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A record of pughead deformity in *Satanoperca jurupari* (Heckel, 1840) (Teleostei: Cichlidae) in the Lower Rio Tocantins Basin

Um registro de deformidade craniana ("pughead") em *Satanoperca jurupari* (Heckel, 1840) (Teleostei: Cichlidae) na Bacia do Baixo Rio Tocantins

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Abstract: The pughead condition is a cranial deformity characterized by a shortened upper jaw relative to the lower jaw. In this study, we report the occurrence of this deformity in a single specimen of the Demon Eartheater, *Satanoperca jurupari*, collected in the Lower Rio Tocantins Basin. The most affected anatomical regions include the upper jaw, suspensorium, and anterior portion of the neurocranium. Although pollution from agrochemicals used in palm monoculture near the collection site is a plausible contributing factor, no direct evidence currently confirms this as the cause. This report represents the first documented case of the pughead condition in *Satanoperca* and the first record of this deformity in a species from rivers of the Brazilian Shield.

Keywords: fish deformity; Papa-terra; benthic-pelagic species; Amazon; Teratology.

Resumo: A condição pughead é uma deformidade craniana caracterizada pelo encurtamento da mandíbula superior em relação à mandíbula inferior. Neste estudo, relatamos a ocorrência dessa deformidade em um exemplar de Papa-terra, *Satanoperca jurupari*, coletado no Baixo rio Tocantins. As regiões anatômicas mais afetadas incluem a mandíbula superior, o suspensório e a porção anterior do neurocrânio. Embora a poluição por agroquímicos utilizados no cultivo de palma próxima ao local de coleta seja uma possível causa contribuinte, não há evidências diretas que confirmem essa hipótese. Este é o primeiro registro da condição "pughead" em *Satanoperca* e o primeiro relato dessa deformidade em uma espécie de rios do Escudo Brasileiro.

Palavras-chave: deformação em peixes; Papa-terra; espécie bento-pelágica; Amazônia; Teratologia.

Different types of deformities have been reported in various fish species, both wild and farmed (Dawson, 1964; Tandel et al., 2025), affecting various anatomical structures such as the jaw, fins, and spines (Catelani et al., 2017). Among cichlids, one of the most frequently observed conditions is the so-called pughead deformity (Näslund & Jawad, 2022), characterized by a shortening of the upper jaw relative to the lower jaw (Bortone, 1972). Although the exact causes of this deformity remain unclear, several studies have associated its occurrence with environmental factors, particularly those linked to human activities. These include the influence of hydroelectric power plants (Catelani et al., 2017), contamination by heavy metals (Kathan et al., 2020), elevated water temperatures (Georgakopoulou et al., 2010), and the presence of chemical pollutants such as polychlorinated biphenyls (PCBs) and hydrocarbons (Incardona et al., 2004; Sun et al., 2009). Contaminated soils containing organometallic compounds and other industrial pollutants have also been implicated (Jawad & Ibrahim, 2017). In addition to environmental influences, genetic factors such as reduced genetic diversity and high inbreeding coefficients may contribute to the development of pughead deformities (Ugbomeh et al., 2022). Nonetheless, the causal relationship between these factors and the occurrence of such deformities remains inconclusive.

Here, we report the occurrence of pughead deformity in a single specimen of *Satanoperca jurupari* (Cichliformes: Cichlidae) (Figure 1), collected during a field expedition on June 26th, 2023, from a small

