Phylogenetic Analysis and Redescription of the Genus *Henochilus* (Characiformes: Characidae)

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***Henochilus*, a characiform genus endemic to eastern Brazil is reviewed. A single species, *Henochilus wheatlandii*, from the Rio Doce and possibly the Rio Mucuri in the states of Minas Gerais and Espirito Santo is recognized and redescribed. The phylogenetic analysis indicates that *Henochilus* is most closely related to the clade consisting of *Brycon* and likely *Chilobrycon*.**

***Henochilus*, um gênero de Characiformes endêmico do leste do Brasil, é revisto. Uma única espécie, *Henochilus wheatlandii*, do Rio Doce e possivelmente do Rio Mucuri, nos Estados de Minas Gerais e Espirito Santo, é reconhecida e redescrita. A análise filogenética indica que *Henochilus* é mais proximamente relacionado com o clado composto por *Brycon* e possivelmente *Chilobrycon*.**

THE unusual characiform genus *Henochilus* has long puzzled ichthyologists engaged in studies of the Neotropical freshwater fish fauna. The genus, and its single included species (*Henochilus wheatlandii*), were described by Garman (1890) based on a single individual that purportedly was collected in the Rio Mucuri of eastern Brazil (Fig. 1). Other than for the report of a second specimen of *Henochilus* with indefinite locality information collected by the same expedition that secured the holotype (Eigenmann and Myers, 1929), no new information about the genus and species was published for over a century. Collecting efforts by the first author and colleagues at the stated type locality and elsewhere throughout the Rio Mucuri basin in 1991 failed to capture *H. wheatlandii*, and this distinctive species was unknown to local fishers. These results raised the possibility that *H. wheatlandii* was threatened (Rosa and Menezes, 1996) or perhaps extinct (Swerdlow, 1998), most likely as a consequence of changes in the aquatic environment associated with the dramatic anthropogenic alterations of the forests in the Rio Mucuri basin (Lima and Castro, 2000). Apparently thriving populations of *H. wheatlandii* were unexpectedly recently discovered by the third author and colleagues in the Rio Doce, an independent basin of eastern Brazil situated to the south of the Rio Mucuri (Vieira et al., 2000). Confirmation of the existence of populations of *Henochilus* is welcome, but the ecosystems inhabited by the species remain threatened by proposed development projects (Vieira and Alves, 2001).

The recently collected specimens of *H. wheatlandii* together with previously misidentified older collections of the species provide an opportunity to adequately describe the species for the first time. This material also permits us to address the phylogenetic questions concerning this enigmatic genus.

MATERIALS AND METHODS

Proportional measurements are given with respect to standard length (SL) except for subunits of the head which are presented as proportions of head length. Lateral-line scale counts include all pored scales along that series, including such scales located posterior of the hypural joint. In counts of median and pelvic fins, lower-case Roman numerals indicate unbranched rays, and Arabic numerals indicate branched rays. Meristic values for the holotype are indicated in square brackets. Some scale- and fin-ray counts could not be determined as a consequence of the poor condition of the holotype. In such instances the reported values for the holotype were taken either from the original description (scales and paired fins) or from a radiograph of that specimen (unpaired fin-rays). Measurements follow the methods outlined in Fink and Weitzman (1974) with the addition of head height measured along the vertical through the base of the supraoccipital spine.

The molecular phylogenetic analysis was based on a tissue sample from a 225.6 mm SL specimen (MZUSP 73105). Total DNA was extracted from ethanol preserved muscle tissue with the Wizard Genomic DNA Purification Kit (Promega). Fragments of mitochondrial ribosomal genes were amplified with the polymerase chain reaction (PCR) using primers described by Kocher et al. (1989) for the 12S rRNA gene and Palumbi et al. (Palumbi, S., A. Martin, W. O. Romano, L. Stice, and G. Grabowski, The
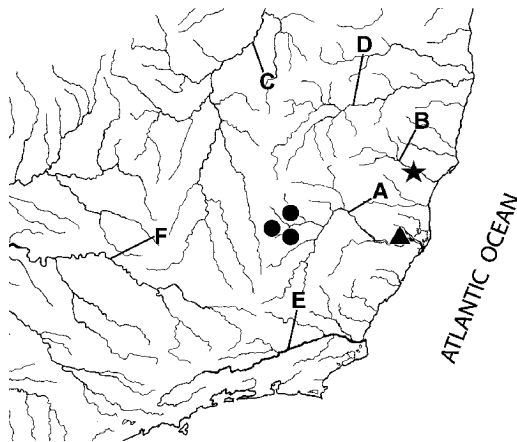
Fig. 1. Map of Rio Doce, Rio Mucuri, and adjoining basins showing distribution of *Henochilus wheatlandii*. Holotype locality indicated by star; nontype Thayer Expedition material indicated by triangle; other samples by dots (A = Rio Doce; B = Rio Mucuri; C = Rio São Francisco; D = Rio Jequitinhonha; E = Rio Paraíba do Sul; F = Rio Grande).

