Leptophilypnion, a new genus with two new species of tiny central Amazonian gobioid fishes (Teleostei, Eleotridae)

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Abstract

Leptophilypnion pusillus new genus and species is described from three specimens, 8.4-9.1 mm standard length, collected in the central lowland Amazon basin at a single locality near Santarém. A gravid female 9.1 mm standard length has 5-6 oval eggs 0.5 mm long. The new fish is distinguished readily from the only other previously described Amazonian eleotrids, the three species of the genus *Microphilpnus* Myers 1927, by adults having head and body anterior to second dorsal fin scaleless; two innermost rays of each pelvic fin simple and filamentous, extending to or beyond anal fin origin; head relatively large and snout truncate or blunt (vs. head smaller and snout relatively pointed); body and caudal peduncle relatively short and deep instead of slender; and only 5 instead of 6 branchiostegal rays. A second species of the new genus, *L. fittkaui*, is described from 10 specimens 8.9-9.7 mm from a locality near Manaus.

Leptophilypnion is perhaps most closely related to the Central American freshwater eleotrid genus *Leptophilypnus* Meek and Hildebrand 1916, particularly the species *L. guatemalensis*, with which *L. pusillus* agrees in having the unusual dorsal fin spine pterygiophore to neural spine insertion formula of 3-12201 and 10+16=26 vertebrae. *L. fittkaui*, a somewhat stouter species, has 3-121110 and 11+15=26.

Zusammenfassung

Die neue Gattung und die neue Art *Leptophilypnion pusillus* werden auf der Grundlage von drei Exemplaren mit 8,4 bis 9,1 mm Standardlänge beschrieben, die im zentralen Tieflandbecken des Amazonas an einer einzigen Lokalität nahe Santarém gefangen wurden. Ein trächtiges Weibchen mit 9,1 mm Standardlänge hatte 5 bis 6 Eier, die 0,5 mm lang waren. Die Vertreter der neuen Fischart unterscheiden sich auffällig von den einzigen bisher beschriebenen Eleotriden Amazoniens, den drei Arten der Gattung *Microphilypnus* Myers 1927: Kopf und Rumpf von vorne bis zur zweiten Rückenflosse ohne Schuppen; die beiden innersten Flossenstrahlen jeder Bauchflosse einfach und fädig, wobei sie sich bis zum Afterflossenansatz oder darüber hinaus erstrecken; relativ großer Kopf, Schnauze wie gestutzt oder stumpf (im Gegensatz zur kleineren und relativ spitzen Schnauze bei den anderen Arten); Rumpf und Schwanzstiel relativ kurz, Schnauze tief, und nicht schlank; nur fünf, nicht sechs Kiemenbogen. Außerdem wird anhand von zehn Exemplaren mit 8,9 bis 9,7 mm Länge eine weitere neue Art: *L. fittkaui,* von einer Lokalität bei Manaus beschrieben.

Leptophilypnion ist wohl am meisten verwandt mit der zentralamerikanischen Süßwasser-Schläfergrundelgattung *Leptophilypnus* Meek and Hildebrand 1916, insbesondere der Art *L. guatemalensis;* mit ihr stimmt *L. pusillus* durch die ungewöhnliche Formel 3-12201 überein, die sich auf die Pterygioophoren der Rückenflosse bis hin zum Ansatz des Neuralstachels bezieht, und durch die Wirbelzahl 10+16=26. Bei den Vertretern von *L. fittkaui* mit ihrer etwas mehr gedrungenen Gestalt lauten die Formeln 3-121110 bzw. 11+15=26.

Résumé

Leptophilypnion pusillus, nouveau genre, nouvelle espèce, est décrit sur base de trois spécimens, 8,4-9,1 de LS, collectés dans le bassin inférieur central de l'Amazone, à un seul endroit près de Santarém. Une femelle gravide, de 9,1 mm de LS, porte 5-6 œufs ovales de 0,5 mm de long. Ce nouveau poisson se distingue aisément des seuls autres Eléotridés amazoniens décrits jusqu'ici, les trois espèces du genre *Microphilypnus* Myers 1927, par le fait que les adultes ont la tête et la partie antérieure du corps jusqu'à la seconde dorsale sans écailles, les deux rayons les plus intérieurs de chaque pelvienne sont simples et filamenteux, s'étendant jusqu'à la base de l'anale ou au-delà, une tête assez grande et un rostre tronqué ou arrondi (contre une tête plus petite et un rostre plutôt pointu); le corps et le pédoncule caudal plutôt courts et gros au lieu d'élancés et n'ayant que 5 rayons branchiostégaux au lieu de 6. Une deuxième espèce du nouveau genre, L. fittkaui est décrite sur base de 10 spécimens, de 8,9-9,7 mm, d'une localité près de Manaus.

