

Keywords: climbing perch *Anabas testudineus*, pelagic eggs, larvae, buoyancy dynamics, larval behavior, fright, startle reaction, dispersed systems, fresh water

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INTRODUCTION

Climbing perch *Anabas testudineus* is a one of the most common freshwater fishes in South and South-east Asia belonging to the suborder anabantoids or labyrinth fish (Anabantoidei)¹. It is characterized by pelagic eggs with positive buoyancy (Zworykin, 2012; Zalina et al., 2012), which is rare in representatives of freshwater ichthyofauna (Davis, 1959; Mellinger, 1994; Makeeva and Pavlov, 2000). It is assumed that the location of eggs close to the water surface in the oxygen-poor tropical water bodies improves the respiration conditions for the embryo. In early larvae, the remaining lipid reserve seems to fulfill a temporary hydrostatic function (Soin, 1968; Soin et al., 1973). Adaptations that provide climbing perch with positive buoyancy in early ontogeny should obviously be reflected in its developmental characteristics and behavior pattern of early larvae. However, buoyancy has not been considered in previous publications on the ontogeny of this fish (Moitra et al., 1987; Amornsakun et al., 2005; Morioka et al., 2009; Zalina et al., 2012; Sarkar et al., 2015). The same is true for other well-known representatives of the suborder, in particular to *Trichopodus trichopterus* (Hodges and Behre, 1953; Morioka et al., 2012), *T. pectoralis* (Amornsakun

et al., 2004), *Trichogaster lalius* (Saha et al., 2017), *Osphronemus goramy* (Amornsakun et al., 2014), and *Helostoma temminckii* (de Sousa and Severi, 2000).

Assessment of buoyancy (especially positive) in small organisms is not an easy task (Power et al., 1991). In climbing perch, it required the development of a special technique that uses the property of substances suspended in water (similar to dissolved substances) to influence the buoyancy of immersed objects (Dzerzhinskiy, 2012). In previous works (Dzerzhinskiy and Zworykin, 2012; Dzerzhinskiy, 2016), we used this technique to determine the climbing perch buoyancy dynamics in early ontogeny, which showed a complex pattern.

The aim of this work is to describe the morphological and functional features of hydrostatically significant structures of eggs and larvae of the climbing perch and to analyze the behavior of the larvae in relation to changes in the hydrostatic characteristics of their body.

MATERIALS AND METHODS

The experiments were carried out in the laboratory of the Coastal Branch of the Russian-Vietnamese Tropical Research and Technological Center (Nha Trang, Vietnam). Biological material was obtained from the Institute of Aquaculture in Ninh Fung

¹ Some researchers allocate a separate order Anabantiformes (Ruggiero et al., 2015; Betancur-Retal., 2017).

(Khánh Hòa Province). The work was carried out on three series of fertilized eggs received on December 18 and 19, 2011, and January 11, 2012. The first series of eggs was transported from the Institute, while the second and third were obtained directly in the Russian-Vietnamese Tropical Research and Technological Center (Tropical Center) as a result of artificially induced spawning (Zworykin, 2012). Eggs from a spawning aquarium were removed immediately after the end of the spawning. The age of embryos and larvae was counted from the moment of spawning.

The development of eggs and hatched larvae² took place in an aerated aquarium, the water temperature in which depended on the ambient room temperature. In the case of the first two series (December 19–23, 2011), it varied between 21.6–and 25.6°C (average 24.1°C), while that for the third series (January 11–16, 2012) varied from 24.1 to 26.1°C (average 24.9°C). To stimulate the development of protozoa, which could be the starting food for larvae, a small amount of dried banana peel and aquarium fish food flakes were added to the bottom of the aquarium. As the eggs and larvae developed, samples were taken: the measurement of buoyancy was carried out simultaneously with observations of the development and behavior of the larvae and on the same living material.

The measurement of density of eggs and developing larvae was carried out titrimetrically using dispersed systems of various densities. Initial density of not swollen eggs was calculated (see Dzerzhinskiy, 2016).

