Description of a new species of annual fish of the genus *Neofundulus* (Cyprinodontiformes: Rivulidae) from the upper río Mamoré basin, Bolivia

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Abstract

Neofundulus splendidus n. sp. is described from Bolivia, departamento de Santa Cruz, río San Miguel, upper Mamoré basin. *Neofundulus splendidus* n. sp. is distinguished from the remaining *Neofundulus* species by a combination of characters: Large black spot at humeral region with diameter of approximately 1.5-2.0 of pupil in males and females (vs. black spot in humeral region, when present, small- half of pupil), 40-41 scales in longitudinal series (vs. 35-38 in remaining congeners).This is the first record for the genus *Neofundulus* in río Mamoré basin.

Resumo

Neofundulus splendidus n. sp. é desrcito da Bolívia, departamento de Santa Cruz, río San Miguel, alto bacia Mamoré. *Neofundulus splendidus* n. sp. destingue das demais espécies de *Neofundulus* pela seguintes combinações de características*:* Grande mancha preta na região de humeral com diâmetro de aproximadamente 1.5-2.0 do diâmetro da pupila em machos e fêmeas (vs. pequena ou ausente mancha preta na região de humeral), 40-41 escamas em séries longitudinais (vs. 35-38 nas demais espécies). Este é o primeiro registro para o gênero *Neofundulus* na bacia do rio Mamoré.

Zusammenfassung

Neofundulus splendidus sp. n. wird vom Fluss San Miguel aus dem Einzugsgebiet des Mamoré-Oberlaufs in Bolivien, Departamento Santa Cruz, beschrieben. Die neue Art unterscheidet sich von anderen Arten der Gattung *Neofundulus* durch eine Kombination folgender Merkmale: großer schwarzer Fleck im Humeralbereich mit etwa dem 1,5- bis 2,0-fachen Durchmesser der Pupille bei Männchen und Weibchen (im Gegensatz zu einem kleinen schwarzen Fleck im Humeralbereich, der – wenn überhaupt vorhanden – etwa dem halben Pupillendurchmesser entspricht), 40-41 Schuppen in Längsreihen (35-38 bei den übrigen Angehörigen der Gattung). Es handelt sich um einen Erstnachweis der Gattung *Neofundulus* im Rio-Marmoré-Einzugsgebiet.

Résumé

Neofundulus splendidus n. sp. est décrit, venant de Bolivie, Departamento de Santa Cruz, rio San Miguel, bassin supérieur du Mamoré. *Neofundulus splendidus* n. sp. se distingue des autres espèces de *Neofundulus* par une combinaison de caractéristiques: une grande tache noire dans la région humérale avec un diamètre d'environ 1,5 - 2,0 de la pupille pour les mâles et les femelles (contre une tache noire dans la région humérale, si elle est présente,, d'une petite moitié de la pupille), 40 - 41 écailles en séries longitudinales (contre 35 - 38 pour les autres congénères). Il s'agit de la première occurrence du genre *Neofundulus* dans le bassin du rio Mamoré.

Sommario

Neofundulus splendidus n. sp. è descritto dalla Bolivia, Departamento de Santa Cruz, Rio San Miguel, bacino superiore del Mamoré. *Neofundulus splendidus* n. sp. si distingue dalle altre specie di *Neofundulus* per una combinazione di caratteri: presenza sia in maschi sia in femmine di una grande macchia nera in regione omerale con diametro di circa 1.5-2.0 volte quello della pupilla (vs. macchia nera nella regione omerale, quando presente, piccola e di diametro metà della pupilla), 40-41 scaglie in serie longitudinale (vs. 35-38 nei congeneri). Il genere *Neofundulus* non era stato ancora segnalato nel bacino del Río Mamoré.

INTRODUCTION

Even though the first *Neofundulus* species *N. paraguayensis*, was described over a hundred years ago (Eigenmann & Kennedy, 1903), few species of the genus are so far known, when compared with other Rivulidae genera, and few studies have been conducted on this genus.

So far, only five *Neofundulus* species have been described: *N. paraguayensis* (Eigenmann & Kennedy, 1903) from the río Paraguay basin, *N.*

ornatipinnis Myers 1935 from the río Paraguay basin, *N. parvipinnis* Costa, 1988 from the rio Cuiabá basin (río Paraguay basin), *N. guaporensis* Costa 1988 from the rio Guaporé basin, and *N. acutirostratus* Costa 1992 from the rio das Velhas, rio São Francisco basin.

