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FIRST RECORD OF THE CRITICALLY ENDANGERED KILLIFISH *Austrolebias univentripinnis* COSTA & CHEFFE, 2005 (CYPRINODONTIFORMES, RIVULIDAE) FOR URUGUAY

*Primer registro del pez killi en peligro crítico Austrolebias univentripinnis Costa y Cheffe, 2005
(Cyprinodontiformes, Rivulidae) para Uruguay*

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Resumen. El análisis de especímenes capturados en una reciente expedición a la cuenca baja del Río Yaguarón (noreste uruguayo), reveló la presencia del pez anual críticamente amenazado *Austrolebias univentripinnis* Costa y Cheffe, 2005, representando el primer registro de esta especie para el país. Se discuten las diferencias morfométricas observadas respecto a poblaciones brasileñas. Proponemos sea tratada como una especie amenazada para Uruguay considerando su distribución restringida, una única charca estacional de aproximadamente 800 m².

Palabras clave. *Cynolebiatini*, cuenca de la Laguna Merín, pez anual, prioridad de conservación.

Abstract. The examination of specimens collected during a recent expedition to the lower Yaguarón river basin (northeastern Uruguay), revealed the presence of the critically endangered killifish *Austrolebias univentripinnis* Costa and Cheffe, 2005, which represents the first record of this species for the country. Morphometric differences with Brazilian populations are discussed. We propose it as an endangered species for Uruguay considering its restricted distribution, found so far only a single seasonal pond of about 800 m².

Key words. *Cynolebiatini*, Merín Lagoon basin, anual killifish, conservation priority.

INTRODUCTION

The Neotropical killifish genus *Austrolebias* Costa, 1998 comprises 51 valid species, distributed in Bolivia, Paraguay, southern Brazil, northeast Argentina, and Uruguay, in the La Plata, Patos-Merín, and southwestern Amazon basins (Alonso *et al.*, 2018; Calviño *et al.*, 2016; Costa, 2006, 2014; Costa *et al.*, 2017b; Fricke *et al.*, 2021; García *et al.*, 2012; Lanés *et al.*, 2021; Loureiro *et al.*, 2011; Nielsen and Pillet, 2015; Serra and Loureiro, 2018; Volcan *et al.*, 2014, 2017, 2021; Volcan and Severo-Neto, 2019). Species of the genus are characterized by the following synapomorphies: absence of scales between the corner of mouth and anterior portion of preopercular region and ventral portion of opercular region, a deep urohyal, reduced ventral process of the angulo-articular, presence of a greyish to blackish infraorbital bar and supraorbital spot, and males with rounded dorsal fin and elongated urogenital papilla (Costa, 2006; Lanés *et al.*, 2021). The monophyly of *Austrolebias* is well supported by morphological and molecular analyses, but subgeneric level relationships are still conflictive (Costa, 2006, 2010; Costa *et al.*, 2017a; Loureiro *et al.*, 2018, Volcan *et al.*, 2021).

For the Uruguayan territory 20 currently valid species have been recorded since 1948 (*Cynolebias bellottii*, de Buen, 1948, 1950; Litz and Koerber, 2014; Koerber *et al.*, 2021), with representatives from all the major clades recognized by Loureiro *et al.* (2018), except from the "*wichi-varzeae*" clade (see Table 1) (Costa, 2006; Fricke *et al.*, 2021; Loureiro and García, 2008; Loureiro *et al.*, 2011, 2018; Serra and Loureiro, 2018). In the present work we record the fourth representative of the subgenus *Austrolebias* for Uruguay.

MATERIALS AND METHODS

The examined specimens were collected with hand nets, euthanized by overdose in eugenol solution, fixed in 4 % formalin solution, preserved in 70 % ethanol after successive rinsing in water to eliminate formalin, and housed in the collections of the Museo Nacional de Historia Natural de Montevideo (MHNM), Montevideo, Uruguay and the Instituto de Bio y Geociencias del NOA (IBIGEIO-I), Rosario de Lerma, Salta, Argentina. Measurements and counts follow Costa (1995, 2006). Measurements are presented as percentages of standard length (SL) except for those related to the head, which are expressed as percentages of head length (HL). Identification was based on Costa and Cheffe (2005), Costa (2006) and Serra and Loureiro (2018), using external characters. The distributions mapped from previous records of the subgenus *Austrolebias* from Uruguay and surrounding areas are based on Costa and Cheffe (2005), Costa (2006) and Serra and Loureiro (2018).