simple fool's guide to PCR, Vers. 2.0, University of Hawaii, Honolulu, 1991) for the 16S rRNA gene. The PCR products were identified in a 1% agarose gel. The amplified segments were extracted from the gel with the kit GFX™ PCR DNA and Gel Band Purification Kit (Amersham Pharmacia Biotech, Inc.). Automated sequencing was performed with the Big Dye™ Terminator v3.0 Cycle Sequencing Kit with Ampli Taq® FS (Applied Biosystems) in an ABI PRISM 377 DNA Sequencer (Applied Biosystems) following manufacturer's specifications. Sequences of 42 species of the order Characiformes obtained by Ortí (1997) and deposited in GenBank were compared to *H. wheatlandii*. The final alignment is available from C. Oliveira upon request. Maximum-parsimony (MP) based phylogenetic analyses were performed using the softwares MEGA2.1 (S. Kumar, K. Tamura, I. Jakobsen, and M. Nei, MEGA: Molecular evolutionary genetic analysis, vers. 2, Pennsylvania State University, University Park, and Arizona State University, Tempe, 2000) and PAUP* beta vers. 4.0b10 (D. L. Swofford, unpubl.) with heuristic searches (1000 replications) using random addition of sequences and the tree bisection and reconnection (TBR) algorithm. In all analyses the character-state optimization method employed was the accelerated transformation (ACCTRAN). Bootstrap resampling (Felsenstein, 1985) was applied to assess support for individual nodes using 1000 replicates with 1000 random additions and TBR branch swapping. Maximum-likelihood (ML) based phylogenetic

relationships were estimated using the software PAUP* beta vers. 4.0b10 (D. L. Swofford, unpubl.). The GTR model (Yang, 1994) incorporating rate variation (Γ) and PINVAR with four Γ-distributed rate classes (Swofford et al., 1996) were used for all likelihood analyses based on a hierarchical hypothesis test of alternative models implemented with Modeltest 3.06 (Posada and Crandall, 1998). The Ti:Tv ratio, gamma shape parameter, and proportion of invariant sites were estimated by maximum likelihood from a maximum parsimony tree. Gaps were treated as missing data. Bootstrap resampling was applied to assess support for individual nodes using 100 replicates with 10 random additions and TBR branch swapping. The clade formed by the African characiform genera *Citharinus* and *Distichodus* was used as outgroup as suggested by Ortí (1997).

Osteological observations were based on a 134.1 mm SL specimen (LIRP 4018) cleared and stained for cartilage and bone using the method of Taylor and Van Dyke (1985). Institutional abbreviations follow Leviton et al. (1985) with the addition of LIRP-Laboratório de Ictiologia de Ribeirão Preto, Departamento de Biologia da Faculdade de Filosofia, Ciências e Letras de Ribeirão Preto, Universidade de São Paulo, Ribeirão Preto, SP, Brazil; and MZUFV-Museu de Zoologia João Mojeen de Oliveira, Viçosa, MG, Brazil. Locality information is followed by institutional catalog numbers, number of specimens in the lot, and the range in standard lengths of that material.

Henochilus Garman

Henochilus Garman, 1890:49 [type species: *Henochilus wheatlandii*, Garman, 1890, by original designation]

Diagnosis.—A member of the subfamily Bryconinae of the family Characidae (see comments under Phylogenetic relationships, below) that differs from *Brycon*, *Catabasis*, *Chalceus*, *Chilobrycon*, and *Triportheus* in having two, rather than three, rows of teeth on the premaxilla. *Henochilus* is further differentiated from *Brycon* (including *Holobrycon*), *Chalceus*, and *Triportheus* by having the teeth of the outer tooth row of the dentary and premaxilla with a greatly enlarged, distally rounded central cusp in large individuals. *Henochilus* also lacks a supramaxilla or an expanded coracoid, features that characterize *Chalceus* and *Triportheus*, respectively. *Henochilus* differs from *Salminus* by having a pair of robust canine or conical symphyseal teeth in the inner

Fig. 2.   *Henochilus wheatlandii*: (A) MZUSP 73118, 75.5 mm SL; and (B) MZUSP 75398, 239.7 mm SL.

tooth row of the dentary contrary to the lack of such dentition in the latter genus.

Henochilus wheatlandii Garman, 1890
Figure 2

Andirá (Brazil, Minas Gerais; Vieira et al., 2000; Vieira and Alves, 2001).

Henochilus wheatlandii Garman, 1890:49 [type locality: Santa Clara, on the Rio Mucuri, Brazil (= Santa Clara, Município de Nanuque, Minas Gerais, Brazil; approximately 17°54′S, 40°13′W)].—Eigenmann and Myers, 1929: 454, pl. 4, fig. 3; pl. 66, figs. 1–4; pl. 94, fig. 1 [redescription apparently based on holotype and one nontype specimen].—Rosa and Menezes, 1996:654 [endangered status].—Vieira et al., 2000:201 [Brazil, Minas Gerais, Rio Doce basin, rediscovery and possible range extension].—Vieira and Alves, 2001:414 [threatened status].

