

## ***Hemibrycon santamartae*: a new species from the Rancheria river of eastern Caribbean Colombia (Characiformes: Characidae)**

### ***Hemibrycon santamartae*: una nueva especie del río Ranchería, de la región oriental del Caribe Colombiano (Characiformes: Characidae).**

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#### **Abstract**

Based on 69 specimens, using morphometric, meristic and osteological characters, we describe a new species of *Hemibrycon* from the Rancheria River, a Caribbean coastal drainage of eastern Colombia that drains the Sierra Nevada de Santa Marta. *H. santamartae* is distinguished from congeners by the following: presence of small hooks on the first six to eight branched anal-fin rays of males, with two to four hooks on only the middle sections of the rays; adipose fin pigmented but with hyaline margins; an apophysis present on the dorsal margin of the rhinosphenoids that is directed toward the anterior tip; ventral process of the angulo-articular reduced and with a straight margin. Data on the phylogenetic position of the new species are presented.

**Key words.** Fish, taxonomy, freshwater, South America.

#### **Resumen**

Con base en una serie de 69 ejemplares, a partir de caracteres morfométricos, merísticos y osteológicos, se describe una nueva especie de *Hemibrycon* para el río Ranchería, de la Sierra Nevada de Santa Marta, Caribe Colombiano. *H. santamartae* se distingue de sus congéneres por: la presencia de ganchos pequeños en los primeros 6 a 8 radios ramificados de la aleta anal de los machos, dos a cuatro ganchos sobre los radios ramificados de la aleta anal en la parte media de los radios, aleta adiposa pigmentada con bordes hialinos, una apófisis en el borde dorsal del rinoesfenoides dirigida hacia el extremo anterior, proceso ventral del ángulo-articular reducido y con borde recto. Se incluyen datos sobre su situación filogenética dentro del grupo.

**Palabras claves.** Pez, taxonomía, agua dulce, Sur América.

#### **INTRODUCTION**

The current diagnosis of the genus *Hemibrycon* is ambiguous, and defined using characters that overlap with other characid genera, such as *Bryconamericus*. Following the traditions of their time, (1) and (2) did not define this or the many other genera they created with phylogenetic criteria. The attributes they used to define *Hemibrycon* are: maxilla with teeth along the greater part of its length; dentary with only one row of teeth; premaxilla with two rows of teeth, males without glandular sac at base of caudal fin. Recently, however, *Hemibrycon* was found to be monophyletic based on osteological and other characters (3).

One of the principal difficulties encountered when describing new species of *Hemibrycon* and a better appreciation of the species diversity of this genus has been the lack of unique characters that would allow adequate diagnosis. Faced with this dilemma, (4) and (5) described subspecies from the upper Cauca and Lake Maracaibo basins.

The genus *Hemibrycon* now includes 26 species (6; 7; 8; 9; 10; 11), reflecting considerable diversity that was previously unrecognized. The purpose of this report is to describe yet another new species of *Hemibrycon*, using in addition to traditional counts and measurements, osteological and sexual dimorphism characters to distinguish it from congeners.

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**MATERIAL AND METHODS**

Fish were preserved in 10% formalin and later transferred to 70% ethanol and deposited in the Ichthyology Laboratory of the University of Quindío, Armenia, Colombia (IUQ). We studied 69 specimens of the new species. Institutional abbreviations follow standard ASIH abbreviations listed at <http://www.asih.org>, with addition of the following institutions: in Colombia: Instituto de Investigaciones Biológicas "Alexander Von Humboldt", Villa de Leyva, Boyacá (IAvH); Laboratorio de Ictiología, Universidad del Quindío, Armenia (IUQ); in Ecuador: Museo de Zoología, Departamento de Ciencias Biológicas, Escuela Politécnica Nacional de Quito, (MEPN); in Perú: Museo de Historia Natural, Universidad Nacional Mayor de San Marcos, Lima (MNH-UNMSM); in Venezuela: Fundación La Salle de Ciencias Naturales-Museo de Historia Natural, Caracas (MHNLS). In the Specimens Examined section, the number of individuals is given in parentheses.

Measurements were taken with digital calipers, and recorded to hundredths of mm. Counts and measurements were done on the left side of specimens when possible and are presented in Table 1, following (12) and (13). A principal component analysis (PCA) was undertaken assuming allometric growth.

All measurements were log transformed, correcting for size using methods of (14) and (15). In the PCA we included the following species: *H. dariensis* (n= 102), *H. metae* (n= 121), *H. jabonero* (n= 86) and *H. santamartae sp. n.* (n= 69). For the number of branched anal-fin rays we used analysis of variance (ANDEVA) at significance level 0.05, and the degrees of freedom given in parentheses.