RESULTS AND DISCUSSION

Austrolebias univentripinnis Costa and Cheffe, 2005

Examined material: **URUGUAY: Cerro Largo: IBIGEIO-I 472**, 1 male 40.4 mm SL and 2 females 31.7-32.6, south of Poblado Uruguay (32°31'S 53°28'W), coll. W.S. Serra, M. Torres and G. Furtado, 10/VII/2021. **MHNM 4626**, 2 males 31.8-33.9 mm SL and 4 females 26.0-30.4 mm SL, south of Poblado Uruguay (32°31'S 53°28'W), coll. W.S. Serra, M. Torres and G. Furtado, 10/VII/2021. **MHNM 4627**, 15 males 27.9-44.9 mm SL and 32 females 28.2-35.8 mm SL, south of Poblado Uruguay (32°31'S 53°28'W), coll. W.S. Serra, M. Torres and G. Furtado, 10/VII/2021.

Table 1 - Valid *Austrolebias* species recorded from Uruguay ordered by subgenus according to Loureiro *et al.* (2018).Subgenus *Acantholebias* Costa, 2008*A. luteoflammulatus* (Vaz-Ferreira, Sierra de Soriano and Scaglia de Paulete, 1965)*A. quirogai* Loureiro, Duarte and Zarucki, 2011Subgenus *Acrolebias* Costa, 2008*A. arachan* Loureiro, Azpelicueta and García, 2004*A. charrua* Costa and Cheffe, 2001*A. reicherti* (Loureiro and García, 2004)*A. viarius* (Vaz-Ferreira, Sierra de Soriano and Scaglia de Paulete, 1965)Subgenus *Argolebias* Costa, 2008*A. nigripinnis* (Regan, 1912)Subgenus *Austrolebias* Costa, 1998*A. bellottii* (Steindachner, 1881)*A. melanoorus* (Amato, 1986)*A. queguay* Serra and Loureiro, 2018Subgenus *Cypholebias* Costa, 2008*A. cinereus* (Amato, 1986)Subgenus *Gymnolebias* Costa, 2008*A. gymnoventris* (Amato, 1986)Subgenus *Megalebias* Costa, 1998*A. cheradophilus* (Vaz-Ferreira, Sierra de Soriano and Scaglia de Paulete, 1965)*A. elongatus* (Steindachner, 1881)*A. prognathus* (Amato, 1986)*A. wolterstorffi* (Ahl, 1924)

Unnamed subgenus

A. affinis (Amato, 1986)*A. alexandri* (Castello and López, 1974)*A. juanlangi* Costa, Cheffe, Salvia and Litz, 2006*A. periodicus* (Costa, 1999)

The examined specimens from the new population herein reported differ from the other species of the genus, except for *A. univentripinnis*, by the following combination of characters: males with urogenital papillae attached to the first anal fin ray, pelvic fins fused, body flanks with vertical rows of iridescent blue-green spots on a dark greenish gray background often forming lines, presence of a single anterior rostral neuromast, low body depth ($\leq 39.8\%SL$) and short anal fin base ($\leq 46.4\%SL$) (Costa and Cheffe, 2005; Costa, 2006; Serra and Loureiro, 2018). Females with nearly triangular anal fin, pelvic fins fused, dorsal fin origin anterior to the anal fin origin, presence of a single anterior rostral neuromast, absence of multiple scale rows on anal fin base, absence of rounded black spots on caudal fin base and caudal peduncle, absence of conspicuous midlateral black spots on body (one or two minute spots, if present) and dorsal profile of body slightly convex (Costa and Cheffe, 2005; Costa, 2006; Serra and Loureiro, 2018).

Morphometric measurements of *A. univentripinnis* from Uruguay (Table 2) partially match those already reported, with the following exceptions: all ranges of measurements presented as percentages of standard length have lower values than those reported by Costa and Cheffe (2005) and Costa (2006), for Brazilian populations, both in males and females, and the following variables presenting not-overlapping ranges: body depth, head length, pre pelvic-fin length and caudal fin length in males and head length, caudal peduncle depth and caudal fin length in females. Measurements presented as percentages of head length have similar ranges and intervals with values reported by Costa and Cheffe (2005) and Costa (2006). Medial pelvic-fin membranes 0–90% coalesced, usually 30% (75–90% in Costa, 2006).