Leptophilypnion est peut-être le plus proche du genre d'Eléotridés d'eau douce d'Amérique Centrale, *Leptophilypnus* Meek et Hildebrand 1916, surtout l'espèce *L. guatema-*

lensis avec laquelle *L. pusillus* partage l'inhabituelle formule d'insertion de l'épine dorsale ptérygiophore avec l'épine neurale de 3-12201 et 10+16=26 vertèbres. *L. fittkaui,* une espèce un peu plus volumineuse, fait 3-121110 et 11+15=26.

Sommario

Leptophilypnion pusillus nuovo genere e nuova specie sono descritti da tre esemplari di 8.4-9.1 mm (lunghezza standard) raccolti nella pianura centrale del bacino amazzonico in una singola località vicino a Santarém. Una femmina di 9.1 mm di lunghezza standard racchiudeva 5-6 uova ovali lunghe 0.5 mm. La nuova specie si distingue facilmente dagli unici altri eleotridi amazzonici descritti in precedenza, le tre specie del genere *Microphilpnus* Myers 1927, poiché gli adulti hanno la testa e la parte anteriore del corpo fino alla seconda pinna dorsale senza scaglie; i due raggi più interni di ogni pinna pelvica semplici e filamentosi, che si estendono fino o al di là dell'origine della pinna anale; testa relativamente grande e muso troncato o smussato (contro la testa piccola e muso relativamente appuntito); corpo e peduncolo caudale relativamente breve e robusto invece di snello, e solo 5 anziché 6 raggi branchiostegi. Una seconda specie, *L. fittkaui,* è descritta da 10 esemplari di 8.9-9.7 mm da una località vicino a Manaus.

Leptophilypnion è forse filogeneticamente più vicino al genere di acqua dolce del centro America *Leptophilypnus* Meek e Hildebrand 1916, in particolare alla specie *L. guatemalensis,* con cui *L. pusillus* condivide l'insolita formula d'inserzione degli pterigiofori sulle spine dorsali di 3-12201 e per avere 10 +16 = 26 vertebre. *L. fittkaui,* una specie un po' più robusta, ha una formula di 3-121110 e 11 +15 = 26 vertebre.

INTRODUCTION

In 1986 Sven O. Kullander of the Naturhistorisk Riksmuseet (NRM) in Stockholm brought to my attention three specimens of a tiny goby he collected in 1980 in the lower Rio Tapajós in the Amazon basin near Santarém. The largest, 9.1 mm standard length, is a ripe female with 5- 6 oval eggs 0.5 mm long. Repeated efforts by Kullander and his colleagues to collect additional specimens at the type locality and further up the Tapajós from 1989 through 1994 were unsuccessful, the only gobies obtained being *Microphilypnus*. Specimens of a closely related species were obtained by the late German dipterist Ernst J. Fittkau while collecting chironomid larvae in the lower part of the Rio Negro river system near Manaus in 1960-1962. These were deposited in the fish collection of the Zoologisch Staatssamlung München (ZSM). The type locality of the first species has in more recent years been completely degraded for many miles around. Searches for additional material in the fish collections of the Museu de Zoologia da Universidade de São Paulo, Museu Paraense Emilio Goeldi in Belem, and Instituto Nacional de Pesquisas da Amazônia in Manaus during the author's lengthy visits to these institutions in 2008 were unsuccessful. These institutions have numerous specimens of small Amazonian eleotrids, many of them less than 10 mm standard length, but apparently all are *Microphilypnus*.

The presence of only five instead of six branchiostegal rays initially led to the supposition that this new gobioid might belong in the family Gobiidae (usually with five branchiostegal rays) instead of Eleotridae (usually with six). However, the pelvic fins are widely separated at the base, as usual in eleotrids, and there is no vestigial median connecting membrane between them, as seen in Gobiidae with secondarily separated pelvic fins. The caudal fin skeleton has two epurals, more common in Eleotridae than in Gobiidae. Thus the two new fish species described here are interpreted as eleotrids with only five branchiostegal rays due to reduction associated with their tiny size. This hypothesis is supported by the finding they they may be most closely related to the eleotrid genus *Leptophilypnus* (although this should be regarded as an hypothesis of relationships that might not stand up to further investigation).