To interpret the behavior of these measurements when differentiating among species of *Hemibrycon*, we used Tukey or box plots. Data were processed using Past software, version 1.28, 2004 and Sigma Plot version 10.0, 2006.

Observations of bone and cartilage were made using cleared and stained specimens (C&S) prepared with procedures of (16) and (17). Bone names follow (18), (19) and (20). Vertebral counts include the four that comprise the Weberian apparatus, and the terminal centrum is counted as one. Descriptions based only on external characteristics of *Hemibrycon* are not adequate; since externally they are all very similar, except for a few meristic characters (21).

For this reason we recommend the use of osteological characters in species descriptions of *Hemibrycon*, as we do here (Tab. 1, Fig. 1).



**Fig. 1.** *Hemibrycon santamartae sp. n.* (Holotype), female, 54.9 mm SL. Colombia, Magdalena, Bongá-Bunkuangueka, Ranchería River drainage, Santa Clara River.

**Table 1.** Morphometric and meristic data of *Hemibrycon santamartae sp. n.* Standard and total length in mm; mean in parenthesis.

	Holotype	Paratype n = 68
<b>Morphometric:</b>		
Standard length (mm)	54.85	36.24-76.32 (53.33)
Total length	66.39	41.18-93.32 (66.20)
<b>Percentages of SL:</b>		
1. Body depth	26.36	22.12-32.37 (28.00)
2. Snout-dorsal fin origin distance	52.94	42.31-58.30 (52.79)
3. Snout-pectoral fin insertion distance	25.21	20.44-27.02 (24.36)
4. Snout-pelvic fin insertion distance	43.85	35.66-47.56 (43.56)
5. Dorsal-fin origin-pectoral-fin distance	39.85	32.11-44.17 (40.12)
6. Snout-anal fin origin distance	58.33	46.48-62.84 (58.22)
7. Dorsal fin origin-hypural plate length	50.1	31.13-55.01 (48.89)
8. Dorsal fin origin-anal fin origin length	28.98	23.07-33.59 (29.18)
9. Dorsal fin length	22.84	17.56-25.66 (22.35)
10. Pectoral fin length	21.44	14.82-23.81 (21.26)
11. Pelvic fin length	14.62	11.85-21.45 (15.16)
12. Anal fin length	16.79	11.56-19.53 (16.24)
13. Caudal peduncle depth	11.45	8.74-15.64 (11.28)
14. Caudal peduncle length	9.12	6.21-14.78 (10.06)
15. Head length	23.52	18.41-25.18 (22.29)
<b>Percentages of HL:</b>		
16. Snout length	23.8	21.37-31.63 (27.28)
17. Orbital diameter	40.78	33.09-45.7 (39.99)
18. Postorbital distance	34.6	31.11-40.86 (36.06)
19. Maxilla length	32.95	25.00-40.46 (32.42)
20. Interorbital distance	34.8	30.73-41.03 (36.07)
21. Upper jaw length	29.44	23.94-35.53 (30.06)
<b>Meristic:</b>		
Lateral line scales	39	38-43
Scales rows between dorsal-fin origin and lateral line	7	6-8
Scales rows between anal-fin origin and lateral line	6	5-7
Scales rows between pelvic-fin insertion and lateral line	5	5-7
Predorsal median scales	13	11-15
Dorsal-fin rays	iii,7	iii,7
Anal-fin rays	iii,22	iii-v,20-23
Pelvic-fin rays	ii,6	ii,6
Pectoral-fin rays	ii,10	ii,9-12

**Holotype.** IUQ 2306, female, 54.9 mm SL, Colombia, Magdalena Basin, Departamento Cesar, Municipio Atanquez, Sierra Nevada de Santa Marta, Ranchería River drainage, Santa Clara-Candela River, Bongá-Bunkuangueka, approx. 11° 00' N, 72° 46' W.

**Paratypes.** All from Colombia in Departamento Cesar, Ranchería River drainage: IUQ 848 (1) collected with holotype; IUQ 924 (1), Municipio Atanquez, Sierra Nevada de

Santa Marta, Candela River, approx. 11° 15' N, 74° 05' W; IUQ 929 (3), Magdalena, Sierra Nevada de Santa Marta, Quebrada Honduras on road to Mutaiahi, approx. 11° 15' N, 74° 10' W; IUQ 1443 (1 C&S), Municipio Atanquez, Sierra Nevada de Santa Marta, Candela River, approx. 11° 00' N, 72° 46' W; ICNMMH 10834 (18), La Guajira. Municipio Distracción, corregimiento Chorreras, puente el Cercado, vía Distracción – Caracolí, approx. 11° 15' N, 74° 05' W; ICNMMH 10839 (19), Colombia, La Guajira, municipio Marocaso, approx. 11° 15' N, 74° 05' W; ICNMMH 10881(24), La Guajira, municipio Marocaso, Marocaso River, approx. 11° 15' N, 74° 05' W.