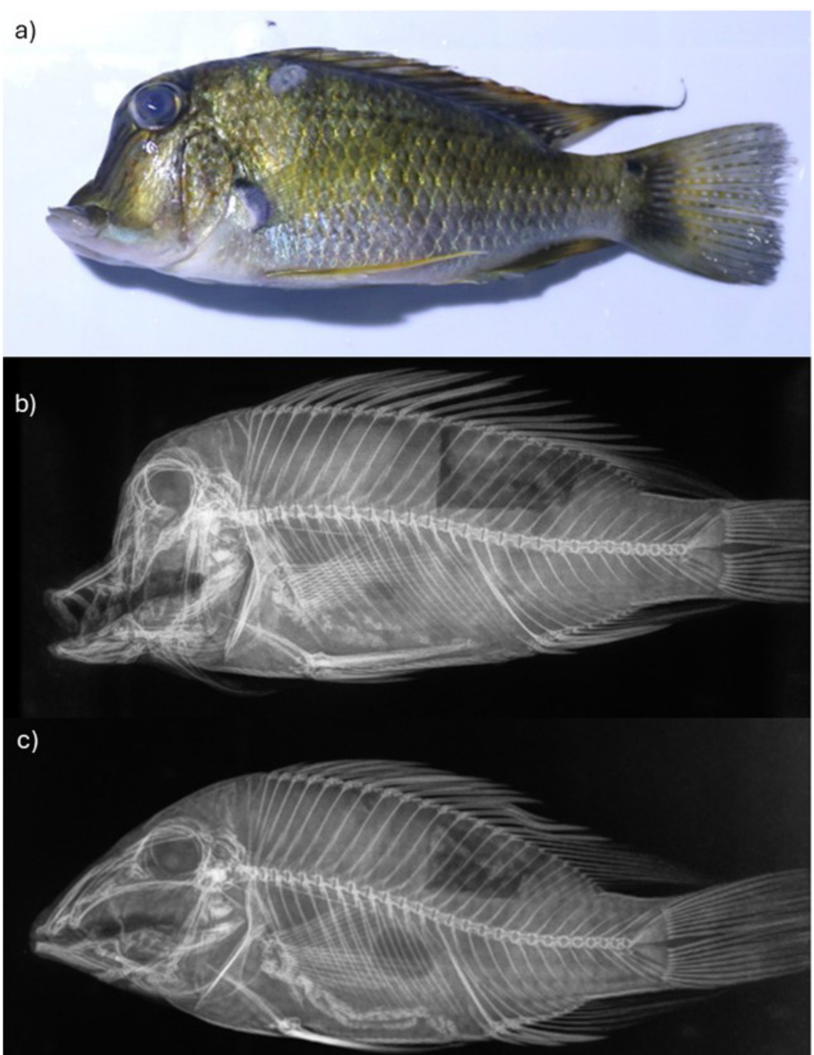

Figure 1. (a) Left lateral view of the pughead *Satanoperca jurupari* specimen collected in the Lower Tocantins River basin, in the municipality of Baião, Pará state, Brazil. (b) X-ray image of the head of the pughead specimen (c) and a normally developed specimen (b, 300 mm SL) of *Satanoperca jupari* in left lateral view.

stream adjacent to the main channel of the Lower Tocantins River. A normally developed *S. jurupari* specimen was also collected at the same site. The sampling location (2°45'51.0"S, 49°40'34.9"W) is situated in the municipality of Baião, within the Cametá microregion in the Northeast Pará state mesoregion (IBGE, 2022). Following collection, both specimens were euthanized on ice, and fixed in a 10% formalin solution, and subsequently transferred to 75% ethanol for long-term preservation. The specimens were deposited in the ichthyological collection of the Museu Paraense Emílio Goeldi (MPEG). Collection and transportation were carried out under permit SISBIO 70940-1, issued by the Brazilian competent environmental authority.

The deformed specimen exhibited secondary "pugheadedness" (Hickey et al., 1977), characterized by hypoplasia of the forehead with a typical ventrally-oriented arch right above the eyes (Figure 1). Like X-ray images, three-dimensional reconstructions obtained using 3D Slicer (Fedorov et al., 2012) from nano-computed tomography (nano-CT) scans allowed us to visualize cranial structures and compare the specimen exhibiting pughead features (Figure 2a) with anatomically normal specimens (Figure 2b). In the neurocranium of the affected *S. jurupari*, a malformation of the frontal bone characterized by a steep, bulging forehead can be observed (Figure 2b). Also, the pughead specimen had a shortening of the premaxillary bone, resulting in a prognathic appearance. The lower jaw and branchial arches seemed mostly unaffected. The shape and arrangement of the premaxilla and maxilla were altered from the normal condition, such that the posterior tip of the upper jaw seemed to be curved downwards.