We focused on the development of the structures that are directly related to buoyancy and affect the position and spatial orientation of the eggs and larvae. An OPTICA microscope with a Nikon Coolpix 4500 digital camera was used to monitor the development of climbing perch. For photographing, the larvae were anesthetized with an alcohol solution of benzocaine. The observation was carried out both from above and laterally. For better observation, the objects were placed in a watch glass. For lateral viewing, a vertical chamber was used. It was a rectangular glass cuvette with an internal dimension of 5 × 18 × 52 mm. The microscope was installed in such a way that the optical axis of the lens was horizontal, and the cuvette with the objects was fixed vertically (Chernyaev, 1962).

The dimensional characteristics of the eggs, as well as the total body length (*TL*) of the larvae, were determined by digital micrographs (Dzerzhinskiy, 2016). The diameters of the egg, yolk, and the oil globule were calculated as the average between two diameters, the largest one and that perpendicular to it. The initial diameter of the nonactivated egg was evaluated by analyzing the contents of the climbing perch ovary, presumably at the final stage of maturation, shortly

before ovulation. Since multiple egg release is typical for the climbing perch (Zworykin, 2012), the diameter of only the highest generation of oocytes was measured. The swim bladder volume was calculated using the formula for revolution ellipsoid (Lindsey et al., 2010): $V = 4/3\pi ab^2$, where *a* and *b* are the largest and the smallest radii of the swim bladder, respectively.

Behavioral activity of the larvae was recorded in the vertical chamber described above by direct observation and photographically. It should be emphasized that we studied only those forms of the simplest behavioral activity of the larvae that were directly related to the change in their hydrostatic features. The following characteristics were recorded: spatial orientation of the larva body (which part of the body is oriented upwards), the vertical distribution of the larvae in the water column at rest and when frightened, and the ability of the larvae for coordinated locomotion and exogenous feeding.

Preliminary observations showed that the larvae stay close to the surface of the water or (from the age of 60 h) in the water column in the rest state, whereas frightened larvae sharply rush to the bottom. Detailed analysis included five experiments (Table 1) in which vertical distribution of the larvae of different ages in the water column at rest and in a frightened state was recorded photographically. To carry them out, a random number (from 10 to 21) of intact larvae were taken from the home aquarium into an experimental cuvette. Tested larvae were never returned back after the experiments. The distribution of resting larvae was photographed at random intervals. The larvae were frightened by double-knocking the side wall of the cuvette with a tweezers; the picture was taken 1–2 s later. The patterns of larvae distribution were determined by counting their number in the upper and lower parts of the chamber.

The statistical analysis included Kruskal–Wallis test (*H*) and the randomization test (*Z*) (Hollander and Wolfe, 1999). Data analysis was conducted using the R software package.

RESULTS

Embryonic Development

Eggs of the climbing perch are relatively small (Table 2). During the first 15 min after spawning, they reach the final size of ~1 mm. The developing eggs have extensive perivitelline space. There is a large oil globule in the yolk that provides positive buoyancy (Fig. 1a). Its diameter is slightly less than the diameter of the entire yolk. During the first 20 h of development, no significant changes were observed in either the egg diameter (*H*: $\chi^2 = 3.6$, *df* = 2, *p* = 0.162) or the diameter of the oil globule (*H*: $\chi^2 = 2.5$, *df* = 2, *p* = 0.291). Further changes in the size of the oil globule were difficult to estimate precisely because its boundaries were not clearly visible, and the shape was

² During the experiment, the transfer of fish to a mixed diet was noted. However, for the entire period after hatching, the term “larva” is used in the work.

Table 1. Characteristics of the biological material used in behavioral experiments

Experiment no.	Date	Larvae number, specimens	Larvae age (after the end of spawning), h:min
1	January 13, 2011	21	62:40
2	January 14, 2011	15	79:10
3	January 14, 2011	15	90:30
4	January 15, 2011	18	107:15
5	January 16, 2011	10	139:10

Table 2. Dimensional characteristics of oocytes, eggs, and larvae of the climbing perch *Anabas testudineus*

Index, age	<i>M</i>	min	max	<i>SD</i>	<i>n</i> , spec.
Diameter of mature, unovulated oocytes, mm	0.72	0.67	0.77	0.019	23
Diameter of developing eggs, mm:					
15 min	1.02	0.97	1.09	0.028	21
17 h 5 min	1.00	0.94	1.08	0.029	21
20 h 25 min	1.01	0.95	1.09	0.036	21
20 h 34 min	1.01	0.97	1.04	0.019	19
Diameter of the oil globule, mm:					
15 min	0.55	0.53	0.58	0.013	21
17 h 5 min	0.55	0.52	0.58	0.016	21
20 h 34 min	0.54	0.51	0.58	0.020	19
Yolk diameter, mm					
15 min	0.64	0.61	0.67	0.019	21
Swim bladder volume, cm ³					
94–140 h	1.2×10^{-6}	0.8×10^{-6}	1.8×10^{-6}	4.13×10^{-7}	9

