

Article

## Larval development of *Characidium orientale* (Actinopterygii: Crenuchidae) a small Neotropical fish

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**ABSTRACT.** The *Characidium orientale* Buckup & Reis, 1997 larvae development is described using specimens collected in the natural environment of Antas River, in the Taquari-Antas river basin, Rio Grande do Sul, Brazil. Were considerate ontogenetic changes in morphology, pigmentation, fin morphology, and meristic characters. Thirty-six larvae (33 in the preflexion stage, two in flexion, and one in the postflexion stage), with a standard length between 4.24 and 11.26 mm were analyzed. The larvae of *C. orientale* are altricial and present fusiform body, subterminal mouth, long intestine with anal opening posterior to the vertical line over the median region of the body. Two chromatophores are evident in the interorbital region, as well pigments concentrated on the side of the body, forming a longitudinal stripe, and between the rays of the caudal fin, which intensify during the development. Also, a conspicuous remnant of the embryonic membrane (finfold) in front of the dorsal fin appears to be characteristic of species of the genus during larval development. The total number of myomeres varies from 32 to 35 (18-23 pre; 11-16 postanal). The main changes in morphometry occur during the preflexion stage and suggest physiological and behavioral changes.

**KEYWORDS.** Characiformes, fish larvae, ontogeny, Taquari-Antas River.

**RESUMO.** Desenvolvimento larval de *Characidium orientale* (Actinopterygii: Crenuchidae). O desenvolvimento das larvas de *Characidium orientale* Buckup & Reis, 1997 é descrito a partir de espécimes coletados no rio Antas, na bacia do rio Taquari-Antas, Rio Grande do Sul, Brasil. Foram consideradas mudanças ontogenéticas na morfologia, pigmentação, morfologia das nadadeiras e caracteres merísticos. Foram analisadas 36 larvas (33 na fase de pré-flexão, duas em flexão e uma em pós-flexão), com comprimento padrão entre 4,24 e 11,26 mm. As larvas de *C. orientale* são altriciais e apresentam corpo fusiforme, boca subterminal, intestino longo com abertura anal posterior à linha vertical sobre a região mediana do corpo. Dois cromatóforos são evidentes na região interorbital, bem como pigmentos concentrados na lateral do corpo, formando uma faixa longitudinal e entre os raios da nadadeira caudal, que se intensificam durante o desenvolvimento. Além disso, um remanescente conspicuo da membrana embrionária na frente da nadadeira dorsal parece ser característico de espécies do gênero durante o desenvolvimento larval. O número total de miômeros varia de 32 a 35 (18-23 pré; 11-16 pós-anal). As principais mudanças na morfometria ocorrem durante o estágio de pré-flexão e sugerem mudanças fisiológicas e comportamentais.

**PALAVRAS-CHAVE.** Characiformes, larvas de peixes, ontogenia, rio Taquari-Antas.

The reproductive dynamics of fish is often evaluated based on studies involving ichthyoplankton since data on the distribution and occurrence of eggs and larvae provide indicators to the delimitation of reproductive sites (NAKATANI *et al.*, 1997) as well as other important information for planning measures to protect the fish fauna (HUMPHRIES & LAKE, 2000). However, such assessments encounter difficulties identifying organisms in the early stages (BIALETZKI *et al.*, 1998).

The morphological similarity between species in the larval stages (FUIMAN, 1983; ORSI *et al.*, 2016), and the dissimilarity of these stages compared to the adult phase, can cause mistakes in the identification of the ichthyoplankton

(LEIS & TRNSKI, 1989; BIALETZKI *et al.*, 2016). Furthermore, the ontogenetic development of a great number of species of fish in tropical regions has not yet been documented (TAGUTI *et al.*, 2009; OLIVEIRA *et al.*, 2012; REYNALTE-TATAJE *et al.*, 2020). This scenario highlights the importance of characterizing the initial stages of fish, whose contribution allows for adequate taxonomic studies and, consequently, the understanding of population dynamics.

Among the largest contributors to the Neotropical fish assembly, the order Characiformes stands out (REIS, 2013), whose high diversity of species with similar morphological characteristics makes its taxonomic classification difficult. Evidence of this difficulty is observed in divergences regarding

the phylogenetic positioning of this order (e.g., BUCKUP, 1998; MIRANDE, 2010), as well as their families (MIRANDE, 2018; TERÁN *et al.*, 2020). One example is Crenuchidae, currently composed of the subfamilies Crenuchinae and Characidiinae, the latter of which was previously considered a subfamily of Characidae (BUCKUP, 1998).