Meristics present wider and overlapping ranges in comparison to the previously reported by Costa and Cheffe (2005) and Costa (2006): dorsal fin rays 23–26 in males



Figure 1 - Male of *Austrolebias univentripinnis* immediately after capture: MHNM 4627, 44.2 mm SL, south of Poblado Uruguay, approx. 32°31'S 53°28'W, Cerro Largo, Uruguay.

and 17–22 in females; anal-fin rays 24–27 in males and 21–25 in females. Caudal fin rays 19–23; pectoral fin rays 11–13; pelvic fin rays 4–6. Scales cycloid. Longitudinal series of scales 28–31, regularly arranged; transversal series 11–16; circumpeduncular series 14–17. Scales on anal-fin base usually absent in females and few irregularly arranged in one row in males. Contact organs present in all analyzed males, 1 to 5 contact organs per scale (usually 2 or 3); contact organs present in four uppermost rays of pectoral fins; no contact organs on unpaired and pelvic fins. Cephalic neuromasts: supraorbital 15–23, parietal 1–3, anterior rostral 1 (0 in one individual of 50), posterior rostral 1–2 (usually 1), infraorbital 0–4 (usually 2–3) + 22–35, preorbital 0–3, otic 1–4, post-otic 2–5, supratemporal 1–2 (usually 1), median opercular 0–1 (usually 1), ventral opercular

1–4, preopercular 18–31, mandibular 12–20, lateral mandibular 2–6.

Coloration pattern of female and male specimens (Figures 1 to 3) are highly variable but coincident with the described in previous works (Costa and Cheffe, 2005; Costa, 2006; Volcan *et al.*, 2010).

We tentatively attribute the differences observed in relation to the previous works (Costa and Cheffe, 2005; Costa, 2006) concerning the fusion degree of pelvic fins and body proportions to the large size and associated allometric effect of the examined individuals, with 28.2–44.9 (N=15, mean=38.4) mm SL vs. 29.9–35.6 (N=7, mean=32.8) mm SL in males and 29.3–35.8 (N=34, mean=33.3) mm SL vs. 25.5–32.2 (N=7, mean=28.5) mm SL in females. Wider variations in count ranges reported in our work are certainly a consequence of the

Table 2 - Morphometric data of *Austrolebias univentripinnis* from Uruguay, Standard Length is expressed in mm; measurement numbered 2–13 are percent of standard length; subunits of head (numbered 14–18) are percents of Head Length.

Character	Males					Females				
	N	Range		Mean	SD	N	Range		Mean	SD
		Low	High				Low	High		
1. Standard length (mm)	15	28.2	44.9	38.4	-	34	28.2	35.8	32.4	-
2. Body depth	15	32.1	35.7	33.7	1.18	34	29.3	37.3	33.3	1.85
3. Head length	15	23.8	28.0	26.1	1.12	34	23.4	29.2	26.2	1.03
4. Caudal peduncle depth	15	10.3	12.3	11.2	0.50	34	10.1	12.5	11.2	0.59
5. Caudal peduncle length	15	10.0	14.1	12.2	1.20	34	14.1	22.3	18.2	2.03
6. Pre dorsal length	15	45.8	51.2	48.3	1.54	34	54.8	61.8	58.2	1.85
7. Dorsal-fin base length	15	38.6	44.2	40.4	1.57	34	21.9	31.3	26.4	2.18
8. Pre anal-fin length	15	47.5	51.6	49.9	1.17	34	54.3	62.8	58.6	1.57
9. Anal-fin base length	15	38.0	42.8	40.3	1.41	34	21.9	28.2	25.4	1.60
10. Pre pelvic-fin length	15	41.3	45.9	43.3	1.08	34	46.7	52.8	49.0	1.37
11. Pectoral fin length	15	17.9	23.0	20.6	1.57	34	19.4	24.9	22.3	1.20
12. Pelvic fin length	15	6.3	8.2	7.4	0.56	34	7.6	13.7	11.0	1.30
13. Caudal fin length	15	17.5	23.4	20.4	1.87	34	21.8	26.9	24.1	1.11
14. Head width	15	63.6	75.1	69.6	3.91	34	68.1	85.2	73.1	3.64
15. Head depth	15	97.1	116.4	109.9	5.66	34	99.5	114.2	106.8	3.87
16. Interorbital width	15	44.7	50.2	47.2	1.72	34	41.6	51.8	46.7	2.35
17. Eye diameter	15	28.3	32.5	30.6	1.38	34	29.7	36.1	32.5	1.55
18. Snout length	15	11.4	16.0	14.1	1.23	34	11.8	18.0	14.5	1.68

large number of examined individuals, 15-16 males (vs. 3-7 in original description) and 34 females (vs. 3-7 in original description).