Material examined.—Brazil, Minas Gerais: Santa Clara, on Rio Mucuri (= Santa Clara, Município de Nanuque, approximately 17°54′S, 40°13′W), MCZ 21109, 1, 333 mm SL (holotype). Minas Geraes (= Minas Gerais; exact locality un-known, but presumably somewhere in east or southeast of state (Higuchi, 1996: An updated list of ichthyological collecting stations of the Thayer Expedition to Brazil (1865–1866): http://www.oeb.harvard.edu/fish.thayer.htm)), USNM 322019 (formerly MCZ 21105), 1, approximately 175 mm SL. Município de Ferros, Rio Santo Antônio, LIRP 4017, 2, 93.5–131.7 mm SL; LIRP 4018, 1, 134.1 mm SL (cleared and stained); USNM 371470, 1, 112.8 mm SL; MCZ 162217, 1, 123.7 mm SL. Município de São Sebastião do Rio Preto, Rio Santo Antônio, estrada Córrego da Fonseca, approximately 11 km from Santo Antônio, casa do Joaquim Quinquim, MZUSP 73112, 1, 245.8 mm SL; MZUSP 73105, 5, 224.6–254.9 mm SL. Município de São Sebastião do Rio Preto, Rio Santo Antônio basin, Rio Preto do Itambé, MZUFV 2724, 1, 139.4 mm SL; MCP 23271, 1, 215.3 mm SL. Rio Doce basin, Município de Ferros, LIRP 4275, 2, 218.4–227.3 mm SL. Município de Santo Antônio do Rio Abaixo, Rio Santo Antônio, Fazenda Mundinho, 0.6 km below city of Santo Antônio do Rio Abaixo, LIRP 4043, 1, 238.1 mm SL. Município de Santo Antônio de Rio Abaixo, Rio Santo Antônio, Area de Lazer Benedito Martins (prainha) in city of Santo Antônio, MZUSP

TABLE 1. MORPHOMETRIC VALUES FOR EXAMINED SPECIMENS OF *Henochilus wheatlandii*; $n = 24$. Standard length is expressed in millimeters; measurements 1–15 as percentages of Standard Length; 16–21 as percentages of Head Length.

		Mean	Standard Deviation
Standard Length	65.8–254.9	169.8	70.9
1. Greatest body depth	25.0–32.2	29.9	2.0
2. Snout to dorsal fin origin	49.6–56.4	53.6	1.8
3. Length of base of dorsal fin	10.8–12.3	11.5	0.5
4. Posterior terminus of dorsal fin to adipose fin	22.0–27.5	24.9	1.4
5. Posterior terminus of dorsal fin to caudal fin base	33.6–40.5	38.2	1.5
6. Snout to origin of pelvic fin	46.9–52.0	49.2	1.7
7. Snout to anus	58.4–66.6	62.6	2.0
8. Snout to origin of anal fin	66.4–73.8	69.8	2.0
9. Length of base of anal fin	21.9–25.6	23.4	1.3
10. Length of caudal peduncle	10.8–13.3	12.0	0.7
11. Length of longest dorsal fin ray	20.5–24.2	22.1	1.1
12. Length of first pectoral fin ray	17.0–22.0	19.3	1.4
13. Length of first pelvic fin ray	13.8–17.5	15.7	1.1
14. Least depth of caudal peduncle	8.7–9.6	9.3	0.7
15. Head length	19.6–28.4	23.4	2.7
16. Head height	71.6–89.2	80.2	5.1
17. Snout length	28.3–37.0	33.2	2.4
18. Gape width	28.3–37.7	35.0	2.9
19. Orbital diameter	21.7–31.9	26.9	3.0
20. Postorbital head length	39.1–45.7	42.9	2.0
21. Interorbital width	29.4–43.1	37.7	3.5

73118, 3, 65.8–78.2 mm SL. Rio Santo Antônio, MZUFV 2689, 3, 68.4–116.9 mm SL. Município de Santa Rita do Rio do Peixe, Rio do Peixe, Bairro Valentim, road along right margin of river approximately 2 km upriver from that city (19°07′55″S, 43°09′W), MZUSP 75398, 2, 211.5–239.7 mm SL.

Brazil. Espirito Santo: Rio Doce (= Rio Doce between Linhares and Aimorés, approximately 19°37′S, 39°49′W), MCZ 21103, 3, 70.4–82.7 mm SL.

Diagnosis.—As for the genus.

Description.—Morphometrics of examined specimens presented in Table 1. Body moderately deep, increasingly so in larger individuals (Fig. 2). Greatest body depth located approximately at vertical through dorsal-fin origin. Dorsal profile of head distinctly convex from margin of upper lip to vertical through posterior nostril, nearly straight from that point to rear of head. Distinct change in dorsal profile situated at tip of supraoccipital spine. Dorsal profile of body straight to slightly convex from tip of supraoccipital spine to dorsal-fin origin, and nearly straight and slightly posteroventrally angled from posterior of dorsal-fin base to posterior of caudal peduncle. Predorsal portion of body transversely rounded overall, but with slight, obtuse, middorsal ridge anterior to dorsal-fin origin. Postdorsal region of body transversely rounded overall, but slightly flattened middorsally. Ventral profile of head slightly convex anteriorly for short distance posterior of margin of lower lip and then nearly straight to vertical through posterior limit of maxilla. Ventral profile of head and body convex from vertical through posterior limit of maxilla to anal-fin origin; convexity increasing progressively in larger specimens. Prepelvic portion of body obtusely flattened transversely, more so proximate to pelvic-fin insertion. Postpelvic region of body with slightly developed median keel terminating posteriorly at anus. Anal-fin base slightly convex along anterior one-third of fin and then straight to posterior terminus of fin base. Caudal peduncle slightly convex ventrally.