(*M*) mean value; (min) and (max) are the limits of variation of the index; (*SD*) standard deviation; (*n*) number of eggs or larvae studied. Swim bladder volume was measured in larvae with *TL* 3.80 (*SD* = 0.076) mm.

changeable. With the development of the egg, fouling emerges on its surface. The envelope is strongly deformed by the end of the embryonic period.

The beginning of egg cleavage was noted on the 35th minute of development at 24.6°C. Starting from this stage of development and prior to the segmentation, the embryo is always turned down in its spatial orientation (Fig. 1a). The oil globule, providing for positive buoyancy, also causes the embryo to rise to the upper wall of the egg within the perivitelline space. The heavier yolk stays down. From the age of ~16 h, when the embryo has developed the Kupffer's vesicle and the tail bud has been formed, body orientation could be different, apparently due to the convergence of the centers of gravity and buoyancy resulting from increased embryo size (Fig. 1b).

Hatching of larvae from the eggs brought from Ninh Fung (series 1) began at the age of 30 h. In series 2, the mass hatching of the larvae was noted at the age of 31 h 30 min; after 1 h, hardly any larvae remained inside the egg envelope. The beginning of hatching of

larvae in series 3 was noted at the age of 29 h 30 min. At the age of 30 h 23 min, 29% of the hatched larvae were recorded, there were 60% of the hatched larvae at the age of 32 h 38 min, and all the larvae hatched by 35 h 30 min.

The first rupturing of the egg envelope during larva hatching was caused by its intense tail movements. It was the tail that often appeared first from the larva egg shell. The front part of the larva body could remain inside the shell for a long time. Such larvae were located near the surface close to vertical position head down, touching the water surface by the tip of the tail. The remains of the egg shell represented a shapeless translucent mass covered with various pieces of fouling. According to the position of such larvae, these remains would have much higher density than the embryo. The larvae free from the shell, initially located in a horizontal position close to the surface of water, yolk sac upwards (Fig. 2a).

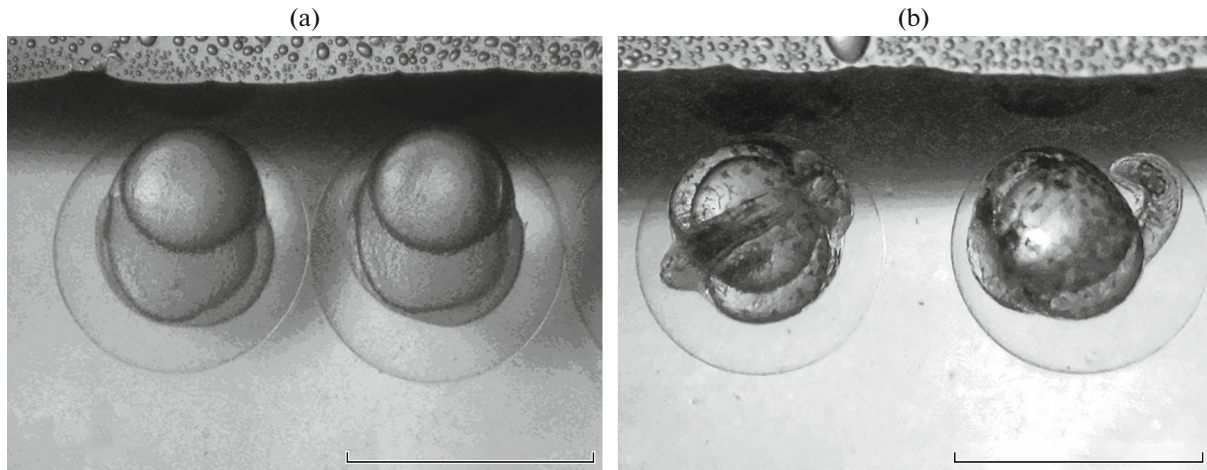


Fig. 1. Embryonic development of the climbing perch *Anabas testudineus* under the surface of water, lateral view: (a) gastrula, 8 h 51 min since spawning, (b) mobile embryo with separated tail section, 20 h 51 min. Scale here and in Fig. 2: 1 mm.