In addition to a visual description of the deformity, we conducted a morphological analysis using both classical and geometric morphometry to assess the disparity between the deformed individual and other individuals exhibiting normal morphological characteristics. The two collected specimens, along with 12 additional specimens from the ichthyological collection of MPEG, were included to represent potential interspecific morphological variation (Table 1). To minimize the influence of the allometry, we selected individuals of similar size, all originating from the same drainage. For the classical morphometric analysis, six linear measurements were obtained using a digital caliper (accuracy: 0.1 mm). The following measurements were recorded for all specimens: standard length (SL), orbit height (OH), orbit length (OL), head

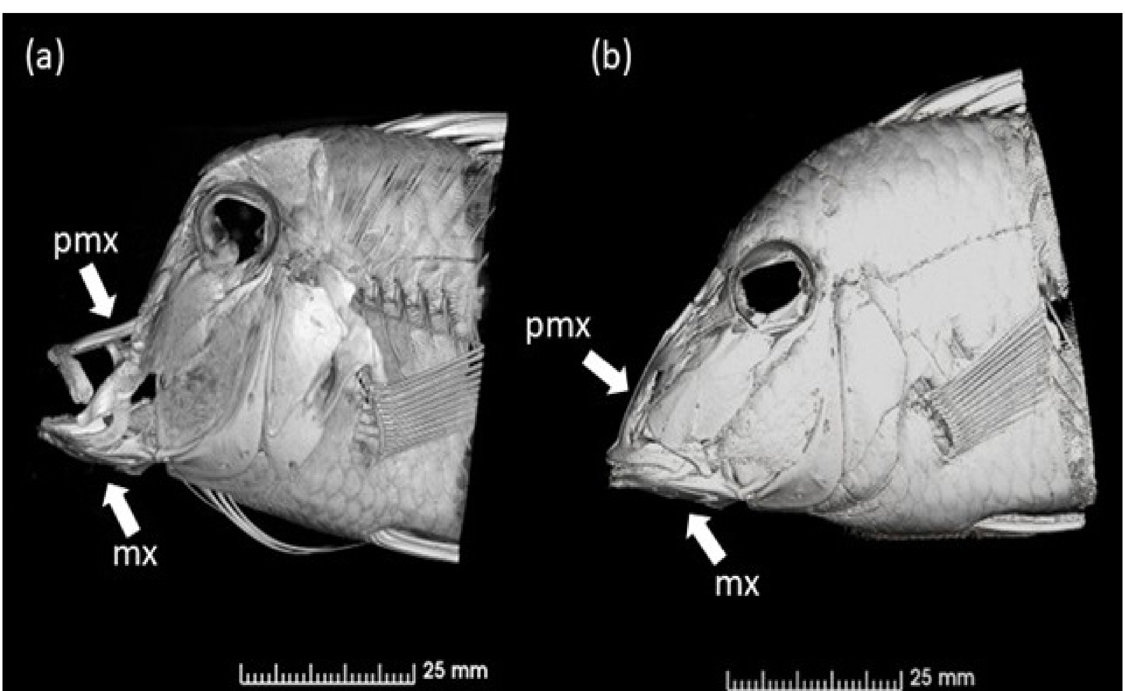

Figure 2. Image from nano-computed tomography (nano-CT) scans allowed us to visualize cranial structures and compare the specimen exhibiting pughead features. (a) pughead specimen (b) and a normally developed specimen. Arrows points out craniofacial structures: mx - maxilla (posterior tip of upper jaw); pmx - premaxilla (upper jaw).

Table 1. Specimens in the morphological analysis are listed, with ID referring to lots of fish at MPEG. Sampled localities and coordinates are included.