**Non types:** Colombia, Ranchería River drainage: ICNMMH 8754 (34), La Guajira, Oreganal, El Cerrejón Mine, before Cerrejon Creek; ICNMMH 8869 (40), La Guajira, Distracción, corregimiento Chorreras, El Cercado; ICNMMH 8706 (5), La Guajira, Oreganal, Mina El Cerrejón, La Batea.

**Diagnosis.** *Hemibrycon santamartae* sp. n. is distinguished from all other *Hemibrycon* by: males with small hooks on the sixth to eight branched anal-fin rays (vs. small hooks present on all the anal-fin rays or well developed on the fourth to twelfth anal-fin rays); two to four hooks present on first branched anal-fin rays located on only the middle section of each ray (vs. with more than four hooks on first branched anal-fin rays located from central part to distal tip, or only at neartip of ray); adipose fin pigmented but with hyaline margins (vs. adipose fin hyaline or completely pigmented); dorsal margin of rhinosphenoids with an apophysis on its anterior tip (vs. dorsal margin of rhinosphenoids without apophysis); ventral process of angulo-articular reduced and with straight margin (vs. angulo-articular well developed and with rounded margin); by the reduction of the ventral process of the anguloarticular (vs. not reduced) and by the number of branched anal-fin rays (20-24 vs. 15-19 or 24-34 except *H. colombianus*, *H. helleri*, *H. huambonicus*). *H. santamartae* sp. n. is similar to *H. jabonero*; however, most specimens can be distinguished from that species by the lower number of branched anal-fin rays (20-23 vs. 23-26).

**Description.** Body short and robust anteriorly. Dorsal profile of head and body curved from snout to dorsal-fin origin, oblique from last dorsal-fin ray to base of caudal fin. Ventral profile of body convex from snout to anal-fin base, more pronounced at level of posterior part of pectoral fins. Oblique dorsal-fin margin, its second simple and first two branched rays longest. Short head and snout; equal jaws; terminal mouth; soft and flexible lips, not covering the external premaxillary tooth row; ventral part of upper jaw flat; posterior tip of maxilla surpasses anterior margin of orbit; posterior nasal openings vertically ovoid.

Dorsal surface of mesethmoids narrow, its posterior border between the frontals concave; dorsal margin of lateral process of mesethmoids undulated and joined to ascendant process of premaxilla. Dorso-posterior extreme of orbitosphenoids and antero-dorsal extreme of pterosphenooids extending as processes of equal dimensions; rectangular, narrow and straight over the frontal. Posterolateral process of sensory canal over dorsal surface of cranium not surpassing canal of lateral process of supraoccipital. Dorso-posterior part of pterotic extends horizontally and projects laterally. Dorso-posterior surface of parasphenoids extends dorsally as laminar lateral processes with convex borders and project posteriorly towards prootic, forming a ventral foramen on each side over the antero-ventral extreme of prootic.

Short supraoccipital spine. Posterior part of rhinosphenoids united by cartilage to orbitosphenoids, dorsal margin with apophysis in anterior section, bordered with cartilage; rhinosphenoid cartilage extends parallel to ventral border of lateral ethmoids and over anterior extreme of parasphenoids. Posterior border of palatine undulated and extending over ectopterygoid, squared on lateral surface with small foramina. Anterior section of ectopterygoids thickened: four to six times wider than posterior part.

Six infraorbitals present; third infraorbital with the posteroventral margin in contact with preopercle, covering the quadrate, the metapterygoid and the ventral portion of the hyomandibular; fourth and fifth infraorbitals about the same size and covering dorsal part of hyomandibular; sixth infraorbital reduced, covering the spine of the sphenotic and projected as a canal to reach the canal of the latero-posterior margin of frontal. Supraorbital is absent. Premaxilla with two rows of teeth; the outer with three to five teeth with three or four cusps, aligned in a straight line; the inner with four teeth with five to seven cusps. Maxilla long, with 6-11 teeth with three to five cusps. Dentary with three large pentacuspoid teeth with very small lateral cusps in front, the fourth tooth is medium-sized and pentacuspoid and is followed by eight to ten lateral teeth that gradually diminish in size and from front to back change from bicuspid to tricuspoid to unicuspid.