Head distinctly obtusely pointed in lateral profile, more so in smaller specimens with proportionally shallower heads. Mouth terminal. Lower jaw slender relative to that of many characids. Premaxilla relatively long, with ventral margin slightly posteroventrally angled. Premaxillae joined by sutures. Maxilla relatively short and extending posteriorly only to under middle of orbit. Premaxilla and maxilla mobile with respect to, and distinct from, each other in

smaller specimens; ankylosed to each other and difficult to distinguish in larger individuals. Nostrils of each side of head very close but separated by distinct flap; anterior opening circular, posterior opening crescent-shaped. Eye relatively large, proportionally more so in smaller individuals. Adipose eyelid barely evident at anterior and posterior margins of eye in smaller individuals and more developed in larger specimens.

Median frontoparietal fontanel extending from rear of mesethmoid to base of supraoccipital spine. Fontanel moderately developed in cleared and stained specimens, but not externally apparent in larger whole individuals. Infraorbital series complete, with first and, to lesser extent, second infraorbitals distinctly narrow, third through fifth infraorbitals, particularly third, very well developed. Sixth infraorbitals smaller than fifth infraorbital. Ventral portion of first infraorbital and anteroventral portion of second infraorbital laterally overlapping dorsolateral surface of maxilla. All infraorbitals having laterosensory canal segments proximate to orbital rim, with unossified side branches of canal system passing through connective tissue overlying infraorbitals apparent in cleared and stained specimen. Supraorbital elongate, extending between nasal bone and sixth infraorbital and excluding frontal from orbital rim.

Branchiostegal rays 4 [4 rays reported on one side and 5 on other side of holotype by Garman, 1890]. Gill rakers on outermost gill arch well developed and elongate, with 13 on upper limb and 16 on lower limb of cleared and stained specimen [17 reported on lower limb for holotype by Eigenmann and Myers, 1929].

Form and arrangement of dentition in both jaws highly variable ontogenetically. Premaxilla with two rows of teeth. Outer tooth row on premaxilla in specimens of 65.8–78.2 mm SL with eight to 10 typically tricuspid teeth with large central cusp; central cusp distinctly pointed in recently erupted teeth but rounded in worn teeth. Pentacuspid teeth sometimes present in larger specimens within this size range. Specimens of 112.8–134.1 mm SL with nine or 10 tricuspid or pentacuspid teeth (Fig. 3) with central cusp proportionally more developed than in smaller individuals; central cusp distinctly pointed in recently erupted teeth and somewhat rounded in worn teeth. Posterior most cusp on each premaxillary tooth failing to overlap anterior margin of subsequent tooth other than in largest specimen in this size range. Specimens of 224.5–254.9 mm SL with eight to 12 proportionally larger, pentacuspid teeth with large second cusp distinctly rounded. Two smaller pos-
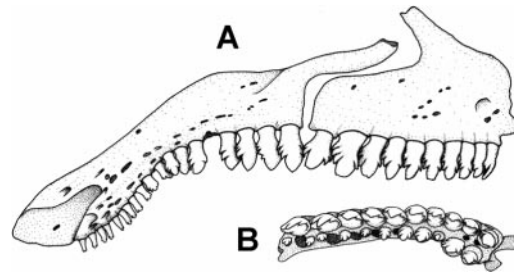

Fig. 3.   *Henochilus wheatlandii*: (A) right premaxilla and maxilla in lateral view, and (B) premaxilla and premaxillary dentition in ventral view; LIRP 4018, 134.1 mm SL.

terior cusps of each tooth overlapping and fitting into depression on, anterolateral surface of basal portion of following tooth.

Teeth of inner premaxillary tooth row in specimens of 65.8–78.2 mm SL with nine or 10 relatively massive, tricuspid teeth. Specimens of 112.8–134.1 mm SL with two larger tricuspid to pentacuspid teeth anteriorly followed by six to nine somewhat smaller, tricuspid teeth (Fig. 3). Inner row of teeth on premaxilla in specimens of 224.5–254.9 mm SL reduced; limited to one or two peglike teeth located proximate to symphysis followed by stiff fleshy ridge situated proximate to medial surface of primary tooth row. Maxilla in specimens of 65.8–78.2 mm SL distinct from, and mobile with respect to, premaxilla; with eight to 12 tricuspid to pentacuspid teeth followed by diastema and then series of six to eight small, conical teeth. Total number of teeth on maxilla 16–21. Specimens of 112.8–134.1 mm SL with maxilla less distinct from, and less mobile with respect to, premaxilla than in smaller individuals; with 10–12 somewhat spatulate, tricuspid or pentacuspid teeth sometimes followed by variably developed diastema and then series of four or five small, conical or elongate, tricuspid teeth (Fig. 3). Specimens of 224.5–254.9 mm SL with maxilla ankylosed to premaxilla and with nine teeth comparable in form to, but smaller than, those on premaxilla, followed in one specimen by diastema and then two small, conical teeth.