Although they are present in most Neotropical freshwater basins, the greater participation of the genera of the Crenuchidae family is observed around the Orinoco and Amazon rivers basins, with the exception for the genus *Characidium* Reinhardt, 1867, which has the greatest range of distribution and number of species, with more than 80 valid nominal taxa (NELSON, 2006; FRICKE *et al.*, 2021). In the Laguna dos Patos basin, drainage located in southern Brazil and eastern Uruguay, five species of the genus have confirmed occurrences (all native): *Characidium* gr. *zebra* Eigenmann, 1909, *C. orientale* Buckup & Reis, 1997, *C. pterostictum* Gomes, 1947, *C. rachovii* Regan, 1913 and *C. tenue* (Cope, 1894) (BECKER *et al.*, 2013; FRICKE *et al.*, 2021). However, only *C. orientale* occurs in the plateau region of the Taquari-Antas sub-basin (LUZ-AGOSTINHO *et al.*, 2010; BECKER *et al.*, 2013). This species has an average size of 54.3 mm, dorsal profile slightly convex, rounded snout, terminal mouth, and compressed body (BUCKUP & REIS, 1997). According to LUZ-AGOSTINHO *et al.* (2010), the species presents with dark vertical bars that extend from the dorsum to just beyond the dark longitudinal strip located along the midline and is considered insectivorous, sedentary, without parental care, and whose reproductive activity occurs mainly in small tributaries.

In Brazil, there is a notable lack of the descriptions of development initial stages of fish larvae (REYNALTE-TATAJE *et al.*, 2020), as evidenced by the small number of specific guides and manuals for identification (e.g. NAKATANI *et al.*, 2001; LEITE *et al.*, 2007; ORSI *et al.*, 2016), beyond articles that reach only a small portion of the Neotropical fish fauna (e.g. OLIVEIRA *et al.*, 2012; SANTOS *et al.*, 2020; LIMA *et al.*, 2021; OLIVEIRA *et al.*, 2021). Also, most studies are aimed at medium and large species of commercial and recreational fishing interest. Unlike those species with relevance to aquaculture, small species such as belonging to genus *Characidium* are poorly known concerning to initial ontogeny, being the only existing description of initial development is restricted to *C. zebra* Eigenmann, 1909 (cited as *C. fasciadorsale* Fowler, 1914 by PONTON & MÉRIGOUX (2001)). Thus, considering the importance of recognizing the early stages of the fish life cycle for subsequent approaches, this study describes the morphological and meristic characteristics, as well as morphometric relationships of the larval stages of *C. orientale*, of specimens collected in the natural environment. Additionally, we seek to identify possible changes in these characteristics during ontogenetic development.

## MATERIAL AND METHODS

The larvae were collected every other week between October 2016 and January 2017 in the Antas River (28°57.0'

and 29°09.0'S – 51°54.0' and 51°06.0'W), located in the Taquari-Antas River basin, state of Rio Grande do Sul, Brazil. The Taquari-Antas River constitutes an important tributary of the Jacuí River that drains to Laguna dos Patos system, an ecoregion in southern Brazil and eastern Uruguay, which drains the southern slopes of the Serra Geral Mountain range. The Taquari-Antas River presents a high average slope and is characterized by torrential rainfall regimes, rapid surface runoff, and abrupt discharge variations throughout the year (FUNDAÇÃO ESTADUAL DE PROTEÇÃO AMBIENTAL HENRIQUE LUIS ROESSLER, 2009; FRESHWATER ECOREGIONS OF THE WORLD, 2015).

Samples were obtained using conical-cylindrical plankton nets (0.5 mm mesh and 36 cm of mouth diameter) equipped with a flowmeter to obtain the volume of filtered water. Each net was exposed for 10 minutes at four times: 21:00, 01:00, 05:00, and 09:00 hours. At each time, samples were obtained using horizontal trawls on the surface. Additionally, stationary nets were installed in the middle of the river and along the margin with greater flow, in sections transverse to the watercourse. A light trap (ÁVILA-SIMAS *et al.*, 2014, adapted from REYNALTE-TATAJE *et al.*, 2012) was also used in the margin of the lower water flow at nightfall (09:00 h) and removed before sunrise (05:00 h). Samples were then stored in flasks and fixed with 4% formalin buffered with calcium carbonate (CaCO<sub>3</sub>). These sampling methods followed procedures laid down in the Catch Authorizations linked to the Operation Licenses numbers 2844/2016-DL, 1353/2015-DL and 1264/2015-DL.