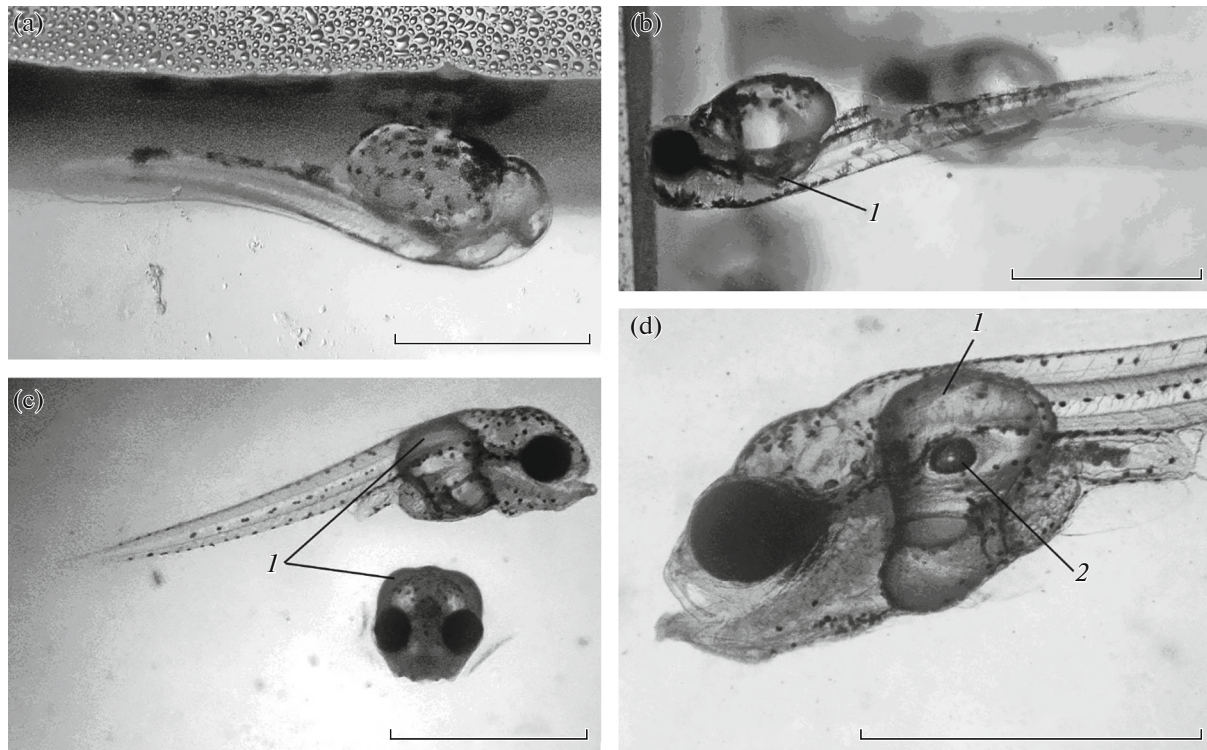


Fig. 2. Larvae of the climbing perch *Anabas testudineus*: (a) 31 h 48 min since spawning, (b) 62 h 44 min, (c) 79 h 51 min, (d) 107 h 55 min; 1, dorsal protrusion of the yolk sac; 2, swimbladder filled with gas.

Postembryonic Development and Behavior

In the beginning, at least until the age of 40 h, the larvae are practically immobile, being located directly under the surface of water in a horizontal position with the yolk sac upward (Fig. 2a). Later, their motor activity increases, which is reflected primarily in periodic energetic dives at different depths, followed by passive

ascent to the surface. This behavior resembles “zoom” that the larvae of many freshwater fish make at this developmental stage (Soin, 1968; Makeeva et al., 2011), with the crucial difference that the latter have an active ascent and a passive descent.