Eight thin, elongate supraneurals with osseous bases and cartilaginous tips are distributed between neural spines 5 to 13. Nine proximal pterygiophores of the dorsal fin are inserted between neural spines 14-21; the terminal piece of the dorsal-fin pterygiophores is osseous. The anal fin has 22 proximal pterygiophores. Pectoral girdle with long, thin posttemporal extending over postero-dorsal section of opercle, ventral margin covering dorsal process of supraclithrum, and in contact with the first postclithrum.

Four proximal radials. The anterior margin of scapular is not in contact with mesocoracoids. The dorsal part of mesocoracoids extends over internal surface of cleithrum, and projects ventrally as a small process without cartilage. Union of mesocoracoids, scapular and coracoids without cartilage. Pelvic fins short, their tips not reaching anal-fin origin. Pelvic bone elongate, lateral margin straight with anterior tip of cartilage, internal surface concave, radial element cartilaginous, ischiatic process osseous, short, curved and prolonged as a pointed cartilaginous process.

Caudal fin with short pointed lobes. Principal caudal-fin rays 10/8. Procurrent caudal-fin rays 9/11. Caudal fin with two enlarged scales at base of peduncle. Lateral line with 39-43 pored scales, the pores forming a gentle curve between the first and the seventh to eighth scales, the rest straight. Total number of vertebrae 39-40.

#### Secondary sexual dimorphism.

Males have hooks on the anal and pelvic-fin rays. There are 4 to 6 hooks located on the middle sections of the sixth to eighth branched anal-fin rays. Branched pelvic-fin rays with small hooks that extend all along the rays.

#### Color in alcohol.

Sides of body brown, lateral band wide with a black lateral line along posterior portion. Humeral spot dark and vertically elongated. Base of dorsal-fin rays pigmented, with posterior margin hyaline. Pectoral and pelvic-fin rays hyaline. Base of anal-fin rays hyaline and posterior part pigmented. Adipose fin pigmented with hyaline margins. Caudal-fin rays pigmented in both dorsal and ventral lobes, and with a black band on middle caudal-fin rays.

#### Distribution.

Known from the Sierra Nevada of Santa Marta mountain range, in the Rancheria river drainage, northeastern Colombia.

#### Etymology.

The name refers to the Sierra Nevada of Santa Marta mountain range, of northeastern Colombia, where this species is found.

#### Comments.

The principal component analysis (PCA) of morphological characters was not informative. Estimation of accumulated variability for these species is 23.57%, 42.2%, 55.41%, 63.96% and 71.91%. ANDEVA did detect significant differences in the number of branched anal-fin rays of *H. santamartae* sp. n., from *H. dariensis*, *H. metae* y *H. jabonero* (20-23 vs. 23-31,  $F=0005(3)(323)=196.1$   $p<0.001$ ) (Fig. 2).

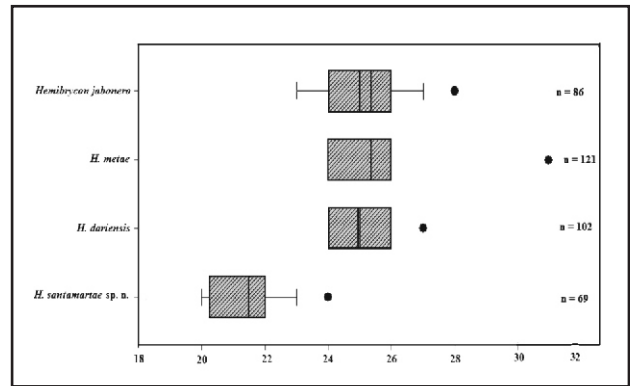


Fig. 2. Box plots for the number of branched anal-fin rays of *Hemibrycon dariensis*, *H. metae*, *H. jabonero* and *H. santamartae* sp. n. ( ) atypic values; (|—|) outside of the boxes indicates minimum and maximum values; ( ) inside the boxes indicates the median.

#### Discussion

The monophyletic species concept states that only species diagnosed by autapomorphies can be accepted as valid, indicating that such species have an independent history from related species, and that these may in some cases have disappeared without a trace (22; 23). However, (24) state that autapomorphies can, in additional analyses using more taxa, reveal that they were in fact synapomorphies of two or more species. *Hemibrycon santamartae* sp. n. has two autapomorphies that serve to separate it from all species of *Hemibrycon*, *Bryconamericus*, *Gephyrocharax*, *Astyanax*, *Hyphessobrycon*, *Hemigrammus*, *Pseudochalceus* and *Brycon*.