The samples were sorted, and the individuals were separated and identified using the developmental sequence technique proposed by AHLSTROM & MOSER (1976). This technique consists of comparing the morphology of smaller individuals to a known juvenile form. After identification, the specimens were classified according to the degree of the notochord development (AHLSTROM *et al.*, 1976, modified by NAKATANI *et al.*, 2001) into preflexion, flexion, and postflexion stages. The individuals that best represented the morphological characteristics were illustrated using a *camera lucida* and photographed using a digital camera Opticam (LOPT 18003) coupled to a stereomicroscope. The individuals used in this study are stored in the Ichthyology Collection of the Núcleo de Pesquisas em Limnologia, Ictiologia e Aquicultura (Nupélia) of the Universidade Estadual de Maringá (UEM) (NUP 022283, NUP 022284, NUP 022285, and NUP 022286).

To characterize the morphometry of early development, the following variables were recorded (AHLSTROM *et al.*, 1976) and expressed in millimeters (mm): standard length (SL), snout length (SnL), eye diameter (ED), head depth (HD), head length (HL), body depth (BD) as well as the length from the tip of the snout to origins of pectoral (SnP), pelvic (SnV), dorsal (SnD), and anal (SnA) fins. For meristic characterization, were counted whenever possible the number of total, preanal, and postanal myomeres, as well as the rays of the pectoral (P), pelvic (V), dorsal (D), and anal (A) fins.

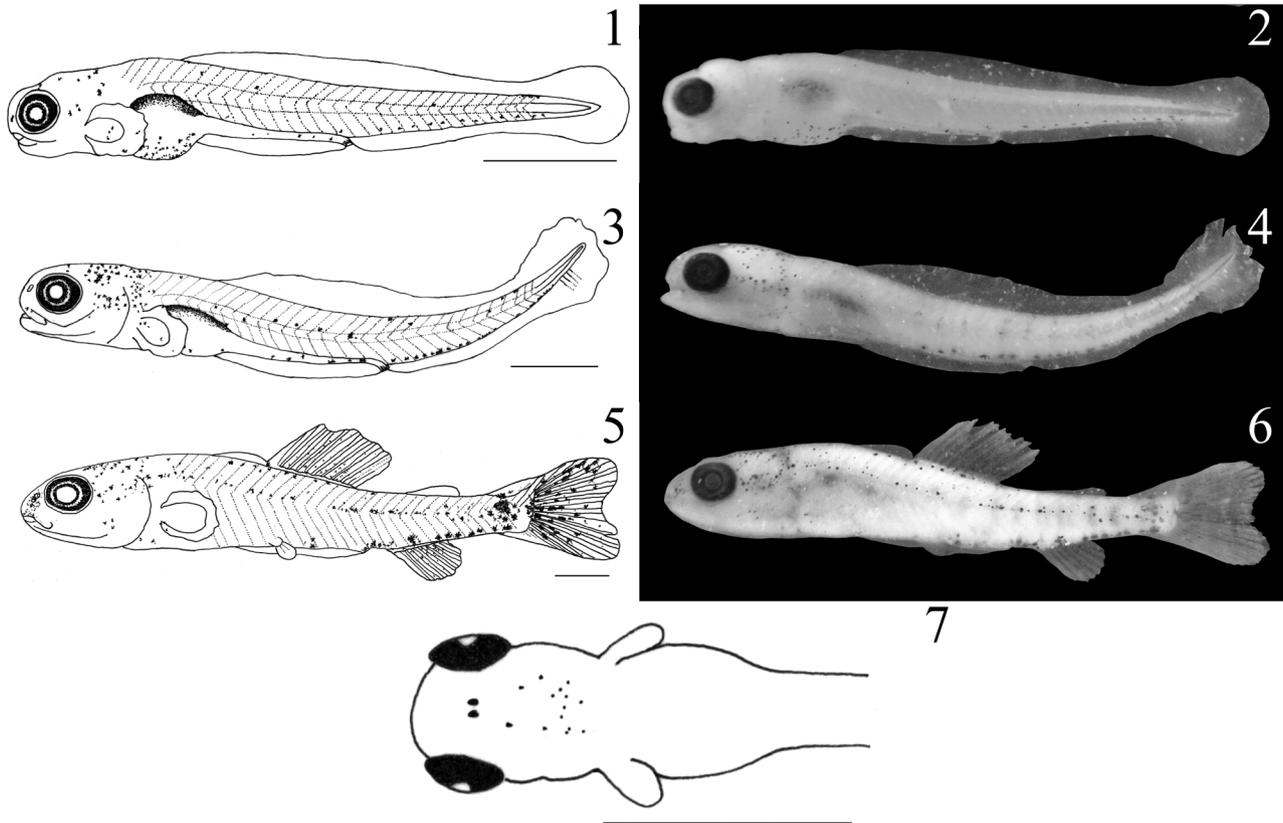
The body shape was determined according to the categories proposed by LEIS & TRNSKI (1989).