By the age of 62–63 h (Fig. 2b), the larvae at rest continued to stay with the yolk sac upwards. But they

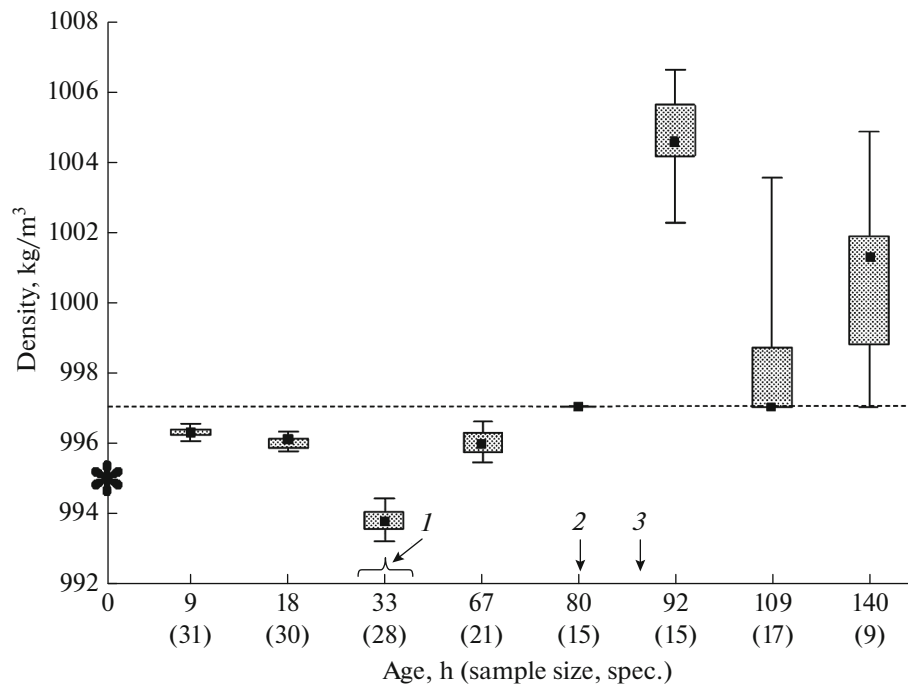


Fig. 3. Density dynamics of the eggs and larvae of the climbing perch *Anabas testudineus*: (*) calculated average value of the density of nonactivated egg at the time of spawn, (■) median, (□) 25–75% quartile, (---) neutral buoyancy; (1) hatching period; (2) transition to exogenous feeding; (3) filling of the swimbladder with air (according to Dzerzhinskiy, 2016, with additions).

also tended to spread to the middle and lower horizons of the experimental cuvette, and spent much less time near the surface. Their vertical distribution at frightening only slightly differed from that at rest. At rest, 58.3 (29.4–71.4)%³ of the larvae stayed in the upper half of the aquarium; after being frightened such were 40.0% (12.5–47.1)% ($Z = -1.9$, $p = 0.059$). Due to positive buoyancy, anesthesia resulted in surface distribution of all the larvae at this age. A similar spatial orientation of the larvae (belly upward) was observed until the age of 67 h.

Significant modification of the yolk sac shape was conspicuous among the behavior-related morphological changes of the larvae after hatching. Up to the age of 43 h, the yolk sac retained a regular, slightly elongated, rounded shape, and the oil globule also had a shape close to spherical (Fig. 2a). Then, the yolk sac gradually decreased in length and increased in height. Both sides of the body in the dorsal part formed protrusions filled with oil. At first, they were small (Fig. 2b), but reached the upper edge of the body in the corpus region by the age of 80 h (Fig. 2c). The result of this overgrowth of the yolk sac was a significant redistribution of oil in the larva body. It was first divided into two symmetrical portions, covering the body from two sides. Second, overgrowth of the yolk sac in the dorsal direction ensured the highest position of the oil glob-

ule. By the age of 80 h these changes altered the pattern of larva swimming rather radically. Larvae began to swim back up; they were evenly distributed over the 2-cm water column at rest. After being frightened, almost all larvae settled to the bottom. At rest, 48.3 (30.8–57.1)% of the larvae were distributed in the upper half of the chamber; after being frightened, this fell to 0 (0–7.1)% ($Z = -3.2$, $p = 0.002$). The buoyancy of anesthetized larvae of this age was close to neutral. At this stage of development, the swim bladder in all examined larvae was not filled with gas. At the same time, many of the larvae started to have non-empty intestine.