Specimen	ID	LS (mm)	LH (mm)	HO (mm)	LO (mm)	HH (mm)	LAT	LONG	Locality/ Municipality
Sp01_n		128.8	47.5	10.8	10.5	50.3			
Sp02_n		135.3	47.7	13.0	12.4	50.4			
Sp03_n	MPEG37367	123.7	46.3	12.1	10.9	49.6	4°22'10.64" S	49° 39'58.92" W	
Sp04_n		130.2	49.0	10.1	11.7	49.9			
Sp05_n		117.5	41.4	10.8	11.2	44.6			Novo Repartimento
Sp06_n		138.4	52.4	11.8	12.9	56.2			
Sp07_n	MPEG37360	135.1	50.1	12.4	12.6	54.8	4°22'10.64"S	49°39'58.92"W	
Sp08_n		124.9	46.3	9.8	10.3	50.7			
Sp09_n		121.6	44.3	10.2	10.0	51.1			
Sp10_n	MPEG38451	99.5	37.4	9.4	9.5	36.2	3°4'17.68"S	49°38'57.79"W	Baião
Sp11_n	MPEG37412	141.7	51.2	11.2	11.0	53.9	3°52'27.32"S	49°45'30.96"W	Tucuruí
Sp12_n	MPEG37371	126.9	47.3	11.5	11.7	49.8	2°47'56.53"S	49°45'22.46"W	
Sp13_n	MPEG40000	133.4	48.4	10.0	10.2	52.1	2°45'51.3"S	49°40'35.1"W	Baião
Sp14_p		122.2	43.4	10.4	10.7	51.4			

Codes: SL: standard length; LH: head length; HO: orbit height; LO: orbit length; HH: head height. The final letter in each specimen ID indicates whether the individual was morphologically normal ('n') or exhibited the pughead deformity ('p').

height (HH), and head length (HL) (Figure 3a). To ensure consistency in comparisons between the pugheaded specimen and those with normal morphology, HH was measured at the level of the upper operculum joint in the skull.

For the geometric morphometric analysis, nine fixed landmarks and seven semi-landmarks were selected based on Gilbert et al. (2020), with minor modifications. Specifically, we excluded landmarks deemed irrelevant to the objective of this our study and adjusted the positioning of the semi-landmarks to enhance the model's ability to capture and visualize the cranial deformation characteristic of pugheadedness (Figure 3b and 3c).

In the laboratory, the left lateral surface of each specimen was photographed in a photo table using a professional camera (Canon EOS 70D) mounted on a fixed support, with a standardized angle of 90º and adjustable height. Each photograph included a scale bar to facilitate subsequent dimensional calibration of the reference data. To account for potential allometric effects, a Generalized Procrustes Analysis (GPA) was performed using the software tpsDig (Rohlf, 2016). GPA aligns landmark configurations from multiple specimens through least squares estimations for translation and rotation (Clabaut et al., 2007). For linear morphometric data, linear regression was used to account for potential allometric effects, with each linear trait as the dependent variable and standard length as the predictor. The resulting residuals were then subjected to an Analysis of Variance (ANOVA) to

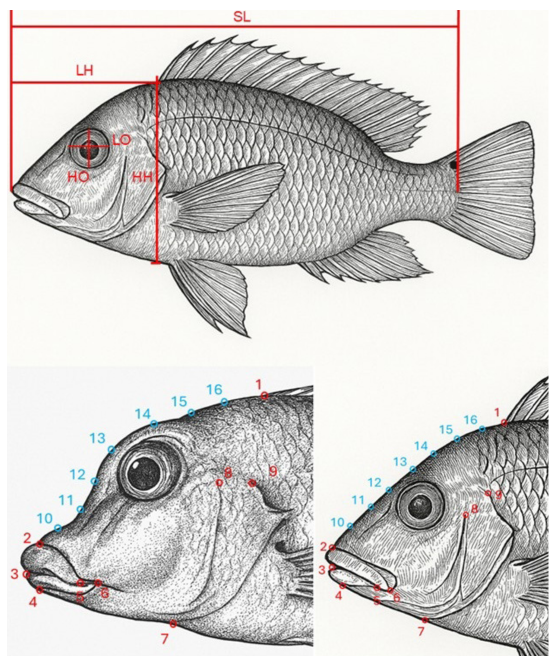

Figure 3. (a) Anatomical landmarks used for linear morphometric analysis. SL: standard length, HO: orbit height, LO: orbit length, HH: head height, LH: head length. Figures (b) and (c) present the anatomical landmarcks delimited used for geometric morphometric analysis: open red circles indicate fixed landmarks; blue circles represent semi-landmarks. Fixed landmarks include: (1) dorsal fin insertion; (2) premaxillary groove; (3) lower jaw insertion; (4) anterior tip of the snout; (5) premaxillary tip; (6) posterior margin of the premaxilla; (7) base of the pectoral region; (8) preoperculum; (9) posterior edge of the operculum. Semi-landmarks (10-16) consist of seven equidistant points outlining the contour of the forehead. Illustrations hand-drawn by Lais Lobato.

test significant differences between the pugheaded individual and fully developed specimens.