To examine the patterns of growth during the early development, the morphometric variables (dependent variables) were plotted against SL and HL (explanatory variables), and their relationships were analyzed using regression models (KOVÁČ *et al.*, 1999). First, we tested the hypothesis that development is continuously isometric using a simple linear regression model. Moreover, we also tested two alternative hypotheses: gradual allometric growth by using quadratic regression analysis and discontinuous isometric growth by using a piecewise linear regression analysis, which is characterized by breakpoints reflecting divergent growth rates. The optimal models for each morphometric variable relative to SL and HL were determined using F tests (SOKAL & ROHLE, 1981). The significance level for the analyses was  $p < 0.05$ .

## RESULTS

Thirty-six larvae of *C. orientale* were analyzed, 33 in preflexion, two in flexion, and one in postflexion stage. The description of each stage is shown below and illustrated in Figs 1–6. The results regarding the morphometric and meristic variables are listed in Tab. I.

Larvae in preflexion stage (Figs 1, 2) (4.24 and 5.62 mm SL) were characterized mainly by the elongated body and notochord in a straight position, that is visible through the transparent skin, besides the presence of the yolk sac, which was practically absorbed. The snout was rounded, the nostrils simple, the mouth subterminal, and the operculum unformed. Eyes were rounded and already pigmented. The swimming bladder appeared inflated and was visible through the transparent skin. The intestine was functional and the anal opening was located posterior to the vertical line that passes over the median region of the body. The larvae presented few punctiform chromatophores throughout the body; those that occurred were concentrated mainly in the upper head region, with two chromatophores evident in the interorbital region and some randomly distributed, including that of the caudal region. In the swimming bladder region, the chromatophores were distributed above it, while in the yolk sac, they were located below it; in the ventral region, they formed a line that begins before the anal opening and extended until a point close to the end of the notochord. The finfold was hyaline and devoid of pigmentation and could be seen surrounding the body dorsoventrally. The pectoral fin button was present but without evident rays. The other fins were not outlined. The total number of myomeres varied between 32 and 35, with 18 to 20 pre and 13 to 16 postanal.



Figs 1–7. Early development of *Characidium orientale* Buckup & Reis, 1997 larvae, collected between October 2016 and January 2017 in the Antas River basin, Rio Grande do Sul, Brazil: 1, 2, preflexion (4.94 mm SL); 3, 4, flexion (7.04 mm SL); 5, 6, postflexion (9.68 mm SL); 7, details of chromatophores on the top of the head (scale = 1 mm).

Tab. I. Morphometric variables, body relations (%), and meristic counts in larvae of *Characidium orientale* Buckup & Reis, 1997, collected between October 2016 and January 2017 in the Antas River basin, Rio Grande do Sul, Brazil (Min, minimum; Max, maximum; X, mean values; SD, standard deviation; PF, preflexion; FL, flexion; FP, postflexion; NV, not visible; n, number of individuals; SL, standard length; SnL, snout length; ED, eye diameter; HL, head length; HD, head depth; BD, body depth; SnP, snout-pectoral fin length; SnV, snout-pelvic fin length; SnD, snout-dorsal fin length; SnA, snout-anal fin length; fins: P, pectoral; V, pelvic; D, dorsal; A, anal).

Variables	Stages (mm)				
	PF (n=33)		FL (n=2)		FP (n=1)
	Min/Max	X ± SD	Min/Max	X ± SD	
SL	4.24-5.62	5.09±0.28	6.18-7.04	6.61±0.61	9.68
SnL	0.07-0.19	0.13±0.03	0.19-0.21	0.20±0.01	0.47
ED	0.30-0.47	0.40±0.04	0.49-0.54	0.52±0.04	0.87
HL	0.73-1.06	0.86±0.09	1.18-1.25	1.22±0.05	2.20
HD	0.56-0.93	0.76±0.07	0.98-1.03	1.00±0.04	1.63
BD	0.61-0.88	0.78±0.06	0.84-1.00	0.92±0.11	1.72
SnP	0.83-1.22	1.03±0.09	1.24-1.43	1.34±0.13	2.47
SnV	NV	NV	NV	NV	4.64
SnD	NV	NV	NV	NV	4.59
SnA	NV	NV	NV	NV	6.81
Relations (%)					
SnL/HL	8.43-21.79	14.77±3.09	16.10-16.80	16.45±0.49	21.36
ED/HL	36.96-56.41	46.78±4.99	41.53-43.20	42.36±1.18	39.55
HD/HL	74.67-103.61	88.65±6.85	82.40-83.05	82.73±0.46	74.09
HL/SL	14.74-20.00	16.82±1.36	17.76-19.09	18.42±0.95	22.73
BD/SL	14.04-17.28	15.29±0.90	13.59-14.20	13.90±0.43	17.77
SnP/SL	15.75-23.27	20.31±1.59	20.06-20.31	20.19±0.18	25.52
SnV/SL	NV	NV	NV	NV	47.93
SnD/SL	NV	NV	NV	NV	47.42
SnA/SL	NV	NV	NV	NV	70.35
Myomeres					
Total	32-35	33.06±0.91	33-35	34.00±1.41	34
Preanal	18-20	19.16±0.75	18-21	19.50±2.12	23
Postanal	13-16	13.94±0.95	14-15	14.5±0.71	11
Rays					
P	NV	NV	NV	NV	NV
D	NV	NV	NV	NV	9
V	NV	NV	NV	NV	NV
A	NV	NV	NV	NV	6