The first larvae with a filled swim bladder were noted at the age of 87 h 30 min. By the age of 90 h 30 min, 20% of the larvae (3 of 15) filled the swim bladder with gas. Just as before, they were evenly distributed across all water horizons at rest, and they settled to the bottom after being frightened. At rest, 38.5 (20.0–53.9)% of the larvae were in the upper half of the aquarium, while 0 (0–7.6)% ($Z = -3.9$, $p < 0.001$) were there after frightening. However, all of the anesthetized larvae lay at the bottom, as a result of negative buoyancy.

By age 107 h, the swim bladder was filled in 47% (8 of 17) of the larvae. The part of the yolk sac filled with lipids had relatively decreased in size. It did not reach the apex of the dorsal surface of the larva in height, but the characteristic extended upward shape of the oil globule was still preserved. Larvae with filled

³ Here and below: outside the brackets is the median; in parentheses, the limits of variation of the indicator.

swim bladder could have almost complete coincidence of the lateral projections of the center of the oil globule and the center of the swim bladder (Fig. 2d). All larvae (regardless of the presence or absence of gas in the swim bladder) had non-empty intestine. The distribution of larvae at rest and after frightening did not change from the previous observations: there were 42.8 (22.2–56.3)% and 5.8 (0–12.5)% of larvae ($Z = -3.2$, $p < 0.001$), respectively, in the upper half of the chamber. More than half of the anesthetized larvae (9 of 17) had neutral or very close to it buoyancy; all others had negative buoyancy.

At the age of 139–140 h, the behavior at rest and after frightening did not change. At rest state, 45.0 (30.0–70.0)% of the larvae were in the upper half of the chamber while none (0–0)% of larvae ($Z = -3.3$, $p = 0.002$) after frightening. The swim bladder was filled in 89% of the larvae (8 of 9). After anesthesia, the buoyancy of eight of nine larvae was negative. Only one larva had close to neutral buoyancy. We found no developmental abnormalities in the single larva with unfilled swim bladder.

Density Dynamics

Density dynamics of the eggs and larvae of the climbing perch are shown in Fig. 3. Positive buoyancy was typical for the early development of the climbing perch up to the age of 80 h: before the start of exogenous feeding, the eggs and larvae were located in direct contact with the surface of water. Temporary immersion of eggs and larvae could be caused by weak stirring of water, but they soon returned to the surface. The egg ascent rate at the beginning of the segmentation stage (age 12 h 40 min), measured visually on a 25 mm section, was 0.95 mm/s ($n = 23$, $SD = 0.198$). The buoyancy of eggs and larvae varied throughout the development. Freshly spawned, nonactivated eggs were characterized by high buoyancy. Egg activation was accompanied by hydration, mainly due to outside water, which decreased buoyancy. This subsequently increased during hatching. The density of the larvae just released from the egg shell was even lower than that calculated for nonactivated eggs. The larva density then gradually increased up to the beginning of filling the swim bladder, and somewhat decreased afterwards.

DISCUSSION

In most freshwater pelagic spawners with negative initial buoyancy of eggs, both hydration and lipid inclusions decrease eggs density (Zotin, 1961; Soin, 1968) and make buoyancy near-neutral. Nevertheless, such eggs retain some negative buoyancy and usually remain in the water column due to the flow turbulence (Battle and Sprules, 1960; Hopson, 1969; Makeeva and Pavlov, 2000). The buoyancy of oocytes in the climbing perch is initially positive. Hydration

decreases it with the same result, shifting to neutral. Thus, the effects of hydration and lipid inclusions on the buoyancy of eggs in the climbing perch are the opposite. However, the eggs retain positive buoyancy even at significant hydration to be able to develop near the surface or in midwater saturated with oxygen (Soin, 1968).

The positive buoyancy of true pelagic eggs in freshwater fish depends first on the large size of the oil globule, the density of which is lower than the density of fresh water. Before accurate measurement of the density of pelagic eggs become possible, the volume of the oil globule divided by the volume of the heavier yolk was used as a proxy for egg buoyancy (Hopson, 1969). Despite the obvious inaccuracy of this approach, a comparison of the relative oil globule volume in eggs of different fish species with truly pelagic eggs is quite informative (Table 3). In particular, it indicates that the climbing perch is characterized by the largest ratio of oil globule to the yolk volume among all freshwater pelagic spawners we are aware of. This suggests that the climbing perch eggs are characterized by the highest buoyancy. However, compared with the eggs of close *Trichogaster lalius* and *T. fasciata*, the relatively large amount of perritelline space in its eggs can partially offset the positive effect that is created by the extra-large oil globule. An accurate measurement of buoyancy by the method we proposed earlier (Dzerzhinskiy, 2016) could significantly clarify the picture.