Outer row of teeth on each dentary in specimens of 65.8–78.2 mm SL consisting of 10–15 tricuspid to pentacuspid teeth anteriorly followed by three to five smaller, conical teeth. Total number of teeth in series 13–20. Specimens of 112.8–134.1 mm SL with 14–18 broadly tricuspid or pentacuspid teeth followed by two to four elongate, tricuspid teeth. Total number of teeth in series 16–20. Specimens of 224.5–254.9

mm SL with 16–20 tricuspid teeth with large central lobe in outer dentary tooth row.

Inner row of teeth on each dentary in specimens of 65.8–78.2 mm SL with two components; single, relatively robust canine tooth proximate to symphysis and more posteriorly positioned series of one to 10 elongate conical teeth paralleling posterior portion of outer tooth row. Specimens of 112.8–134.1 mm SL with inner row of dentary teeth comparable to that just described other than for having nine to 15 conical teeth in posterior series. Inner row dentary dentition in specimens of 224.5–254.9 mm SL limited to robust conical symphyseal tooth; tooth sometimes eroded dorsally and with irregular functional surface.

Scales cycloid, relatively large, with distinct anastomosis of radii. Lateral line decurved and completely pored from supracleithrum to base of caudal fin. Lateral line scales 46–53 [47 reported for holotype by Garman, 1890]; scales in transverse series from dorsal-fin origin to lateral line eight or nine [eight reported for holotype by Garman, 1890]; scales in transverse series from pelvic-fin origin to lateral line four or five [four or five scales below lateral line to unstated landmark reported for holotype by Garman, 1890]; scales in transverse series from anal-fin origin to lateral line four to six [four or five scales below lateral line to unstated landmark reported for holotype by Garman, 1890]; scales along middorsal line between tip of supraoccipital process and dorsal-fin origin 16 to 22; scales along middorsal line between posterior termination of dorsal fin and adipose fin 13–17; horizontal scale rows around caudal peduncle 13–16.

Dorsal-fin rays ii,9 [11 total rays apparent in radiograph of holotype; count of 12 total rays reported for species by Eigenmann and Myers (1929) erroneous based on our examination of both specimens available to those authors]; anal-fin rays iii,21 to iii,26, or ii,23 (in only one specimen) [iii,23 apparent in radiograph of holotype]. First proximal pterygiophore of dorsal fin supporting two fin rays and a small ossification positioned anterior to first developed ray. Pectoral-fin rays i,11 to i,14 [14 total rays reported for holotype by Garman, 1890]. Pelvic-fin rays i,7 [eight total rays reported for holotype by Garman, 1890]. Principal caudal-fin rays 10/9 [10/9 in radiograph of holotype].

Dorsal-fin margin slightly emarginate to straight, first unbranched ray approximately one-half as long as second unbranched ray; second unbranched and first branched rays subequal and longest in fin. Dorsal fin situated on posterior half of body; fin origin located immediately posterior of vertical through middle of SL and slightly posterior of vertical through pelvic-fin insertion. Longest dimension of adipose fin slightly longer than eye width. Adipose-fin origin located approximately at vertical through posterior terminus of anal fin base. Pectoral-fin profile distinctly acute. Tip of adpressed pectoral fin extending posteriorly to vertical located slightly more than two-thirds of distance between pectoral- and pelvic-fin insertions. Pelvic-fin profile obtusely pointed. Tip of adpressed pelvic fin extending to anus. Ventral margin of anal fin distinctly concave anteriorly and approximately straight for posterior two-thirds. Last unbranched and first branched anal-fin rays longest and subequal, following five or six branched rays rapidly decreasing in length, remaining anal-fin rays very gradually decreasing in length. Caudal fin forked, with lobes pointed. Vertebrae 43(2), 44(10), 45(13), 46 (2) [45].

Coloration in life.—Description based on color slides taken in the field by the third author of a recently collected specimen. Dark coloration on body as described under Coloration in alcohol other than that dark pigmentation on ventral two-thirds of head and body obscured by bright, silvery, guanine layer that covers scales. Guanine layer extending further dorsally on opercle than in case of preserved specimens. Iris silvery. Dark humeral spot apparent but variably obscured by overlying guanine. Dorsal, anal, and particularly pectoral fins slightly reddish.

Coloration in alcohol.—Overall ground color of specimens fixed in formalin ranging from brown to smokey grey. Head with silvery coloration variably present on infraorbitals and up to lower two-thirds of opercle. Some specimens with variably developed dark pigmentation on upper portion of opercle. Iris silvery overall and somewhat yellowish golden in one large specimen. Dorsal portion of head and snout dark, but margin of upper lip distinctly lighter. Body distinctly darker dorsally. Prominent humeral spot beginning immediately above first and second scales of lateral line. Humeral spot in smaller specimens formed solely of deep lying pigmentation. Deep lying humeral pigmentation supplemented by dark surface pigmentation in larger individuals. Specimens up to 136 mm SL with variably developed concentrations of dark chromatophores along posterior margin of scales on dorsal one-half to two-thirds of body. Distribution of such dark pigmentation variable and sometimes forming vertically elongate

cross-hatched patterns that differ between individuals. Body distinctly lighter ventrally in larger available specimens, all of which retain guanine on scales in that region. Deep lying and surface dark pigmentation somewhat more developed on caudal peduncle in larger specimens, but not in form of definite spot.