Austrolebias univentripinnis is recorded for Uruguay from a single locality, a pond south of Poblado Uruguay near the Yaguarón River (Figure 4), being the sixth known population of the species. The other five populations are from Município de Jaguarão in Rio Grande do Sul (Volcan *et al.*, 2015), close to the Uruguayan locality. The Uruguayan pond contained this single species, had an area of about 800 m² and present dense submerged vegetation (Figure 5). The other killifish species recorded for the area (twelve sampled ponds in 94 ha) was *Austrolebias reicherti* (Loureiro and García, 2004) from only one pond.

According to the criteria defined by Soutullo *et al.* (2013), we propose that *A. univentripinnis* must be considered as a

priority for conservation and classified as an endangered species for the species list of Sistema Nacional de Áreas Protegidas (SNAP) of Uruguay: its global distribution occupies less than 200.000 km² (Criteria 1), its occurrence area represents less than 10% of the national territory (Criteria 4), is an ecologically singular species (Criteria 7) and has economic value (Criteria 8). Reinforcing this, the species had been previously classified as Critically Endangered for Brazil because of its small Area of Occupancy (less than 10 km²) and the impact of rice cultivation, cattle farming and urbanization on its known populations (Volcan *et al.*, 2010, 2015; ICMBio, 2018). The Uruguayan population does not extend significantly the species' area of distribution, being located about two kilometers south of the type locality. Productive activities in the property where the species was found

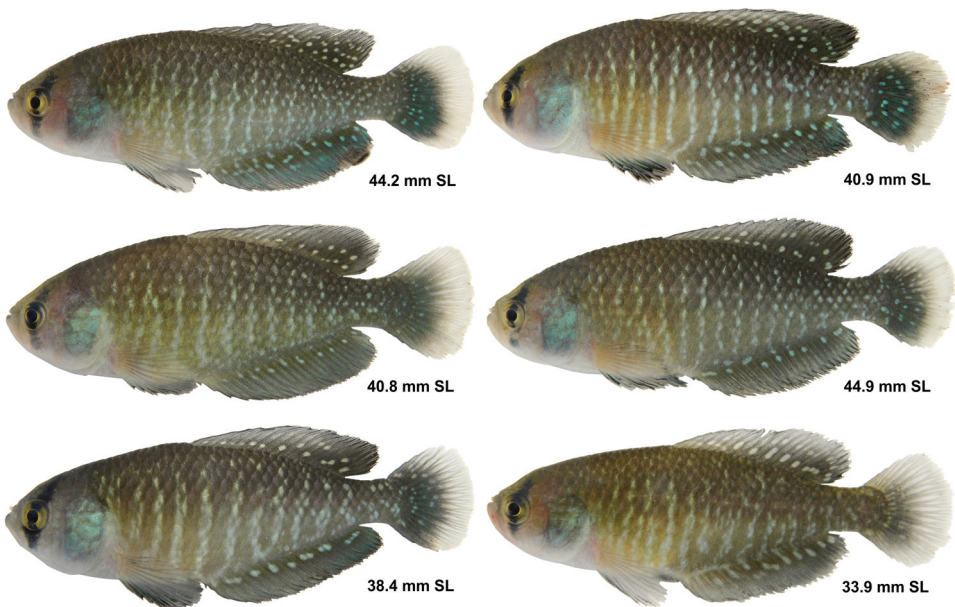


Figure 2 - Males of *Austrolebias univentripinnis* before fixation: MHN 4627, south of Poblado Uruguay, approx. 32°31'S 53°28'W, Cerro Largo, Uruguay.