Myers (1924) erected *Neofundulus* for *Fundulus paraguayensis*, which has as its type locality a pool near the arroyo Trementina Paraguay. Aramburu *et al.* (1962) studied material of the genus collected in Argentina and verified through meristic analysis and ray fin counts that *Neofundulus paraguayensis* could not be distinguished from *Neofundulus ornatipinnis*, which they considered to be a synonym of the earlier.

Costa (1988) revalidated the two species, based on number of rays of dorsal fin, number of scales in transversal series and caudal peduncle and in the form of the fins he considered *N. ornatipinnis* e *N. paraguayensis* separate species and described two additional species *Neofundulus parvipinnis* and *Neofundulus guaporensis*.

The last *Neofundulus* species of the genus to be described, *N. acutirostratus*, was based on a single specimen collected in 1942 by A. L. Carvalho and G. S. Myers (Costa, 1992).

The genus *Neofundulus* is one of the several annual genera of Rivulidae, that is, they are considered annual fish for living in temporary habitats and resisting desiccation by laying their eggs on the substratum. Annual fish are unique in the fact that they develop their eggs very slowly and can go up through to three diapauses, depending on genetic and environmental factors (Myers 1952; Wourms 1972).

The new species described below was found in a roadside annual pool between the towns of San Miguel and San Juan, departamento de Santa Cruz, in the area near the río San Miguel, a tributary of the right bank of the río Mamore, Bolivia.

MATERIAL AND METHODS

Measurements were taken point-to-point under a stereomicroscope with a digital caliper to the nearest 0.1 mm on the left side of the specimen following Costa (1995; 2007). Measurements are expressed as percents of standard length (SL), except the measurement of the head, which are recorded as percents of head length (HL).

Counts of vertebrae and pleural ribs were taken from radiographs of the holotype, five males paratypes and six females paratypes. Terminology for frontal squamation follows Hoedeman (1958) and Costa (2006). For vertebral counts the caudal compound centrum was counted as a single element. Institutional abbreviations follow Sabaj-Pérez (2010), with addition of UNITAU (Universidade de Taubaté), MNKP- Museo Noel Kempff, Santa Cruz de La Sierra, Bolivia. Osteological features included in the description are those considered phylogenetically informative in studies on *Neofundulus* (1988, 1992).

Neofundulus splendidus, n. sp.

(Figs 1-2, Table I)

Holotype: MNKP-11205, 01 male, 63.5mm SL, Bolivia. Deposited at Museo Noel Kempff, road San Miguel to San Juan, Departamento Santa Cruz, San Miguel city, río San Miguel, río Mamoré basin (16°46’22.0”S 61°11’12.6”W), altitude 440 m, 20 February 2012, Didier Pillet, Bruno Accorsi, Jean Marc Beltramon, Michel Beuchey& Christine Lambert.

Paratypes: MNKP-11206, five males 33.9-40.5 mm SL and six females, 33.5-37.1 mm SL, Bolivia. Deposited at Museo Noel Kempff, Santa Cruz de La Sierra, ZUEC-7303, one male, 35.7 mm SL and one female, 36.2 mm SL. Deposited at Museu de Zoologia da Universidade de Campinas, Campinas, São Paulo, Brazil, collected with the holotype.

Diagnosis: *Neofundulus splendidus* differs from the remaining species of the *Neofundulus* genus by the male body with large black spot at humeral region with diameter of approximately 1.5-2.0 of pupil in males and females (vs. black spot in humeral region, when present, small), 40-41 scales in longitudinal series (vs. 35-38 in remaining congeners), by color pattern, except *N. guaporensis* and *N. acutirostratus*, consisting in three to four yellow lines in the body, alternated with two to three slender red lines, that become thicker and dark towards the caudal peduncle (vs. yellow lines lacking in *N. paraguayensis*, a single yellow line in *N. ornatipinnis*, three yellow lines alternated with three red lines in *N. parvipinnis)*, the color of pectoral fin in males, with finely delicate drawing with narrow white grading to yellow lines within a dark orange background (vs. pectoral fin with black spots in remaining congeners), presence of four to five transverse black bars in dorsal fin in males (vs. black bars absent or presence of small black spots, or else combinations of two black bars with small black spots in the remaining *Neofundulus* species),

Table I. Morphometric and meristic data for the holotype (H) and paratypes of *Neofundulus splendidus.*