Dorsal fin with small dark chromatophores scattered over membranes; chromatophores more concentrated distally and forming dusky band along distal portion of fin in individuals of moderate to large sizes. Anal fin with small dark chromatophores scattered over membranes; chromatophores more concentrated distally and forming dusky band along distal portion of fin in individuals of moderate size. Band of dark chromatophores on anal fin variably more obvious in larger individuals; extending to base of fin rays in some specimens but more prominent anteriorly in other individuals. Caudal fin with dark pigmentation on basal portions of all fin rays and along length of dorsal- and ventral-most fin-rays. Distal portions of all caudal-fin rays bordered by black. Pectoral fin with scattered dark chromatophores on fin rays; larger individuals with entire fin darker, particularly laterally. Pelvic fin with scattered dark pigmentation limited to membranes of lateral portion of fin in smaller individuals; pigmentation more broadly distributed in larger specimens in which distal portion of fin variably dusky.

Distribution.—All samples of *H. wheatlandii* collected in recent years originated in the upper Rio Doce basin in the state of Minas Gerais, Brazil. The holotype of *H. wheatlandii* (Garman, 1890) was, however, reportedly collected by the Thayer Expedition at Santa Clara on the Rio Mucuri (= Santa Clara, Município de Nanuque, Minas Gerais, Brazil), an independent river system two basins to the north of the Rio Doce (Fig. 1). Subsequent collecting efforts in the Rio Mucuri basin, some focused on the capture of *H. wheatlandii*, failed to yield specimens of the species either at the type locality or at numerous other sites within that river system. Other Thayer Expedition population samples of *H. wheatlandii*, in turn, either are from indefinite localities in the eastern portions of the state of Minas Gerais (USNM 320019) or originated in the Rio Doce (MCZ 21103), the basin that was the source of all recent samples of the species. Given that all recently collected *Henochilus* specimens originated in the upper reaches of the Rio Doce, a region well separated from the Rio Mucuri, Vieira et al. (2000) suggested that the originally stated type locality for *H. wheatlandii*

might be erroneous or that the species had undergone local extirpation in the Rio Mucuri.

Ecology.—According to Vieira et al. (2000), Vieira and Alves (2001) and F. Di Dario (pers. comm.), the typical habitats occupied by *H. wheatlandii* are 10–20 m wide rivers, 0.5–1.5 m deep with dark colored, slightly acidic water of low nutrient concentration flowing over a sandy bottom interrupted by large boulders forming alternating riffles and eddy pools. The surrounding riparian vegetation was largely undisturbed. In light of the apparent restriction of *H. wheatlandii* to such habitats, Vieira and Alves (2001) proposed that the species requires specialized habitats in terms of stream order, substrate features, and water quality.

Vieira and Alves (2001) reported that *H. wheatlandii* feeds on insects and plant matter. Further observations indicate that the species undergoes dramatic ontogenetic changes in the diet and digestive tract. A 78.2 mm SL specimen (MZUSP 73118) had the stomach and intestine full of whole and fragmented chironomid larvae other than for a small quantity of filamentous algae and sand. A 134.1 mm SL specimen (LIRP 4018) had the intestinal tract largely filled with parts of semisubmerged riparian macrophytes (Poacea and Commelinaceae, genus *Tradescantia*). The stomach contents of a 225.6 mm SL specimen (MZUSP 73105) had numerous seeds of submerged or semisubmerged macrophytes (Podostemacea) that grow on boulders in areas of rapid water flow, along with one coleopteran (Cerambicidae) and one ant (Formicidae). The intestines were full of pieces of leaves and stems of the Podostemacea, confirming observations by the third author that *H. wheatlandii* feeds on submerged macrophytes (Podostemacea).

Associated with these possible dietary shifts in the three specimens that had their digestive system contents examined are ontogenetic modifications of the teeth and relative length of the digestive system. The dentition of smaller *H. wheatlandii* specimens is that typical of characiforms, which are generalized insectivores with pointed multicuspidate teeth, whereas larger individuals have more rounded dentition (Fig. 4). In a 78.2 mm SL specimen (MZUSP 73118), the digestive tract is 1.10 of SL, whereas in a 134.1 mm SL individual (LIRP 4018), it is 2.24 of SL, and in a 225.6 mm SL specimen (MZUSP 73105), it is 3.21 of SL. In contrast, the stomach represents a proportionally smaller percentage of the total length of the digestive tract in larger individuals (21.2% in 78.2 mm SL specimen and 9.7% in 225.6 mm SL individual). The
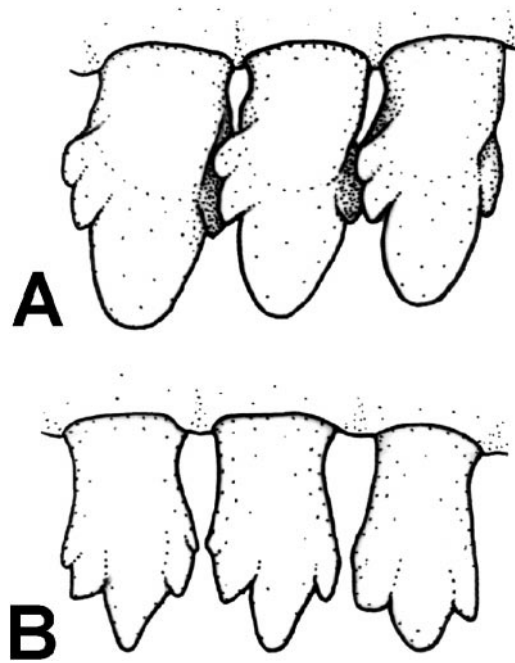
Fig. 4. Fifth through seventh premaxillary teeth of *Henochilus wheatlandii*: (A) 254.9 mm SL specimen (MZUSP 73105); and (B) 65.8 mm SL specimen (MZUSP 73118); drawings not to scale, anterior to right.