(3) stated that species of the genus *Hemibrycon* constitute a monophyletic group based upon four synapomorphies associated with orbital region, cranium, suspensorium and color patterns. *Hemibrycon santamartae* sp. n., has all of the synapomorphies observed in other *Hemibrycon*. However, its phylogenetic position remains uncertain, since it lacks apomorphies shared within this genus.

The reduction of the ventral process of the angulo-articular is an evolutionary novelty evolved by *H. santamartae* sp. n., and is not common in Characidae, where usually the angulo-articular has two processes directed horizontally or one process oriented vertically. The presence of an apophysis on the anterior section of the rhinosphenoids is not common among species of *Hemibrycon*. In *Hemigrammus*, *Pseudochalceus*, *Moenkhausia* and *Deuterodon* a triangular shaped medial process is observed on the dorsal surface of the rhinosphenoids, but it is different from the condition in *H. santamartae* sp. n.

*Hemibrycon santamartae* sp. n. can be distinguished from all other *Hemibrycon* by characters related to the sexual dimorphism of mature males. The reduction of the number of hooks on the first branched anal-fin rays separates *H. santamartae* sp. n. from all congeners that usually have hooks on up to the twelfth to fourteenth branched rays or even up to the last branched anal-fin ray. The number of hooks on each branched anal-fin ray varies from two to four. Sexual dimorphism is a well known pattern of morphological variation in the biotic world, and different attempts have been made to quantify and describe the differences between the sexes and use them phylogenetically (25; 3). There is evidence that mature males of *Aphyrocharax anisitsi*, *Creagrutus brevispinnis*, *Bryconamericus caucanus*, *Roeboides dayi* and *Hemibrycon boquiae* retain these secondary sexual characteristics for their entire lives once they develop (26; 27; 28; 29).

#### COMPARATIVE MATERIAL EXAMINED

**Hemibrycon jabonero:** (all from Venezuela), EBRG 4324 (20), (2 C&S); Aragua, Limón River Pozo 350 masl, Profauna, El Limón; MCNG 24838 (12), Maracaibo Lake basin, Escalante River, Crta. 1, Mérida-Táchira; MCNG 16972 (7), Monagas, Cocoyal or Cocollar River, in campo Elías before San Antonio tributary Guarapiche River, San Juan slope; MCNG 7475 (2); Urbante, on the way to Tovar, Mérida; MCNG 42720 (17), Maracaibo Lake basin, Chama river, Mérida; MCNG 48498 (39), Apure-Portuguesa, caño north of Ospino, La Estación-La Reinosa way (9° 22' 17"N, 69° 29' 38"W); MBUCV 24896 (3), Carabobo, Lago de Valencia basin, El Ercigue river, north of San Joaquin; MBUCV 22854 (3), Carabobo, San Diego river La Cumaca, 5 km north of San Diego; MBUCV 12530 (4), Miranda, Grande river, approximately 500 m from the mouth of the Santa Cruz River in the Rio Grande, National Park Guatopo.

**H. taeniurus:** MCNG 43512 (24), Venezuela, Sucre, Cristalino river; MCNG 43512 (2 C&S), Venezuela, Sucre, Cristalino river; IUQ 1201 (1 C&S), Venezuela, Punceres river, Cachipo, Monagas.

**H. guppyi:** USNM 290406 (1 C&S), Trinidad & Tobago, Trinidad, Matura river; USNM 290406 (7), Trinidad & Tobago, Trinidad, Matura river.

**H. metae:** Colombia: IAvH 3124 (1 C&S), Casanare, Pauto, Tocaría, Chave y Cravo Sur rivers basins; IUQ 1448 (1 C&S); Venezuela, mainstem Orituco river; IAvH 2986 (3), Pauto river, Tocaría, Charre y Cravosur; IAvH 2973 (7), Casanare, quebrada Palmicha, Aguazul, tributary Cachiza river; IAvH 2991 (3), Casanare, Únete river, Cusiana y Tua; IAvH 3122

(10), Casanare, quebrada Chichaza, Aguazul tributary Cachiza river. IAvH 3125 (33), Casanare, Únete, Cusiana y Tua rivers; IAvH 3134 (4), Casanare, Nunchia river; IAvH 3127 (2), Casanare, quebrada Barreña; IAvH 3129 (42), Casanare, quebrada Cupiagua-Únete River basin; IAvH 3124 (17), Casanare, Pauto river, Tocaría, Crave y Cravo Sur; Venezuela: MCNG 26774 (2 C&S), Santa Bárbara river, 3 km NE Santa Bárbara, Barinas (1° 50' 13" N, 71° 11' 13").