The larvae in flexion stage (Figs 3, 4) (6.18 and 7.04 mm SL) had a fusiform body, flexed and visible notochord, and a fully absorbed yolk sac. Morphologically, the snout, nostrils, mouth, eyes and intestine do not have change from the previous stage, only the operculum was in formation. There was an increase in pigmentation, mainly in the head, with the increase of punctiform chromatophores and the appearance of some dendritic chromatophores randomly distributed throughout the body and dorsoventrally. The finfold was still present, but it was already possible to see the formation of some hypural bones. The pectoral fin button was present, but evident rays and the other fins were not yet outlined. The total number of myomeres varied from 33 to 35 (18 to 21 pre and 14 to 15 postanal).

The only individual captured at postflexion stage (Figs 5–6) (11.26 mm SL) owned body elongated and it was no longer possible to visualize the notochord. The nostrils were divided into two parts, the mouth remained in a subterminal position, and the anal opening was located posteriorly to the vertical line that passes over the median region of the body. Pigmentation intensified throughout the body, spreading over the dorsal region of the head, around the mouth, and on the back of the body. The two chromatophores, evident in the upper region of the head in the previous stages (Fig. 7), have become less noticeable with the development of more chromatophores in the region. There was the appearance of punctate chromatophores in the middle of the body forming a line that extended from the pectoral fin button to the caudal

peduncle and also some chromatophores between the rays of the caudal fin. Remnants of the finfold could be seen in the ventral region of the body along the intestine and, in the dorsal region, anteriorly the dorsal fin, which was relatively conspicuous. All fins were outlined, but not fully formed, and it was possible to visualize only nine rays on the dorsal fin and six on the anal fin. The total number of myomeres was 34, with 23 pre and 11 postanal.

Morphometric analysis revealed that during larval development, *C. orientale* owned a long body (14.04 to 17.77%), head length ranging from small to moderate (14.74 to 22.73%) and a large eye diameter (36.96 to 56.41%). The snout length varied from 8.43 to 21.79%, the head height from 74.09 to 103.61% and the snout-pectoral fin length from 15.75 to 25.52% (Tab. I). The pelvic, dorsal, and anal fins were only visible in the only individual in postflexion and represented 47.93%, 47.42% and 70.35%, respectively in relation to the standard length (Tab. I).

Regarding the growth patterns between the morphometric variables, the snout length, eye diameter, and head length were better represented by the piecewise linear regression model (two different isometric stages), and showing an increase in the growth rate from its inflexion point 0.14 mm HL, 0.42 mm HL, and 0.91 mm SL, respectively, while for the head depth the best model was the linear (Tab. II, Figs 8–11). Finally, the body depth and the snout-pectoral fin length were better represented by the quadratic model with positive allometry, indicating that these variables grow faster than the standard length (Tab. II, Figs 12, 13).

## DISCUSSION

We describe the larval development of *C. orientale*, focusing on morphological and morphometric changes during ontogeny. Larvae of this species can be typified as altricial; that is, they are small, transparent, and poorly developed at hatching (BALON, 1990; BIALETZKI *et al.*, 2016), and then go through a pronounced metamorphosis where the adult species characteristics are acquired throughout the initial development. This pattern is keeping with the