In our opinion, increased buoyancy after hatching is associated with the loss of the chorion ballast. Increased chorion density with respect to the rest of the egg is known in pelagic spawners (Craik and Harvey, 1987; Kjesbu et al., 1992; Nissling et al., 1994).

Hydrostatic function of the oil globule is affected by its size, shape, and location. Gradual development of the dorsal protrusions of the yolk sac brings about increasing convergence of the center of buoyancy and the center of gravity. Therefore, the larva that was able to swim exclusively up-belly can proceed to the normal up-back swimming pattern. The positions of the centers of the swim bladder and the oil globule (Fig. 2d) are crucial for the common hydrostatic function of these structures. Obviously, they are linked with the center of gravity of the larval body. A different location of these structures could greatly complicate the control over the spatial position of the body, especially when one structure is gradually replaced with the other.

Thus, the hydrostatic function of the oil globule in climbing perch larvae is not limited to positive buoyancy. Its shape and relative position indicate that it would play an important role in coordinated swimming. The special properties of the oil globule and swim bladder can complement each other and optimize the hydrostatic capabilities of the larvae. The principal difference of the oil globule from the swim

Table 3. Relative volume of the oil globule in the eggs of some freshwater pelagic spawners

Species (family)	Volume of the oil globule, % of volume			Data source
	egg	yolk	pervitelline space	
<i>Lates niloticus</i> (Latidae)	13.9	27.7	28.0	Hopson, 1969
<i>Aplodinotus grunntiens</i> (Sciaenidae)	10.9	30.8	16.9	Davies, 1959 (cit ex: Hopson, 1969)
<i>Macquaria ambigua</i> (Percichthyidae)	0.9	38.5	0.9	Lake, 1967 (cit ex: Hopson, 1969)
<i>Trichogaster lalius</i> (Osphronemidae)	27.2	47.6	63.6	Analysis of the images from Saha et al., 2017
<i>Trichogaster fasciata</i> (Osphronemidae)	32.7	56.8	77.2	Analysis of the images from Islam et al., 2017
<i>Anabas testudineus</i> (Anabantidae)	15.7	63.5	20.8	Our data

bladder is its incompressibility and, as a consequence, independence of its hydrostatic properties on the depth. The gas-filled swim bladder, however, introduces vertical instability in the body position of the fish, which is especially pronounced with rapid changes in depth (Steen, 1970). The oil globule has no such drawback, this can be considered an important benefit. On the other hand, the compressibility of the swim bladder, in contrast to the oil globule, allows the larva to control its buoyancy.

The location of the oil globule varies significantly among different fish species (Hopson, 1969). Its location at some distance from the developing swim bladder can be observed, for example, in fish larvae where it is relatively small (Summerfelt, 1996; Hamm and Hinton, 2000; Villalobos et al., 2000; Chalde et al., 2014) and does not carry important hydrostatic function. Certain other species, e.g. the Nile perch *Lates niloticus*, have large oil globule, making for positive buoyancy of the larvae. However, it protrudes so far forward that the larva initially develops vertically with the head up (Hopson, 1969). Hopson suggests that as the larva grows and its head section increases, the centers of gravity and buoyancy should converge. However, by the time this may be really important (the beginning of free swimming and exogenous feeding), the oil globule almost disappears.

Studies of the early development of the climbing perch (Morioka et al., 2009; Zalina et al., 2012) indicate that the oil globule coexists with the swim bladder in the larvae for quite a long time, at least until 7 days after fertilization. The initial filling of the swim bladder in the larvae occurs during a limited period, usually coinciding with the transition to exogenous feeding (Chatain, 1986; Trotter et al., 2005) or precedes it (Palińska-Żarska et al., 2014). This enables coordinated swimming ability required for successful search for food. In the climbing perch, coordinated swimming and the associated exogenous feeding were observed even before the swim bladder was filled with

air, pointing to important and independent hydrostatic role of the oil globule.