**H. boquiae:** IUQ 536 (4) (C&S), Colombia, Quinchía; vereda Opirama, Opirama river, quebrada Talabám, (5° 17' 47" N, 75° 45' 08" W) 1172 masl; IUQ 871 (15), Colombia, Quindío; quebrada Boquia, vereda Boquia, Quindío River tributary.

**H. rafaelse:** IUQ 499 (2 C&S), (Paratypes), Colombia, Risaralda, San Rafael river on the way to Santuario; IUQ 1212 (2 C&S), Colombia, Quindío, quebrada Canceles, La Vieja river tributary, Reserva El Ocaso, (40° 33' 4" N, 75° 52' 71" W) 999 masl; IUQ 1213 (2 C&S), Colombia, Quindío, quebrada Canceles, La Vieja river tributary, Reserva El Ocaso, (40° 33' 4" N, 75° 52' 71" W) 999 masl.

**H. colombianus:** ICNMMNH 753 (4), Colombia, Santander, Nevado river, Capita Negro; ICNMMNH 755 (1 C&S); Colombia, Santander, Nevado river, Capita Negro; IAvH 3130 (28, 1 C&S), Colombia, Santander, Moniquira river and Suárez river tributaries.

**H. dariensis:** IUQ 849 (26), Colombia, Zungo river, road, Sistema León river; IUQ 852 (2 C&S), Colombia, Urabá, Villa Ortega; IUQ 1281 (17), Colombia, Ranchería river; ICNMMNH 3141 (11), Colombia, Urabá, Sucio river, municipio Dabeiba; ICNMMNH 3053 (8), Colombia Antioquia, Mutata river; ICNMMNH 3074 (4), Colombia Antioquia, quebrada Murcia, Mutata; ICNMMNH 3057 (5), Colombia, Antioquia, Mutata river, on the way to Chigorodo, Uraba; USNM 293245 (28), Panamá, Darien, Tuirá river (08° N, 77° W) province of Darien, Pucuro river nearly 3-4 km up confluence with River Tuirá, Pacifico; USNM 293218 (2), Panamá Atlántico, Kuna Yala region, Mandinga river between Pingando river, Mandinga town (09° 28' N, 70° 06' W); USNM 293234 (1), Panamá Darien, Pirre river half a kilometer through El Real, (Tuirá river) Pacifico; ICNMMNH 804 (11), Colombia Guajira, quebrada Arroyo Mamón.

**H. polyodon:** IUQ 1142 (2 C&S), Ecuador, Zamora; quebrada Antonio-Guadalupe; MEPN 27 (14, 1 C&S), Ecuador, Sucumbíos, Coec river, estero Venado, 3 km downstream from codo bajo-INECEL camp.

**H. orcesi:** MEPN 1538 (17), Ecuador, Morona-Santiago, Tayusa river, Upano river tributary, under the bridge on the

road Mendez-Sucua; MEPN 1538 (4 C&S), Ecuador, Morona-Santiago; Tayusa river, Upano river tributary, under the bridge on the road Mendez-Sucua.

**H. pautensis:** IUQ 533 (2 Paratypes, C&S), Ecuador, Morona-Santiago, Paute river, at the mouth of the river Namangoza; MEPN 1549 (1), (Holotype), Ecuador, Paute river at the mouth of the river Namangoza

**H. huambonicus:** USNM 273726 (6, 2 C&S), Perú, Huanco, quebrada Huancachypa, Huanco.

**H. jelskii:** IUQ 1141 (2 C&S), Ecuador, Orellana, Divino river 1600 meters from the well Chontayacu I, bloque 18. MCNG 6128 (1 C&S), Ecuador, Orellana, Divino river 1600 meters from the well Chontayacu I, bloque 18. USNM 361171, (3)

Perú, Cusco, La Convención, Echarate Peruanita, quebrada Igoripato. MEPN 1544 (15, 3 C&S), Ecuador, Orellana, Jivino river 1600 meters from the well Chontayacu.