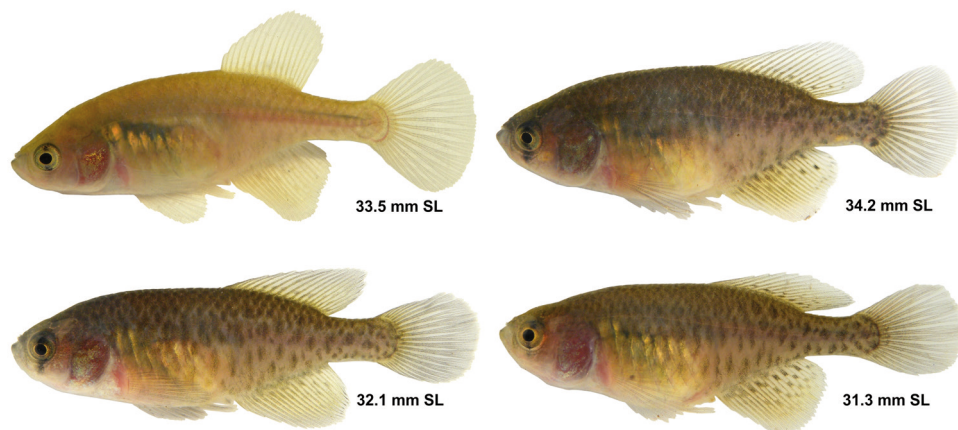


Figure 3 - Females of *Austrolebias univentrifinnis* before fixation: MHNM 4627, south of Poblado Uruguay, approx. 32°31'S 53°28'W, Cerro Largo, Uruguay.

seem not be a significant source of stress for the species (i.e., extensive cattle-farming), but rice production in the surrounding area is potentially a serious threat to this population. As suggested for Rio Grande do Sul populations (Volcan *et al.*, 2010, 2015; Lanés *et al.*, 2021), specific actions are needed to protect the site, such as creating a small private protected area, working with local people on conservation strategies, and lobbying with government agencies to promote legal protection of the species.

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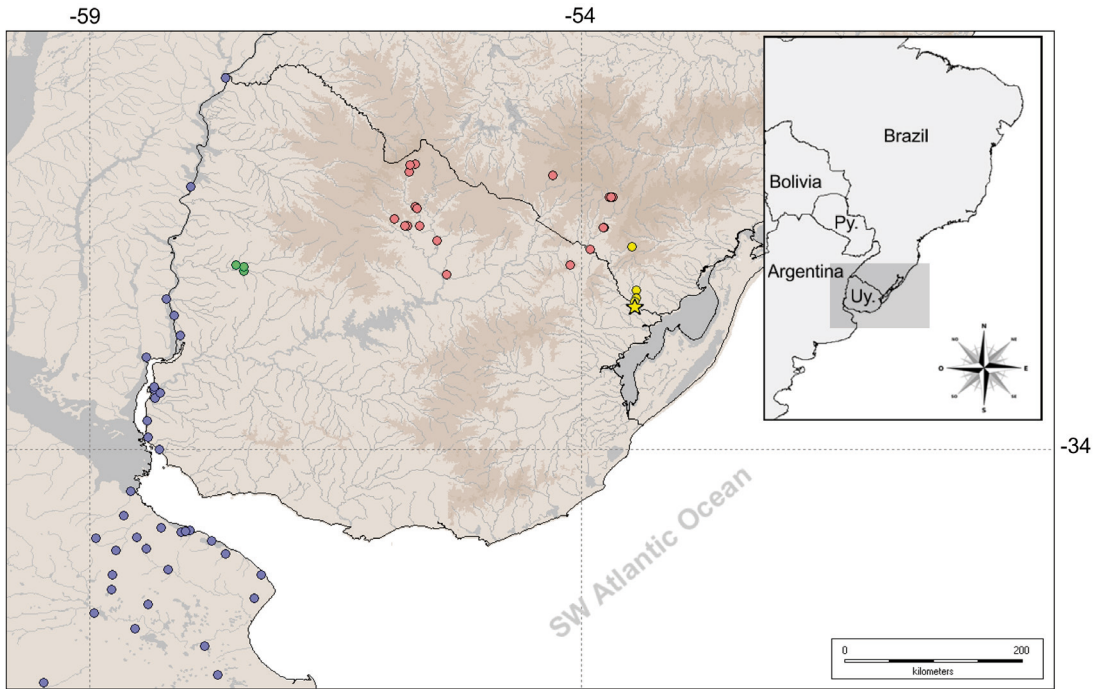


Figure 4 - Geographic distribution of *Austrolebias univentripinnis* (yellow dots) and the other species of subgenus *Austrolebias* recorded for Uruguay: *A. bellottii* (blue dots), *A. melanoorus* (red dots), and *A. queguay* (green dots). Yellow star indicates de new locality. Py. = Paraguay; Uy. = Uruguay.



Figure 5 - Temporary pond where *Austrolebias univentripinnis* was collected, south of Poblado Uruguay, approx. 32°31'S 53°28'W, Cerro Largo, Uruguay.

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