The only eleotrids hitherto known from the interior of the Amazon basin belong to the genus *Microphilypnus* Myers 1927, and also are quite small. Three species are recognized now, *Microphilypnus ternetzi* Myers 1927 (type species of the genus); *M. macrostoma* Myers 1927; and *M. acangaquara* Caires & Figuierdo 2011 (a fourth nominal species, *M. amazonicus* Myers 1927, has been placed as a synonym of *M. terrnetzi)* (Caires & Figuierdo, 2011). The maximum standard length of *Microphilypnus* is 18-25 mm; the body is elongate or fusiform, and the head pointed or markedly less truncate. Adults usually have scales on the entire sides of the body, on top of the head, and on the gill cover. Juvenile *Microphilypnus* may lack scales on top of the head and on the anteriormost part of the body, and some adults of *M. ternetzi* and *M. acangaquara* lack scales on top of the head (Caires & Figueiredo, 2011). Sensory papillae occur in rows on the top and sides of the head and inside the lower jaw. The pelvic fins are non-filamentous, the second and third rays being longest, and the innermost two pelvic rays much shorter, so that the tips of the longest pelvic fin rays fall far short of the anal fin origin, rather than reaching to or beyond it.

Initially it seemed that specimens of the new genus collected near Santarém and near Manaus were conspecific. Further study, especially of specimens cleared and stained for osteological observations, indicated that they represent distinct species, both named here.

MATERIALS AND METHODS

Study material of the two new species is listed as type material in the species accounts. Information provided includes institutional catalog numbers, standard lengths in mm, and all available locality data including name of collector and date of collection. Type material is deposited in the Museu de Zoología of the Universidade de São Paulo, MZUSP; Naturhistoriska Riksmuseet, Stockholm, NRM; and Zoologische Staatsumlung, Munich, ZSM. Measurements and counts follow Hubbs & Lagler (1949), Pezold & Cage (2002) and Caires & Figueiredo (2011) unless otherwise indicated.

The holotypes of the new species were photographed in 2012 and 2013. They were mounted under water in a petri dish with Silgard mounting medium and photographed using a a Leica binocular digital microscope which takes images at different distances and combines them into a single image with greater depth of field. The holotype of *L. pusillus* was drawn in 1986 using a Bausch and Lomb binocular dissecting microscope with drawing tube. The drawing was revised in 2013, after re-examination and photography of the holotype.

Much of the information in the following taxonomic accounts comes from three specimens cleared and stained with alizarin red S and methylene blue. None of the preparations were entirely satisfactory, making uniform comparison and more complete drawing of osteological characters impossible. A paratype of *L. pusillus* was cleared and stained with alizarin and methylene blue, and the resulting osteological preparation drawn with the same microscope and drawing tube in 1986. Osteological specimens were mounted with *minuten nadeln* under water or a mixture of glycerine and water in a petri dish with Silgard mounting medium. The cleared and stained material of this specimen, prepared while it was still relatively fresh, retained its stain for at least one year, but by 2012 it was completely destained. In 2012 a paratype of *L. fittkaui* was similarly cleared and stained, but with unsatisfactory results. The specimen partially disintegrated, and the parts that remained intact retained stain only for two days, just long enough for some observations to be recorded but not long enough to make drawings. No photographs were taken of any of the osteological preparations. Attempts to clear and stain additional specimens failed. Efforts to radiograph specimens for counts of vertebrae, fin rays, and pterygiophores also were unsuccessful.

Around 1993 Peter J. Miller received some specimens of *L. fittkaui* from the same collection as the type specimens. He observed and drew sensory papillae on the head based on several specimens, and cleared and stained one specimen, from which he made drawings of the palatoquadratohyomandibular arch and branchiostegal bar and of the caudal fin skeleton. The author did not examine this cleared and stained specimen. Miller's drawings are reproduced here, with his permission. The present whereabouts of these specimens are unknown, and they are not included in the type material.

Available morphometric and meristic data for the best-studied specimens of *Leptophilypnion*, including both holotypes, are presented in Table I. Most of the measurements of the holotype of *L. pusillus* were taken from the drawing of it (Figure 2), while those of the holotype of *L. fittkaui* were taken much later from photographs, so they might not be strictly comparable. For this reason and also because they are based on so few specimens, the proportional measurements have not been incorporated directly into the species descriptions.