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#### BIBLIOGRAPHY

- (1) Günther A. Catalogue of the fishes in the British museum. Catalogue of the Physostomi, containing the families Siluridae, Characinae, Haplochitonidae, Sternoptychidae, Scopelidae, Stomidae in the collection of the British Museum London. 1864; (5): 1-455.
- (2) Eigenmann CH. The American Characidae. 1927; Mem Mus Comp Zool; XLIII (3): 311-428.
- (3) Arcila-Mesa DK. Análisis filogenético y biogeográfico de las especies de Hemibrycon (Characiformes, Characidae). Trabajo de grado B.Sc. Universidad del Quindío, Programa de Biología, Armenia, Colombia. 2008; 90 pp.
- (4) Dahl G. Los peces del norte de Colombia. Ministerio de Agricultura, Instituto de Desarrollo de Recursos Naturales Renovables (Inderena). 1971, Bogotá, Colombia; 391pp.
- (5) Schultz LP. The fishes of the family Characinae from Venezuela, with descriptions of seventeen new forms. 1944; Proc U S Nat Mus; 95 (3181): 235-367.
- (6) Román-Valencia C, Ruiz-C. R., Barriga R. Una nueva especie de pez del género Hemibrycon (Characiformes: Characidae). 2006; Rev Biol Trop, 54 (1): 1-9.
- (7) Román-Valencia C, Ruiz-C R. Una nueva especie de pez del género Hemibrycon (Characiformes: Characidae) del Alto río Atrato, Noroccidente de Colombia. 2007; Caldasia; 29 (1): 121-131.
- (8) Román-Valencia C, Ruiz-CR, Giraldo A. Dieta y reproducción de dos especies sintópicas: Hemibrycon boquiae y Bryconamericus caucanus (Pisces: Characidae) en la quebrada Boquia, río Quindío, Alto Cauca, Colombia. 2008; Rev Mus Argentino Cienc Nat; 10 (1): 55-62.
- (9) Román-Valencia C, Arcila-Mesa DK. A new species of Hemibrycon (Characiformes: Characidae) from the upper Cauca River, with keys to Colombian species. 2008; Animal Biod Conser; 31(1): 67-75.
- (10) Román-Valencia C, Arcila-Mesa DK. Two new species of Hemibrycon (Characiformes, Characidae) from the Magdalena river, Colombia. 2009; Animal Biod Conser; 32 (2):77-87.
- (11) Román-Valencia C, Arcila-Mesa DK. Five new species Hemibrycon (Characiformes: Characidae) from the Magdalena River basin, Colombia. 2010; Rev Biol Trop; 58 (1) In press.
- (12) Fink WL, Weitzman SH. The so-called cheirodontin fishes of Central America with descriptions of two new species (Pisces: Characidae). 1974; Smith Cont Zool; (172): 1-46.
- (13) Román-Valencia C. Descripción de tres nuevas especies de Bryconamericus (Pisces: Ostariophysi: Characidae) de Colombia. 2003; Mem Fund La Salle Cienc Nat; (155): 31-49.
- (14) Burnaby TP. Growth-invariant discriminant functions and generalized distances. 1966; Biometrics; 22: 96-116.
- (15) McCoy MW, Bolker BM, Osenberg CW, Miner BG, Vonesh JR. Size correction: comparing morphological traits among populations and environments. 2006; Oecologia; (148): 547-554.

- (16) Taylor WR, Van Dyke GC. Revised procedures for staining and clearing small fishes and other vertebrates for bone and cartilage study. 1985; *Cybiurn*; (9): 107-119.
- (17) Song J, Parenti LR. Clearing and staining whole fish specimens for simultaneous demonstration of bone, cartilage and nerves. 1995; *Copeia* (1995): 114-118
- (18) Weitzman SH. The osteology of *Brycon meeki*, a generalized characid fish, with an osteological definition of the family. 1962; *Stanford Ichthyol Bull*; 8 (1): 1-77.
- (19) Vari RP. The Neotropical fish family Ctenoluciidae (Teleostei: Ostariophysi: Characiformes): supra and interfamilial Phylogenetic relationship, with a revisionary study. 1995; *Smith Contr Zool*; (564): 1-96.
- (20) Toledo-Piza M. The neotropical fish subfamily Cynodontinae (Teleostei: Ostariophysi: Characiformes): a Phylogenetic study and a revision of *Cynodon* and *Rhaphiodon*. 2000; *Amer Mus Nat Hist*; (3268): 1-88.
- (21) Román-Valencia C, Arcila-Mesa DK, Hurtado T. Variación morfológica de los peces *Hemibrycon boquiae* y *Hemibrycon rafaelse* (Characiformes: Characidae). 2009; *Rev Biol Trop*; 57 (3): 541-566.
- (22) Rosen DE. Vicariant patterns and historical explanation in biogeography. 1978; *Syst Zool*; (27): 759-188.
- (23) de Pinna MCC. Species concepts and phylogenetic. 1999; *Rev Fish Biol Fishery*; (9): 353-373.
- (24) Sidlauskas BL, Vari RP. Phylogenetic relationships within the South American fish family Anostomidae (Teleostei, Ostariophysi, Characiformes). 2008; *Zool J Linn Soc*; (154): 70-210.
- (25) Delph LF. Processes that constrain and facilitate the evolution of sexual dimorphism. 2005; *Amer Nat*; (166): S1-S4.
- (26) Román-Valencia C. Alimentación y reproducción de *Creagrutus brevipinnis* (Pisces: Characidae) en Alto Cauca, Colombia. 1998; *Rev Biol Trop*; 46 (3): 783-789.
- (27) Román-Valencia C, Muñoz A. Ecología trófica y reproductiva de *Bryconamericus caucanus* (Pisces: Characidae). 2001; *Boll Mus reg Sci nat Torino*; 18 (2): 459-467.
- (28) Gonçalves TK, Azevedo MA, Malabarba LR, Fialho B. Reproductive biology and development sexually dimorphic structures in *Aphyrocharax anisits* (Ostariophysi: Characidae). 2005; *Neotrop Ichthyol*; (3): 433-438.
- (29) Román-Valencia C, Botero A, Ruiz-CR. Trophic and reproductive ecology of *Roebooides dayi* (Teleostei: Characidae) from upper Rio Cauca, Colombia. 2003; *Boll. Mus. Reg. Sci. Nat. Torino*; 20 (2): 487-496.