Filling of the swim bladder in the climbing perch was documented from the age of 87 h and 30 min. Therefore, the larva hydrostatics was determined by at least three factors: the yolk sac, the swim bladder, and the gastrointestinal tract and its content. This, could account for the high variability of the larva density (see Dzerzhinskiy, 2016, for more details). Coordination of the different hydrostatic systems in the larvae of the climbing perch is still unclear.

According to various estimations, the swim bladder volume required for neutral buoyancy in freshwater fish is usually 7–8% of the body volume (Schmidt-Nielsen, 1979; Evans, 1998). However, some catfish (Siluriformes) and labyrinth fish with additional respiratory organs may experience a noticeable reduction in the size of the swim bladder (Gee, 1976; Hasan, 1966). It was suggested (Qasim and Hasan, 1961) that these organs, like the air collected in the oral cavity (Gee and Gee, 1995), significantly affect fish hydrostatics. Thus, the hydrostatic load on the swim bladder decreases, resulting in reduction of its volume (Alexander, 1964).

It is noteworthy that with a similar age and body size ($TL = 3.8$ mm), the volume of the swim bladder of the climbing perch larvae is less than that in larval zebrafish *Danio rerio* (Lindsey et al., 2010) by almost 13 times (1.2×10^{-6} vs. 1.5×10^{-5} cm³). Unlike labyrinth fish, zebrafish larvae lack additional hydrostatic devices. A visual comparison of the available images of climbing perch larvae with images of larvae of other freshwater fish available in the literature also unveils relatively small swim bladder volume in the climbing perch. Even though reduction of the swim bladder in the climbing perch larvae can be accounted for by morphological and physical causes, additional experiments are necessary, considering the volume and the specific density of the larva in addition to linear dimensions.

The specificity of the climbing perch hydrostatics is not limited by the early ontogeny. As the oil globule reduces, it develops an additional respiratory organ, the labyrinth (Morioka et al., 2009). Unlike the larval oil globule, the labyrinth also affecting the body hydrostatics, strongly protrudes forward relative to the swim bladder, and provides an additional buoyancy center. This should introduce further complications in the control and stabilization of the horizontal body position of the fish.

The features of the swim bladder and the oil globule documented in this paper along with their complex combined effects are characteristic of other representatives of anabantid fish (Bhimachar et al., 1944; Soin, 1968; Soin et al., 1973). The structure and characteristics of the egg development, buoyancy, timing of the exogenous feeding and locomotion, along with a number of other patterns, constitute a complex reproductive strategy. In turn, the reproductive strategy plays a crucial role in the evolution of this group and is important for understanding their phylogeny (Britz, 1997; Rüber et al., 2006; Zworykin, 2017).

The behavior of the larvae is also of undoubted interest for further research. The startle escape, an anti-predator response, is one of the most developmentally early behaviors known in many fishes (Noakes and Godin, 1988). This kind of rapid reflex movement is typically directed away from the source of danger and includes a S-shaped bending phase (Hale et al., 2002; Witt et al., 2015). Even though this response is characteristic of fish not only at the larval stage, its development and ontogenetic modifications are of particular interest (Kimmel et al., 1974; Li et al., 2014; Roberts et al., 2016). Noteworthy, the rapid escape in response to frightening stimuli appear in the climbing perch larvae relatively early. For comparison, such reaction appears in zebrafish on the fourth to fifth day after fertilization (Kimmel et al., 1974; Roberts et al., 2011), while we began to observe it in climbing perch at least in the middle of the third day. Of course, its manifestation would depend both on the type of stimulus and on the development of the systems that control the response to the stimulus.

The fish species in which this response has been traditionally studied are the zebrafish, the goldfish *Carassius auratus*, and several other. We are not aware of prior publications analyzing this type of early behavior in the climbing perch. Obviously, in the case of climbing perch larvae which stay under the surface of water, the direction of the startle jerk is limited to downwards into the water at some angle. Additionally, larva with positive buoyancy would passively float towards the surface after the jerk. We suggest that for the larvae of this species that often breed in temporary reservoirs with muddy water and few aquatic predators, jerks down would provide an adaptive tactic to avoid threat from the air. Testing this hypothesis would be an interesting study.

Thus, early ontogeny of the climbing perch is of great interest for understanding of the relationships between various provisional and definitive organs and systems, the development and transformation of these relationships, interactions with the age, and their effects on fish behavior.

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