While alizarin generally stains bones red, and methylene blue stains cartilage blue, this distinction is not always clear-cut in fishes, especially in larvae and paedomorphic or very small species. A skeletal element stained red or rosy with alizarin probably can be confidently identified as bone, but it cannot be assumed that an element lightly stained with methylene blue is cartilage. In some instances membrane bones such as the opercle pick up methylene blue while at the same time failing to pick up alizarin.

Leptophilypnion n. gen.

Type species: *Leptophilypnion pusillus.*

Diagnosis: *Leptophilypnion* differs from all other eleotrids in its extremely small size (all known specimens less than 10 mm standard length) and in having only 5 instead of 6 branchiostegal rays. Pectoral fin rays attached to a single non-calcified radial

plate with three fenestra (larvae of perhaps all other gobioid taxa have similar plates, but in adults of all other gobioids this plate has developed largely or entirely into four separate pectoral ossifications or radials that are calcified and stain with alizarin. It further differs from *Microphilypnus*, the only other genus of Eleotridae known from the interior of the Amazon basin, in having a relatively stout head and body; blunt snout; scales absent on head and anterior third of body; a highly distinctive insertion pattern of first dorsal fin pterygiophores to interneural spines; pelvic fins filamentous, last two (innermost rays) of each fin nearly twice as long as others, and extending well beyond anal fin origin when adpressed to body; and vertebrae 10+16=26 or 11+15=26 (vs. 11-13+14-17=26-29).

The relationship of interneural spines to first dorsal fin spine pterygiophores in *Leptophilypnion*, using the system developed by Ray S. Birdsong (Birdsong, 1975; Birdsong, et al., 1988), is 3-12201 in one species and 3-121110 in a second species. The 3-12201 pattern has not been reported in *Microphilypnus*. It has been reported previously only in the elongate gobiid genus *Gobioides* (Birdsong et al., 1988: 195-196; Birdsong & Robbins, 1995: 682) and in some specimens of the eleotrid species *Leptophilypnus guatemalensis* Thacker and Pezold in Thacker et al. (2006). The 3-121110 pattern occurs in *Microphilypnus* (Caires & Figueiredo, 2011) and in the gobiid genus *Evermannichthys* (Birdsong et al., 1988). The number of spines in the first dorsal fin of *Microphilypnus* varies from 3 to 7 (Caires & Figueiredo, 2011: 40), but the usual number is 6, as in *Leptophilypnion* and the great majority of gobioids. In *Microphilypnus* with 6 spines the only other reported relationships of neural spines to first dorsal fin pterygiophores reported are 3-22110, 3-12210 and 3-21210 (Birdsong et al., 1988: 202; Caires & Figueiredo, 2011: 40). The first two of these occur in numerous genera of eleotrids and gobiids, while 3-21210 in otherwise known only in the gobiid genera *Oligolepis* and *Tridentiger*. No "interneural gap" (Birdsong et al., 1988: 177, fig. 2A) occurs between the first and second dorsal fins. In *Leptophilypnion* insertion of second dorsal fin pterygiophores to neural spines is 111212.

Cephalic laterosensory canals, as in gobioids generally, are present in *Leptophilypnus*, *Microphilypnus*, and *Leptophilypnion*. *Leptophilypnion* has a supraorbital canal with no pores, and no preopercular canal. Its cephalic laterosensory canal system is among the most reduced observed in eleotrids. *Leptophilypnus fluviatilis* and *L. panamensis* have supraorbital and preopercular canals with at least two supraorbital pores and two or three preopercular pores. *Microphilypnus acangaquara* has a supraorbital canal with a single large posterior pore and a preopercular canal with large upper and lower pores (Caires & Figueiredo, 2011: fig. 11). *Leptophilypnus guatemalensis* has a supraorbital canal with two pores and lacks a preopercular canal (Thacker et al., 2006: fig. 2B).

Cephalic neuromasts usually are readily visible in gobiids and eleotrids including *Microphilypnus* and *Leptophilypnus* and their pattern may be helpful in recognition of taxa and their classification. They are, however, either not developed or have not preserved well in the holotype and two paratypes of *L. pusillus* from Santarém (and thus are not illustrated in the figure of the holotype (Fig. 1). They are, however, readily visible in some of the specimens of *L. fittkaui,* particularly in ZSM 28290, 9.2 mm. Peter J. Miller examined several of the specimens from near Manaus and made a composite diagrammatic drawing of their cephalic neuromasts, which he has kindly given permission to reproduce here. His drawing also shows the posterior nostril, which I was unable to observe in any of the specimens. The only fine features detected on the head when the holotype was drawn were melanophores, indicated as dots in my drawing. Similar melanophores are present on the fins and on the posterior margins of the scales on the body.