	H	Paratypes	
	Male	Male n = 5	Females n = 6
Standard length (mm)	63.5	33.9-40.5	33.5-37.1
Percents of standard length			
Body depth	66.4	22.1-23.6	18.5-20.5
Caudal-peduncle depth	14.3	13.6-14.5	12.7-14.4
Pre-dorsal length	67.1	73.7-77.0	71.4-75.4
Pre-pelvic length	53.5	52.3-54.0	53.7-57.0
Length of dorsal-fin base	17.8	10.8-13.3	12.7-15.7
Length of anal-fin base	21.2	17.7-23.9	17.1-22.3
Caudal-fin length	18.4	24.2-28.4	22.4-30.4
Pectoral-fin length	21.4	19.8-24.1	15.7-26.9
Pelvic-fin length	11.8	11.3-17.9	11.6-16.1
Head length	24.7	29.3-32.4	28.4-33.9
Percents of head length			
Head depth	68.0	57.3-64.1	52.6-62.3
Head width	34.6	32.5-40.7	31.3-35.9
Lower jaw length	17.2	16.9-20.8	17.1-19.4
Eye diameter	28.0	27.9-29.2	26.5-29.4
Counts			
Dorsal fin	10	9-10	11-12
Caudal fin	22	22	22-23
Anal fin	14	14-15	15-16
Pelvic fin	07	07	6-7
Pectoral fin	14	12-14	12-14
Meristic			
Scales in longitudinal series	40	40-41	40-41
Scales in transversal series	09	09	09
Horizontal scales around caudal peduncle	16	16	16

Fig. 1. *Neofundulus splendidus*, male, holotype, 63.5 mm SL: Bolivia, Departamento de Santa Cruz, San Miguel. Photo by G. Dethu.

three transverse brown bars in the body of females (vs. bars lacking in females of the remaining congeners), transverse brown bars in dorsal and anal fins in females (vs. brown bars absent or presence of small black spots in dorsal fin of the females of remaining *Neofundulus* species).

Description: Morphometric and meristic data presented in Table I. Largest specimen examined 63.5mm SL. Body elongated, slender, subcylindrical, deeper than wide, compressed posteriorly. Greatest body depth at level of pelvic-fin base. Dorsal profile gently convex from snout to end of dorsal-fin base, approximately straight to slightly concave on caudal peduncle. Ventral profile slightly convex on head, almost straight from anterior portion of abdominal region to end of anal-fin base, slightly concave along caudal peduncle. Greatest body depth at level of pelvic-fin base. Snout very blunt. Jaws long.

Tip of dorsal and anal-fins slightly pointed in male, rounded in female, lacking filamentous rays. Anal fin approximately rectangular in males, longer than deep, reaching vertical through base of caudal fin. Anal-fin rounded, short, and without filamentous rays in females.

Caudal-fin rounded to subtruncated in males and females, without filamentous rays. Pectoral-fin elliptical. Pelvic-fin pointed, tip reaching between base of 3rd and 4th anal-fin rays in male and 1st and 2nd anal-fin rays in female. Pelvic-fin bases in close proximity in males, and separated by small interspace, in females. Dorsal-fin origin on vertical through base of 5th or 6th anal-fin ray, and between neural spines of 22nd and 24th vertebrae. Anal-fin origin between pleural ribs of 17th and 19th vertebrae. Dorsal fin rays 10-13; anal fin rays 14-16; caudal fin rays 22-24; pectoral fin rays 12-14; pelvic fin rays 7.

Scales large, cycloid. Body and head entirely scaled, except antero-ventral surface of head. Body scales extending over caudal-fin base; no scales on dorsal and anal fins. Frontal squamation E-patterned; E-scales not overlapping medially; scales arranged in irregular circular pattern around A-scale without exposed margins. Longitudinal series of scales 40-41; transverse series of scales 9; scale rows around caudal peduncle 16. Total vertebrae 34-35. Contact organs absent.

Coloration in life (Figs 1-2):

Males: Side of body with yellow, red and iridescent clear blue spots longitudinally aligned, becoming olive-green on the back, abdominal area whitish. Large black spot on humeral region with diameter of approximately 1.5-2.0 of pupil. Sides of head olive-green. Iris yellow, with dark, vertical brown bar crossing center of eye. Dorsal fin pale yellow, with four to five transverse dark bars. Anal-fin dark olive-green, large golden yellow bar surrounded by dark pigmentation near the basis, in some specimens seemingly to continue into the caudal-fin. Caudal-fin dark gray, with white and yellow spots irregularly distributed, and distal portion of the fin black. Pelvic fin dark orange, without spots or lines. Pectoral fin with finely delicate drawing composed by narrow white grading to yellow lines within a dark orange background.

Females: Side of body light brown, with three longitudinal brown stripes. Large black spot on humeral region with diameter of approximately

Fig. 2. *Neofundulus splendidus*, female, paratype, 37.1 mm SL: Bolivia, Departamento de Santa Cruz, San Miguel. Photo by G. Dethu.