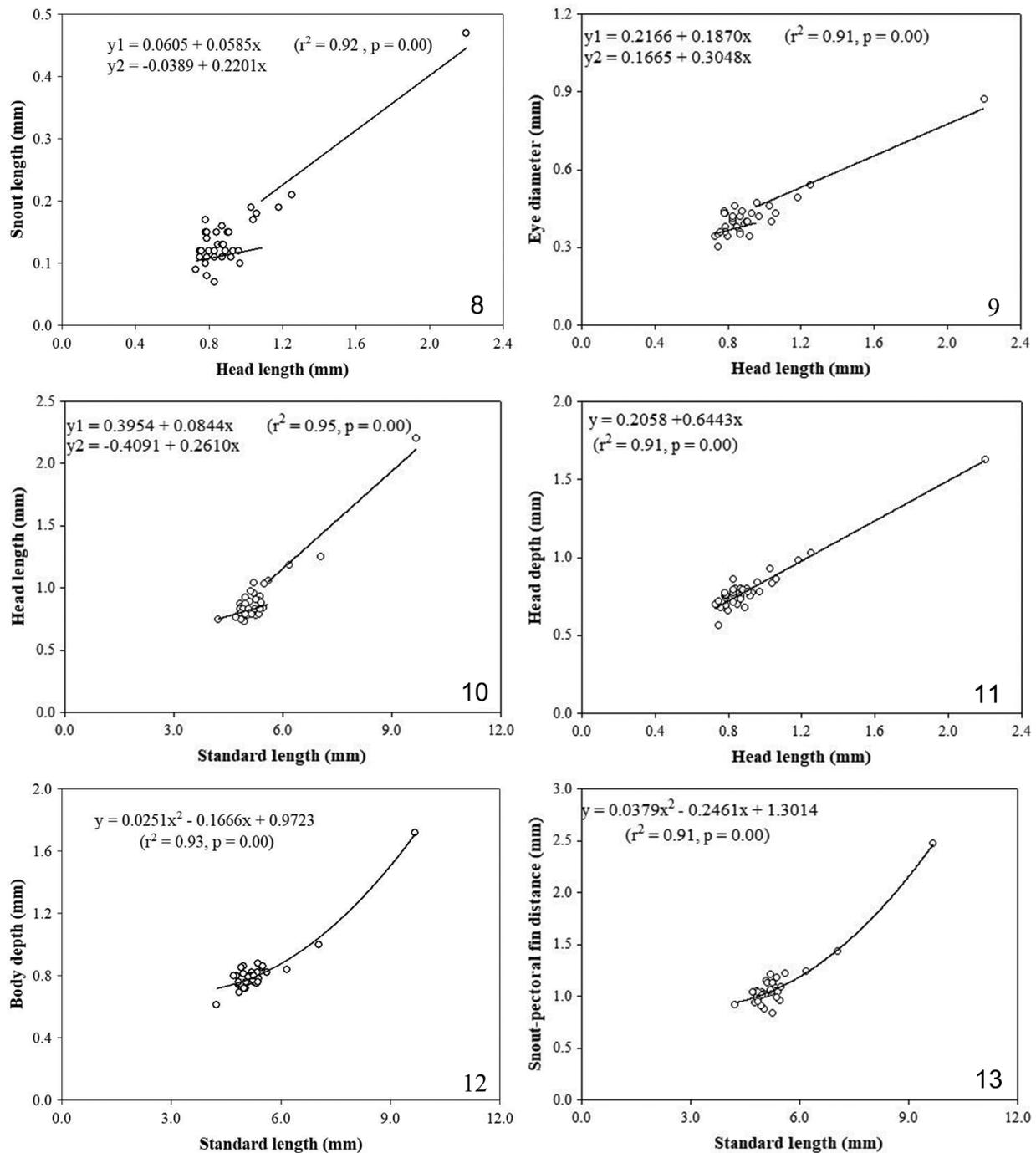
type of reproductive strategy presented by the species of *Characidium*, without parental care for offspring (LUZ-AGOSTINHO *et al.*, 2010).

*Characidium orientale* larvae exhibit an elongated body and subterminal mouth throughout their development. This pattern is similar to that observed in adults, as well as in the early stages of *C. zebra* (= *C. fasciadorsale* in PONTON & MÉRIGOUX, 2001). Morphologically, the fusiform body presents a greater performance in pelagic regions that do not present obstacles as they reach greater swimming speed (WERNER, 1977). ARANHA *et al.* (2000) suggest that the fusiform body, besides the enlarged and ventrally located pectoral and pelvic fins, allow species of *Characidium* to remain close to the substrate and resist the flow of water in rapids. These anatomical characteristics, in addition to the subterminal mouth, favor the capture of food in places that are difficult for other fish to access, such as in small spaces between rocks (ARANHA *et al.*, 2000), revealing a nektobenthic habit and exhibit a lurking predation tactic to capture aquatic insect larvae (ARANHA *et al.*, 2000; CASATTI & CASTRO, 2006; CARMO *et al.*, 2015).