Compared to the cephalic neuromasts of *Microphilypnus*, *Leptophilypnus*, and other eleotrids, those of *Leptophilypnion* are much fewer and less organized. In species of *Leptophilypnus* and *Microphilypnus* neuromasts on the cheek and preopercle are numerous close-set organs forming continuous horizontal and vertical intersecting rows (Thacker et al., 2006, fig. 2A-D; Caires & Figueiredo, 2011, figs 4, 7, 8, 11). *Leptophilypnus* have the inframandibular main horizontal row of neuromast organs with numerous short vertical branches along all or part of its length. *Microphilypnus* and *Leptophilypnion* have a simple inframandibular row of neuromasts organs (without short vertical rows). This similarity presumably relates more to size reduction in the two genera rather than phyletic relationship. *Leptophilypnion* has a close grouping of a half-dozen neuromasts organs on the chin, where they have not been reported or illustrated in *Microphilypnus* or *Lepto-*

Table I. *Leptophilypnion.* Standard length, measurements (as times in standard length), and counts. DFPN = relationship of first dorsal fin pterygiophores to neural spines).

	L. pusillus			*L. fittkaui*	
	holotype	**paratype**	**paratype**	**holotype**	**paratype**
standard length (mm)	9.1	9.0	8.4	9.2	9.5
proportional measurements					
head length	3.3	3.7	3.2	3.4	
head width	7.0		5.6	6.9	
eye diameter	10.3	9.7	9.8	9.5	
interborbital	66.5		32.0	27.5	
snout	17.3	19.3	15.8	18.3	
upper jaw	13.0	12.1	8.0		
lower jaw	9.0	7.9	6.0		
body depth	4.6		4.6	6.9	
body width	7.2		6.7	8.5	
predorsal length	2.6	2.4	2.4	3.8	
prepelvic length	3.2	3.3	3.0	4.8	
preanal length	1.7	1.7	1.7	2.7	
dorsal fin spine	4.5	4.6	5.4		
pectoral fin length	5.2		6.5		
pectoral fin base	12.9	12.9	10.9	16.2	
pelvic fin spine	7.0		6.7		
pelvic fin length	3.1	3.4	3.3	6.5	
interpelvic width	28.8		25.5	18.3	
anal fin length	4.3		4.4	4.5	
anal fin base	6.7	8.9		6.9	
caudal peduncle depth	9.3	9.8	8.9	12.0	
caudal peduncle length	3.7	3.1	3.9	5.0	
counts					
lateral scale rows	18		?		
transverse scale rows	5		5		
circumpeduncular scales	10		10		
first dorsal fin	vi	vi	vi		
DFPN		3-12201			3-121110
second dorsal fin	i7	i7			
anal fin	i6	i7?			
pectoral fin	13/13	13/13			
pelvic fin	i5/5i	i5/5i			
caudal fin	6/5	8,8/6,10			
branchiostegal rays		1.3.1/1.3.1			
gill rakers		0+8/8+0			
epurals		2			
vertebrae		10+16=26			11+15=26

philypnus. The distribution of cephalic neuromast organs in *Leptophilypnion* could be derived by reduction either from that of *Microphilypnus* or *Leptophilypnus*, so it seemingly offers little information relevant to determining relationships among these taxa.

The *Leptophilypnion* branchial skeleton has generalized gobioid morphology. Fully formed gill rakers, only present on leading edge of first ceratobranchial, are 8 (from one cleared and stained *L. pusillus)* or 9 (from one cleared and stained *L. fittkaui)*. Rakers on trailing edge of ceratobranchial 1 and on leading and trailing edges of ceratobranchials 2-3 and leading edge of ceratobranchial 4 irregularly and weakly developed. Fifth ceratobranchial with no gill rakers, teeth relatively few and tiny, except for enlarged teeth clustered towards middle of trailing edge. Basibranchials 1-3, usually

separate in gobies, combined as a single elongate rod. Basibranchial 4 small and separate.

The caudal fin skeleton is of generalized gobioid morphology, with two epurals, fused hypurals 1 plus 2 and 3 plus 4, and a small separate hypural 5. Large dorsal and ventral procurrent cartilages support most of the unsegmented procurrent caudal fin rays, 8/10 in the cleared and stained specimen from Santarém. The first two lower procurrent rays are supported by the hemal spine of the last fully formed vertebra. Principal caudal fin rays are 8/6=14. *Microphilypnus* has only 12-13 principal caudal fin rays (Caires & Figueiredo, 2011: 49), *Leptophilypnus* 13-15 (Thacker et al., 2006: 490).