1.5-2.0 of pupil. Dorsum light brown, abdominal region whitish. Sides of head grey, with a pale metallic yellow tinge on opercle. Iris yellow, with pale brown horizontal bar crossing center of eye. Dorsal and anal fins hyaline, with longitudinal brown bars. Caudal-fin pale yellow, with transverse narrow, light brown bars. Pectoral fin yellow with small brown spots. Pelvic fin pale yellow.

Distribution: *Neofundulus splendidus* is currently only known from its type-locality.

Habitat (Fig. 3): The type-locality lies at the plateau area (440 ma.s.l.), just southeast of Llanos de Mojos, which occupy much of the río Mamoré basin in Bolivia (see Loubens et al., 1992). The pool is located beside the highway, between the cities of San Miguel to San Juan. Water temperature of the pool was 24.6°C, at the depth of 40 cm. In the marginal area of the pool, at the depth of 10 cm, water temperature was 29°C. *Neofundulus splendidus* was the only fish species present in the pool, where it was found over its entire area. The pool presented clean water, pH 6.4, 0.5 mg/l of dissolved iron (Fe) and electric conductivity 67 μS. Other animals recorded at the pool were *Phylomedusa* sp. tadpoles, clams and freshwaters crabs. A dense coverage of aquatic vegetation mainly formed by *Echinodorus* sp. and *Nymphaea* sp., was present at the pool.

Behavior in captivity: The male initiates the courtship expanding its unpaired fins next to the female, forming with their bodies, angles of 10° or 15°. Sometimes the male touch, with its mouth, the female abdominal region. After the male display, it touches the substratum with its snout, and the couple dives in the substratum. The female lay two or three large-sized (1.5 mm diameter) eggs per day. Males can be sometimes aggressive towards the female, which makes necessary to separate the couple after the end of courtship.

Etymology: In reference to the exuberant colors of the male. An adjective.

DISCUSSION

Neofundulus splendidus can be easily identified from its congeners by the color pattern of the pectoral-fin in males, and by the presence of a large black spot in the humeral region of males and females. *Neofundulus* was previously found in southern Bolivia in the Paraguay basin (Costa et al. 1996), but this is the first record of the genus for the río Mamore basin. Although the genus *Neofundulus* had already been found in another tributary of the rio Madeira basin, the rio Guaporé basin, the new species extends the distribution of the genus and suggests that the genus might be more widespread in other hydrographic basins on the right bank of the Amazon basin.

Other species of annual fish of several genera, *Papiliolebias*, *Spectrolebias*, *Trigonectes*, *Moema*, *Aphyolebias*, and *Pterolebias* were found recently in Bolivia, at the río Mamore, rio Guaporé and río Paraguay basins (Costa 1996, 1998a, Costa et al. 1996, Costa et al. 1997). Since the area searched was relatively small when the total area of these hydrographic basins is considered, and also considering the remoteness of most of the area, it is very likely that further fieldwork will discover additional new rivulid species in this region.

Neofundulus is distinguished from the remaining genera of Rivulidae family by having the following synapomorphies: fourth pectoral radial not expanded ventrally, a subasal longitudinal white-yellowish stripe on the anal fin in males, transverse black bars on caudal fin of females, and black spots on pectoral fin of males (Costa 1998b) *Neofundulus splendidus* presents very narrow yellow lines on the pectoral fins, instead of black spots present on the other *Neofundulus* species, thus an autapomorphy of this species. *Neofundulus splendidus* appears to be closer to *Neofundulus parvipinnis*, since because both species share 14 to 16 anal-fin rays and a color pattern of the males presenting longitudinal yellow stripes.

Fig. 3. Type locality of *Neofundulus splendidus* near río San Miguel, upper río Mamoré basin, Bolivia. Photo by D. Pillet.

Key to *Neofundulus* species based on Costa 1992, with the addition of *Neofundulus splendidus:*

1a. Origin of dorsal fin on vertical through base of first or second anal-fin rays 2
1b. Origin of dorsal fin on vertical through base of fifth or sixth anal-fin rays..... 4
2a. Caudal-fin rounded; tip of pelvic fin in males reaching fourth to sixth anal-fin ray;16 horizontal scales around caudal peduncle .... *N. paraguayensis*
2b. Caudal-fin truncated; tip of pelvic fin in males reaching first or second ray of anal fin; 18 to 20 horizontal scales around caudal peduncle 3
3a. Lateral profile of the head pointed; caudal peduncle depth 17.0 % SL..... ..... *N. acutirostratus*
3b. Lateral profile of the head rounded; caudal peduncle depth 12.5-15.0 % SL .... *N. ornatipinnis*
4a. 12 rays anal fin..... *N. guaporensis*
4b. 14 to 16 rays anal fin 5
5a. 35 to 36 scales in longitudinal series .... *N. parvipinnis*
5b. 40 to 41 scales in longitudinal series .... *N. splendidus*

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REFERENCES

ARÁMBURU, A. S. A., ARÁMBURU, R. H. & RINGUELET, R. A. 1962. Peces paranaenses nuevos para la fauna argentina. *Physis XXIII*, 65: 223-239.