In this aspect, despite the larvae also having a fusiform body and the subterminal mouth, the fins are not fully developed at the beginning of the postflexion stage, suggesting that these have not yet occupy the same habitat as adults and possibly also do not exhibit the same foraging behavior, as mentioned above. For *C. zebra*, for example, all fins (except pectorals) are formed from 10.0 mm (PONTON & MÉRIGOUX, 2001). The same authors report insect larvae in the diet of larvae of *C. zebra* of about 14 mm, and an increase in this food item only in individuals over 30 mm. CASATTI & CASTRO (1998) categorize the *Characidium* as sit-and-wait predators, when fish stays stationary on the bottom, capturing arrested food in short and successive leaps. However, although these species possibly consume similar food items, it is possible that the capture strategy is different, as *C. orientale* has a rounded snout, while *C. zebra* has a pointed snout (PONTON & MÉRIGOUX, 2001). In this sense, studies addressing the ontogeny of feeding can clarify this relationship between morphology and food capture.

Tab. II. Linear, quadratic, and piecewise regression statistics for the morphometric variables in relation to the head length and standard length of the larvae of *Characidium orientale* Buckup & Reis, 1997, collected between October 2016 and January 2017 in the Antas River basin, Rio Grande do Sul, Brazil ( $R^2$ , coefficient of determination; L, linear regression; Q, quadratic regression; S, piecewise regression; BM, best model; BP, breakpoint; N, number of individuals; SnL, snout length; HL, head length; ED, eye diameter; HD, head depth; SL, standard length; BD, body depth; SnP, snout-pectoral fin length; bold values,  $p < 0.05$ ).

Relation	$R^2$			F-test			BM	BP	N
	L	Q	S	Q/L	S/Q	S/L			
SnL/HL	0.84	0.86	0.92	3.66	<b>27.97</b>	<b>17.36</b>	S	0.14	36
ED/HL	0.83	0.85	0.91	2.10	<b>19.90</b>	<b>11.62</b>	S	0.42	36
HD/HL	0.91	0.92	0.93	0.16	<b>7.20</b>	3.69	L		36
HL/SL	0.90	0.92	0.95	<b>7.72</b>	<b>19.29</b>	<b>15.71</b>	S	0.91	36
BD/SL	0.88	0.93	0.89	<b>20.43</b>	<b>-9.69</b>	1.95	Q		36
SnP/SL	0.87	0.91	0.92	<b>16.56</b>	2.57	<b>9.97</b>	Q		36



Figs 8–13. Morphometric relationship (mm) between head length and snout length (8), head length and eye diameter (9), standard length and head length (10), head length and head depth (11), standard length and body depth (12) and standard length and snout-pectoral fin length (13) during the early development of *Characidium orientale* Buckup & Reis, 1997 larvae, collected between October 2016 and January 2017 in the Antas River basin, Rio Grande do Sul, Brazil.

Eye pigmentation and mouth opening development are events that occur simultaneously and are directly related to exogenous nutrition (LASKER *et al.*, 1970). These characteristics can be observed in the preflexion stage, with the functionality of the intestine and the partial absorption of the yolk sac, indicating the beginning of larval feeding activity.

The larval pigmentation pattern may represent a fundamental taxonomic character in the identification of species (OLIVEIRA *et al.*, 2012). KENDALL JR *et al.* (1984) argue that each species has a distinct pigmentation pattern, with the size and location of the melanophores being genetically determined. For *C. orientale* larvae, the most characteristic pigments are concentrated mainly in the head region, with

two chromatophores evident in the upper region and also in the body, forming a longitudinal band. Among the rays of the caudal fin, this pattern intensifies throughout development. The absence of two evident chromatophores in the head region of *C. zebra* is the main difference in pigmentation between these species (PONTON & MÉRIGOUX, 2001). NAKATANI *et al.* (1997) suggest that the pigmentation pattern of the larvae varies according to the environment where it is found. Species of the Crenuchidae family have a close relationship between the substrate and the presence of a cryptic coloring, used to camouflage over sand or rocks (GÉRY, 1977; BUCKUP, 1993; BUCKUP, 2003). This result reinforces the possibility that a change in the position of the water column occurs during development, changing from pelagic (larval period) to nekto-benthic (juveniles and adults).