# Identificación de Bacterias que afectan el establecimiento in vitro de segmentos nodales de *Guadua angustifolia* Kunth

## Identification of bacteria affecting in vitro establishment of nodal segments of *Guadua angustifolia* Kunth

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### RESUMEN.

El cultivo in vitro de segmentos nodales (medio y basal), de ramas de chusquines de *Guadua angustifolia* Kunth., presenta un alto grado de contaminación por bacterias afectando su micropropagación, en la fase de establecimiento. Para la identificación de las bacterias contaminantes, se seleccionaron siete grupos con características de aspecto y color, a las que se les aplicaron medios de cultivo diferenciales, como los utilizados para géneros bacterianos fitopatógenos, como: YDC; B de King; D1; y D5; además, se efectuaron pruebas bioquímicas para confirmar la identificación, como: tinción de Gram; prueba de Hugh y Leifson; Indol; y motilidad. Mediante la presente investigación, se llegó a la identificación de los géneros: *Xanthomonas*, *Pseudomonas*, *Agrobacterium*, y una asociación entre *Erwinia-Pseudomonas*. El género *Pseudomonas* fue el principal agente contaminante para los segmentos nodales basales (26.8%) y la asociación *Erwinia-Pseudomonas* para segmentos nodales medios (26.6%). La sensibilidad a los antibióticos por parte de las bacterias, se determinó a través del antibiograma, donde los géneros encontrados presentaron sensibilidad a Amikacina, Gentamicina y Vancomicina.

**Palabras clave:** in vitro, segmentos nodales, *Guadua angustifolia*, fitopatógenos, antibióticos.

### ABSTRACT

The in vitro culture of nodal segments (medium and basal) of branches chusquines *Guadua angustifolia* Kunth, has a high degree of bacterial contamination, affecting their micropropagation, in the establishment phase. For identification of the contaminating bacteria, seven groups with aroma and color characteristics were selected, which were applied differential culture media, such as those used for phytopathogenic bacterial genera, such as: YDC, King B, D1 and D5. In addition, biochemical tests were performed to confirm identification such as: gram stain, Hugh and Leifson test, Indol; and motility. This investigation led to the identification of the genera: *Xanthomonas*, *Pseudomonas*, *Agrobacterium*, and *Erwinia-Pseudomonas* partnership. The genus *Pseudomonas* was the main pollutant for basal nodal segments (26.8%) and *Erwinia-Pseudomonas* association for media nodal segments (26.6%). The sensitivity to antibiotics by bacteria, were determined by antibiogram, where genera was found to be sensible to Amikacin, Gentamicin and Vancomycin.

**Keywords:** in vitro, nodal segments, *Guadua angustifolia*, plant pathogens, antibiotics.

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### INTRODUCCIÓN

Las técnicas de cultivo in vitro han desarrollado una exitosa y rápida propagación asexual de un gran número de especies vegetales, donde uno de los requisitos básicos es mantener los cultivos libres de microorganismos contaminantes (1,2).

El efecto de los microorganismos contaminantes sobre las plantas in vitro es considerable, si se tiene en cuenta que compiten con ellas por los nutrientes del medio de cultivo y proporcionan daños directos e indirectos por la colonización de sus tejidos y la expulsión al medio de metabolitos tóxicos (2-4).

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