To visualize the data, we performed a Principal Component Analysis (PCA) on the Procrustes residuals (with allometry corrected). The first two principal components were used to project the region of morphospace occupied by each specimen, allowing visual assessment of individual overlaps (Gilbert et al., 2020) and highlighting morphological differences.

The first two PCA axes accounted for 74.72% of total shape variation (Figure 4). PC1 explained 59.4% and captured variation in the shape of the forehead and mouth. On the positive end, the forehead was more protruded and the mouth longer and narrower, whereas the negative end was characterized by a more undulating forehead and a broader, shorter mouth. PC2 accounted for an additional 15.3% variation, emphasizing differences in forehead and lip shape. Negative scores were associated with a jaw featuring thinner lips and a wavy forehead, while positive scores reflected a wider mouth and a more concave forehead shape. The negative scores on both axes were driven by the individual exhibiting the pughead deformity, highlighting its morphological divergence from the species' average.

The procrustes ANOVA showed no difference between the pughead individual and the normal specimens in relation to the size of the centroid ($F_{(1, 12)} = 0.67$, $p = 0.42$), however, in relation to the shape, there was a remarkable difference ($F_{(28,336)} = 5.28$, $p < 0.01$). In Figure 4, the morphology of the deformed individual is highlighted in dark blue for better visualization of the landmarks in contrast to the species mean, shown in light blue.

Cranial deformities such as the pughead phenotype have been documented in various fish species over the years (Näslund & Jawad, 2022), typically as isolated or rare occurrences. However, three additional studies have reported the presence of this malformation in Amazonian Neotropical cichlids introduced into river systems in southern Brazil, suggesting that such deformities may be more prevalent in non-native populations under certain ecological or genetic conditions (Catelani et al., 2017; Oliveira et al., 2021, 2022). This cranial malformation may result from