changes in the dentition and digestive tract and the likely correlated possible shifts in the diet of *H. wheatlandii*, if they prove consistent in a larger number of specimens examined for dietary studies, are comparable to similar ontogenetic changes present in the Neotropical characiform *Mylesinus paraschomburgkii*; a species that shifts from a predominately insectivorous diet when young to a nearly exclusively herbivorous diet as an adult (Santos et al., 1997; Jégu et al., 1989). Given that both species are dependent on the Podostemacea as adults, it is likely that the building of dams within the range of *H. wheatlandii*, which will eliminate the habitat necessary for that group of plants, would result in the extirpation of *H. wheatlandii* in a mode similar to that forecast by Santos et al., (1997) for *M. paraschomburgkii*.

Remarks.—An evaluation of whether the holotype of *H. wheatlandii* is conspecific with material of the genus from the Rio Doce basin is complicated both by the poor condition of the holotype and the fact that it is distinctly larger (333 mm SL; K. Hartel, MCZ, pers. comm.) than other available specimens of the genus (largest 254.9 mm SL). Data on many meristic features can no longer be taken from the ho-

lotype because of its poor condition, but the counts reported for the holotype by Garman (1890) all fall within the range for such values in the other examined specimens, as do all features amenable to examination via radiography. The holotype of *H. wheatlandii* as originally illustrated by Garman (1890) and subsequently by Eigenmann and Myers (1929) has a distinctly deeper body than that present in all other available samples. *Henochilus wheatlandii* demonstrates, however, a distinct ontogenetic increase in the relative body depth across the range of recently collected specimens (Fig. 2), a trend that would result in a notably deeper body in a specimen the length of the holotype. Furthermore, the holotype had its body cavity opened as evidenced Garman's (1890) detailed description of the gas-bladder and stomach contents. In conjunction with the very soft condition of the holotype, such a dissection would likely exaggerate the apparent relative body depth. The relatively few body measurements that could be accurately taken on the holotype (head length, snout length, orbital diameter, and postorbital head length) all fall along the regression lines for these values across all examined specimens. In the absence of evidence to the contrary, we consequently consider all available specimens of *Henochilus* to be conspecific.

Phylogenetic relationships.—In his description of *Henochilus*, Garman (1890:52) proposed that "the position of the genus is close to the genera *Tetragonopterus* and *Scissor*" but did not elaborate on that suggestion. Subsequent authors proposed a variety of alternative placements for the genus, all necessarily based on information in the original description or from the two Thayer Expedition specimens given the lack, until recently, of additional material of the genus. Eigenmann and Myers (1929:454) considered *Henochilus* to be "an aberrant member of the Tetragonopterinae" and tentatively proposed that it might be aligned with the genus *Psalidodon*. Géry (1977) shifted both of those genera into the Cheirodontinae and united them in the tribe Henochilini without, however, being explicit as to the basis for such an association. Most recently Malabarba (1998) removed *Henochilus* from the Cheirodontinae as delimited in his paper and left that genus *insertae sedis* in the Characidae.

Molecular information indicates a close phylogenetic relationship of *Henochilus* with the characid genus *Brycon*. The combined sequences of the 12S and 16S rRNA mitochondrial data of the 43 taxa in the analysis resulted in 872 characters, of which 461 were conserved, 308
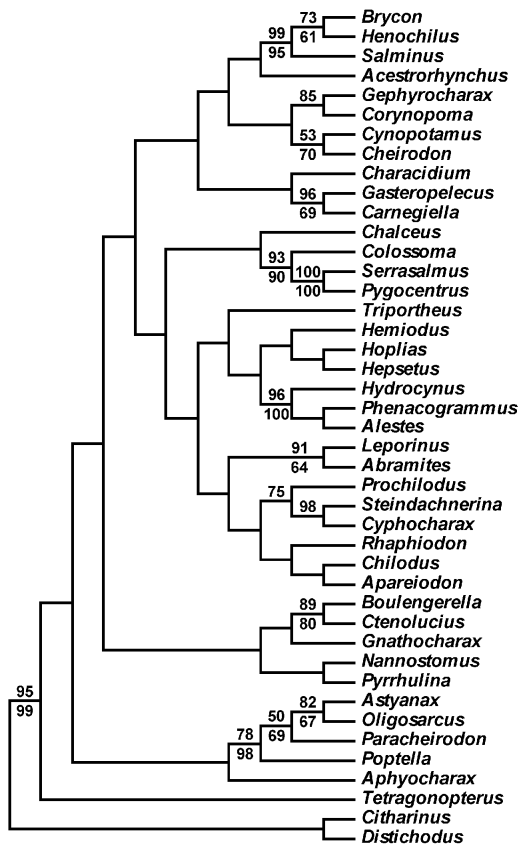
Fig. 5. Strict consensus MP tree of five equally parcimonious trees obtained when gaps were considered as missing data and Ti:Tv ratio was 1:1 (TL = 2392, CI = 0.28, RI = 0.36). Numbers above branches are bootstrap values from 1000 replicates. Numbers below branches are bootstrap values based on 100 replicates in the ML analysis. Bootstrap values below 50% are not shown.