Regarding the fins, the first to be present, and consequently, the one that supports the formation of the unpaired fins, is the embryonic fin or finfold. It is present in newly hatched larvae, serving as a propellant organ for locomotion and orientation (WEBB & WEIHS, 1986). The pectoral fin, which consists of only one membrane in the preflexion stage, is probably present at hatching, similar to that found in other genera of Characiformes (NAKATANI *et al.*, 2001; OLIVEIRA *et al.*, 2012; SANTOS *et al.*, 2020). Among the other fins, the formation of the caudal fin is observed in the flexion stage. Then, in the postflexion stage, the formation of the rays of the dorsal and anal fins occurs almost simultaneously. The formation of the pelvic and pectoral rays is likely to occur at the end of the postflexion stage (individuals larger than 11.26 mm SL). In *C. zebra* (PONTON & MÉRIGOUX, 2001), it is possible to verify a similar pattern in the development of the fins, since in the largest individual analyzed (14.3 mm) the pectoral fin still did not have any rays, despite the other fins already being formed. Another similar characteristic between these two species was the relatively large remnant finfold in front of the dorsal fin, which is only absorbed at the end of the larval development. This characteristic may be exclusive to this genus and, therefore, could be used as a character for its identification. However, as little is known about the early development of *Characidium* species, this characteristic must always be associated with others presented in this description.

The total or partial number of myomeres is also considered an important character in the identification of fish larvae (SNYDER, 1979; KELSO *et al.*, 2012; SANTOS *et al.*, 2020; CAJADO *et al.*, 2021). Besides, partial myomere counts can also be used as reference points to locate other structures, such as the position of the anal opening or the origin of the fins (BERRY & RICHARDS, 1973). *Characidium orientale* showed a wide variation in the number of preanal and postanal myomeres between the pre and postflexion stages (from 18 to 21 in preflexion to 23 in postflexion and also the postanal, from 13 to 16 in preflexion to 11 in postflexion). SEVERI & VERANI (2006) mention that, during the larval metamorphosis of Clupeiformes species, the position of the dorsal fin, anal opening, and origin of the anal fin moves anteriorly about to the length and the myomeres

number. Such characteristics are opposite to what was observed for *C. orientale* larvae, which presented a posterior movement of the anal fin as well as the anal opening. In fish, this transition is usually associated with changes in eating behavior (LOY *et al.*, 1998).

Allometric growth during the initial development is considered an adaptive response to reduce environmental pressures, increasing the likelihood of survival and growth (FUIMAN, 1983; OSSE & VAN DEN BOOGAART, 1999). The same authors suggest this form of growth produces changes in the shape of the body due to the differential growth of the organs and systems involved in primary functions (that is, food, breathing, and locomotion), rather than organs with a lower priority for survival. These changes are associated with morphological, physiological, ecological, or behavioral changes of the species (KOVÁČ *et al.*, 1999). In the initial development of *C. orientale*, it is possible to verify that for the morphometric variables related to the head (SnL and ED) and standard length (HD, BD, and SnP) presented differentiated growth (abruptly or gradually), indicating a greater metamorphosis, coinciding with the preflexion stage, distinctly to other Characiformes which show major changes between flexion and postflexion (BIALETZKI *et al.*, 2008; TAGUTI *et al.*, 2009; SANTOS *et al.*, 2020; CAJADO *et al.*, 2021). Thus, it is possible to suggest that during this stage, *C. orientale* larvae undergo food modifications or even begin exploring new habitats. However, as the number of individuals analyzed in the flexion and postflexion stages was small in relation to the preflexion stage, this result can be seen with parsimony, being considered only a growth trend, allowing a superficial but important approach, of morphological changes during the initial development of the species.

Therefore, *C. orientale* has altricial larvae and, consequently, indirect development, presenting important meristic, morphological and morphometric changes during its initial ontogeny. Their larvae are characterized by having an elongated and fusiform body, subterminal mouth, long intestine with an anal opening located posterior to the vertical line that passes over the median region of the body, concentrated pigments on the top of the head where two chromatophores stand out, as well as a lateral longitudinal strip. Also, they have a conspicuous remnant of the finfold in front of the dorsal fin, which seems to be characteristic of species of the genus during larval development. The total number of myomeres varies from 32 to 35 (18-23 pre and 11-16 postanal). The main changes in morphometry (metamorphosis) occur during the preflexion stage and suggest physiological and behavioral changes.

Rarely all stages of development of the species are found in field surveys, as these organisms have different environmental requirements, requiring a large sampling effort (CABACINHA *et al.*, 2019; REYNALTE-TATAJE *et al.*, 2020). Furthermore, our study, despite the small number of individuals, covers all the early development, and is also the first study addressing a species of the Crenuchidae family in Brazil (REYNALTE-TATAJE *et al.*, 2020), thus contributing

to the knowledge of the early development of Neotropical species. Another highlight is that this study is a pioneer in the description of the early development of a species of fish from the Taquari-Antas River, creating possibilities for future taxonomic and ecological studies of the ichthyoplankton community.

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