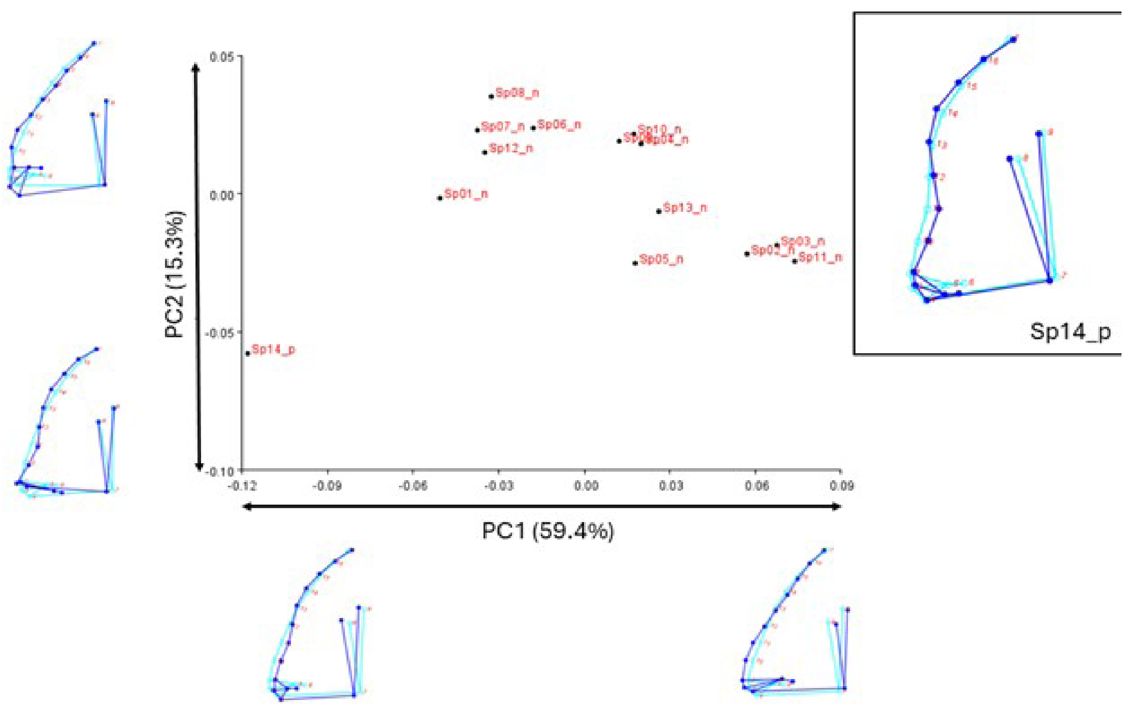

Figure 4. Graphical representation of the principal component analysis (PCA) for 14 individuals of *Satanoperca jurupari*. The X-axis represents PC1 (59.4%) and the Y-axis (left) represents PC2 (15.3%), illustrating the distribution of individuals across the morphospace. Each point corresponds to an individual, and the occupied regions reflect morphological variation. The morphological disparity test revealed no significant difference between the deformed specimen and the remaining individuals (p = 0.42). We used the specimen column and defined the final letter of everyone to indicate which specimen was normal (n) or which presented the pughead deformity (p). Morphological profile of the pughead individual (dark blue) compared to the mean profile of the remaining individuals (light blue).

inbreeding among closely related individuals, particularly in populations derived from introduced specimens, which often retain only a fraction of the species' original genetic diversity. This reduction in genetic variability can increase the likelihood of anatomical abnormalities (Catelani et al., 2017; Langen et al., 2011). Additionally, anthropogenic impacts, especially water pollution caused by the discharge of pesticides and other agrochemicals into aquatic ecosystems, have also been suggested as potential contributing factors (Oliveira et al., 2021).

The biological implications of the pughead deformity in cichlid species suggests that this cranial anomaly may not have a negatively effect on feeding efficiency and overall fitness, as most recorded specimens with this condition have been captured at mature stages (Näslund & Jawad, 2022). However, modifications in jaw structure can hinder prey capture or food manipulation, whereas changes in skull morphology may affect hydrodynamics and swimming performance (Bortone, 1972; Lijalad & Powell, 2009). Given the ecological adaptability and benthic feeding strategy of *S. jurupari*, it is likely that the deformity did not significantly impair the substrate shifting efficiency of the affected specimen. Statistical analysis supports that the pughead condition in *S. jurupari* represent a distinct cranial shape abnormality, unrelated to body size, emphasizing the importance of assessing shape variation independently of size in morphological studies.

Considering the area where the pughead *S. jurupari* specimen was collected, a major part of the original land cover has been replaced by Palm oil crops (*Elaeis guineensis*). Glyphosate-based herbicides and endosulfan insecticides are commonly utilized in this monoculture. Such agrochemicals are known to potentially cause developmental disorders in fishes, especially during early developmental stages, by altering embryonic development (Lanzarin et al., 2023; Lopes et al., 2022). Since agrochemicals have been used in the region, it is plausible that the pughead deformity observed in the collected specimen may be associated with pollution from these substances. Nonetheless, it must be emphasized that, currently, there is no direct evidence to substantiate this as the definitive cause.

As discussed in previous studies (e.g., Slooff, 1982; Boglione et al., 2013; Näslund & Jawad, 2022), the causes of pugheadedness can be multifactorial and difficult to determine definitively, although human impacts may be among the possible contributing factors. Further studies, including experimental ones, are needed to investigate the potential triggers of this head deformity and to ascertain whether environmental impacts resulting from anthropic activities, such as agrochemical pollution, play a role. Besides, so far only a single fish specimen with this deformity has been recorded in the area. The record of additional individuals with this or other deformities, especially from other fish species occurring in the area, could support the hypothesis that these deformities are caused by environmental factors resulting from anthropic activities, such as water pollution.

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Data availability

The entire data set supporting the results of this study has been published in the article itself.

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