COSTA, W. J. E. M. 1988. Sistemática e distribuição do gênero *Neofundulus* (Cyprinodontiformes: Rivulidae), com a descrição de duas espécies novas. *Rev. Brasil. Biol.*, 48 (2): 103-111.

COSTA, W. J. E. M. 1992. Descrição de uma nova espécie de *Neofundulus* (Cyprinodontiformes: Rivulidae), da bacia do rio São Francisco, Brasil. *Revista Brasileira de Biologia* 52 (4): 615-618.

COSTA, W. J. E. M. 1995. Pearl killifishes: The Cynolebiatinae. Systematics and Biogeography of a Neotropical annual fish subfamily (Cyprinodontiformes: Rivulidae). *TFH, Neptune* City, 128 pp.

COSTA, W. J. E. M. 1996. A new Specie of the annual fish genus *Pterolebias* (Cyprinodontiformes: Rivulidae) from the Rio Mamoré basin, Bolivian Amazon. *Neotropical Ichthyology*, 7: 91-96.

COSTA, W. J. E. M. 1998a. Revision of the Neotropical annual fish genus *Plesiolebias* (Cyprinodontiformes: Rivulidae). *Ichthyological Exploration of Freshwaters*, 8: 313-334.

COSTA, W. J. E. M. 1998b. Phylogeny and classification of Rivulidae revisited: Origin and evolution of annualism and miniaturization in rivulid fishes (Cyprinodontiformes: Aplocheiloidei). *Journal of Comparative Biology*, 3: 33-92.

COSTA, W. J. E. M. 2006. Descriptive morphology and phylogenetic relationship among species of the Neotropical annual killifish genera *Nematolebias* and *Simpsonichthys* (Cyprinodontiformes: Aplocheiloidei: Rivulidae). *Neotropical Ichthyology*, 4: 1-26.

COSTA, W. J. E. M. 2007. Taxonomic revision of the seasonal South American killifish genus *Simpsonichthys* (Teleostei: Cyprinodontiformes: Aplocheiloidei: Rivulidae). *Zootaxa*, 1669: 1-134.

COSTA, W. J. E. M., BARRERA, S. & SARMIENTO, J. 1996. A new species of annual fish genus *Pterolebias* (Cyprinodontiformes: Rivulidae) from the Rio Mamoré basin, Bolivian Amazon. *Ichthyological Exploration of Freshwaters*, 7: 91-95.

COSTA, W. J. E. M., BARRERA, S. & SARMIENTO, J. 1997. *Simpsonichthys filamentosus*, une nouvelle espèce des Llanuras Benianas, bassin du Rio Mamoré, Bolivia. *Revue Française d'Aquariologie*, 24: 83-86.

EIGENMANN, C. H. & KENNEDY, C. H. 1903. On a collection of fishes from Paraguay with a synopsis of the American genera of cichlids. *Proc. Acad. Nat. Sci. Philad.*, 477-537.

HOEDEMAN, J. J. 1958.The frontal scalation pattern in some groups of tooth carps. *Bulletin of Aquatic Biology*, 1: 23-28.

MYERS, G. S. 1924. A new poeciliid fish from Congo, with remarks on funduline genera. *Amer. Mus. Novitates*, 116: 1-9.

MYERS, G. S. 1935. Four new freshwater fishes from Brzil, Venezuela, and Paraguay. *Proc. Biol. Soc. Wash.*, 48: 7-14.

MYERS, G. S. 1952. Annual fishes. *Aquarium Journal*, 2: 125-141.

NIELSEN, D. T. B. 2008. *Simpsonichthys e Nematolebias. Cabral Editora e Livraria Universitária, Taubaté.* 235 pp.

SABAJ-PÉREZ, M. H. 2010. Standard symbolic codes for institutional resource collections in herpetology and ichthyology: an Online Reference. Verson 1.5 (4 Oct 2010). Electronically accessible at http://www.asih.org/, American Society of Ichthyologists and Herpetologists, Washington, DC.

WOURMS, J. P. 1972. The developmental biology of annual fishes III. Pre-embryonic and embryonic diapause of variable duration in the eggs of annual fishes. *Journal of Experimental Zoology*, 182: 389-414.