were parsimony-informative and 103 were parsimony-uninformative. The MP heuristic searches using all sites with equal weight and treating alignment gaps as missing resulted in five equally shortest trees with tree lengths of 2372 steps, a consistency index of 0.28, and a retention index of 0.36 (Fig. 5). Weighting transversion twice transitions or excluding ambiguous positions in the original alignment resulted in different tree topologies (data not shown); however, in all of those trees *Henochilus* was placed as the sister group of *Brycon*. ML analysis conducted with PAUP* resulted in a consensus phylogeny similar to those obtained in the MP analyses (−Ln likelihood = 10580.13). The groups supported at least 50% in the 100 bootstrap replicates in the ML analyses were almost the same supported in the MP analysis (Fig. 5). Extensive

molecular studies about mutation rates in the 12S and 16S rRNA genes conducted by Ortí and coworkers and summarized in Ortí (1997) indicate that saturation may be a problem beyond the family level in Characiformes, a factor that was also observed in the present analysis. However, these genes contain sufficient information to access the relationship at the generic level (Ortí, 1997), which was confirmed in the present study since all phylogenies obtained showed high bootstrap values supporting several of the intergeneric nodes, most notably the clade (*Salminus*, (*Brycon*, *Henochilus*)) (Fig. 5). An evaluation of the phylogenetic placement of *Henochilus* based on morphological data is encumbered by the uncertainty that surrounds the composition of the family Characidae and its subunits. *Henochilus* is assignable to the Characidae under the concept of that family advanced by Buckup (1998), which is more restrictive than that of the family proposed by Greenwood et al. (1966); however, as pointed out by Weitzman and Malabarba (1998) the limits of that family and its monophyly are questionable. As noted, *Henochilus* was previously assigned to two of the major subunits within the Characidae, the Tetragonopterinae and Cheirodontinae. Malabarba (1998) demonstrated that the relationships of *Henochilus* lie outside of the Cheirodontinae as delimited by that author. *Henochilus*, in turn, possesses a supraorbital, an autogenous ossification situated along the dorsal surface of the orbit, which is typically absent in tetragonopterine characids (Weitzman and Malabarba, 1998), and there are no known derived attributes of the genus shared with the members of the Tetragonopterinae. As such an alignment of *Henochilus* with either the Cheirodontinae or Tetragonopterinae is not supported by the available data. Looking to the various other subfamilies recognized within the Characidae by Weitzman and Malabarba (1998), we find that *Henochilus* lacks the characters that diagnose the Agoniatinae, Aphyocharacinae, Characinae, Glandulocaudinae, Paragoniatinae, Rhoadsiinae, and Stethaprioninae but that *Henochilus* does possess many of the attributes of the Bryconinae, that Weitzman and Malabarba (1998) noted may also be a component of the Characidae.

The absence of a published phylogenetic treatment of the Bryconinae makes it impossible to advance a definitive morphologically based evaluation of the phylogenetic position of *Henochilus* within that subfamily. Indeed the limits of the Bryconinae are indefinite (Weitzman and Malabarba, 1998), but the subfamily as delimited by Géry (1977) and Géry and de Rham

(1981) included *Brycon, Catabasis, Chalceus, Chilobrycon, Salminus*, and *Triportheus. Henochilus* lacks a supramaxilla, an autogenous ossification unique to *Chalceus* in the Characidae and also lacks the distinctly expanded and transversely expanded pectoral ventral portion of the pectoral girdle that characterizes the species of *Triportheus*. We find that *Henochilus* possesses some of the attributes identified by Howes (1982) as characterizing *Brycon* (including *Holobrycon* according to that author) including the presence of two symphyseal teeth in the lower jaw and premaxillae that are linked via a convoluted symphyseal joint. However, *Henochilus* shares a distinctive overall form of the jaws, lips, and dentition with *Chilobrycon*. The two genera differ, however, in the number of rows of teeth on the premaxilla (Géry and de Rham, 1981), with *Chilobrycon* sharing the presence of three rows of teeth in the upper jaw with *Brycon*, a feature, however, which has not been demonstrated to be derived. A resolution of these apparently contradictory alignments of *Henochilus* with *Brycon* on the one hand and *Chilobrycon* on the other lies beyond the scope of this study, but the evidence indicates that the relationships of *Henochilus* lie within the Bryconinae, most likely with *Brycon* and *Chilobrycon*; a finding that is congruent with the results of the molecular analysis.

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