There is no separate parhypural in the caudal fin skeleton of the only cleared and stained specimens of *L. pusillus;* perhaps it has fused with hypurals 1 and 2. The single cleared and stained specimen of *L. fittkaui*, on the other hand, has a well-developed separate parhypural.

Etymology: *Leptophilypnion* is formed from the eleotrid genus *Leptophilypnus* Meek and Hildbrand 1916 to which this new genus seems to be related, and the Greek diminutive *-ion*. *Leptophilypnus* is masculine, and *Leptophilypnion* also is masculine.

Leptophilypnion pusillus n. sp.

Figs 1-8

Holotype: MZUSP 40297, 9.1 mm, mature female, right bank of lower part of igarapé tributary to Rio Tapajós bordering city of Santarém, Sven O. Kullander, 27 Sep 1980.

Paratypes: NRM 23245, 8.4 mm, male?, collected with holotype; NRM 60133, 9.0 mm mature female, collected with holotype (cleared and stained with alcian and alizarin; only cranium, pectoral girdles and axial skeleton, completely destained by September 2012.

Diagnosis: Minute stout-bodied eleotrids with large ctenoid scales confined to posterior three-quarters of body (commencing after end of first dorsal fin but before origin of second dorsal fin), and no cycloid scales; head and body anterior to origin of second dorsal fin scaleless. Side of body with 5 scales in transverse scale row and 18 scales in midlateral series; circumpeduncular scales 10. No scales on exposed portion of fin rays at base of median and paired fins including caudal fin. Branchiostegal rays 5. Innermost two pelvic fin rays filamentous, extending posteriorly to or beyond anal fin origin. 3-12201 first dorsal fin pterygiophore insertion pattern. Vertebrae 10+16=26. Second dorsal fin and anal fin pterygiophores 7 and 8. No parhypural.

Description: First dorsal fin with 6 slender spines, first two longest and extending posteriorly when fin is depressed to beyond origin of second dorsal fin, last spine very small and widely separated from others, apparently attached by membrane to preceding spine of first dorsal fin but not to first ray of second dorsal fin. Second dorsal and anal fins with a thin flexible spine followed by 7 and 6 soft rays, respectively. Pectoral fin with 13 soft rays. Pectoral fin rays supported by a single cartilaginous plate with three foramina (Fig. 3). Pelvic fins rays i5; innermost two rays filamentous, extending posteriorly to beyond anal fin origin. Pelvic fins widely separate, without frenum or membrane connecting innermost rays of left and right fins. Caudal fin with slightly rounded posterior margin, with 8 upper principal rays articulating with upper hypural plate, and 6 lower procurrent rays articulating with lower hypural plate; eight upper and 10 lower procurrent caudal rays (latter counts obtained only for cleared and stained paratype of *L. pusillus).*

Head and body laterally compressed; eyes large, lateral in position, not projecting above dorsal profile of head; Cephalic laterosensory pores apparently absent. Snout and jaws relatively short; upper jaw extending to below or slightly beyond anterior margin of eye, lower jaw to below middle of eye. Upper and lower jaws with one or two rows of close-set uniformly small canine teeth with sharp, recurved tips. Outer tooth row of premaxilla with about 22 teeth, inner row with somewhat fewer teeth. Leading edge of lower limb of first gill arch with 8 elongate gill rakers on each side in single cleared and stained specimen. Upper limb of first gill arch without gill rakers.

Despite repeated searching with a good microscope and excellent lighting in 1986, when it was still relatively fresh, I could not detect laterosensory pores or neuromast papillae on the head of the holotype or other specimens of *L. pusillus*. The posterior nostril also could not be seen, although it presumably is present. Neither could any of these structures be detected when the holotype was observed and photographed with a Leica digital microscope in October 2012. Failure to detect these structures might be due to poor condition of the preserved material. On the other hand, the posterior nostril and sensory papillae were readily

observed in 1990 and 2012 in specimens of *L. fitt-kaui* that had been collected in the early 1960's and were also not in a very good state of preservation.

Ctenii large, strong, and spine-like, a single row of 4-9 on posterior margin of each scale. For scale counts see diagnosis of species above.

After 11 years in preservative (in 1991), 9.1 mm holotype (female) and 8.4 paratype (sex undetermined) were uniformly tan. A distinctive pattern of melanophore distribution present on head, body, and fins. Largest melanophores, on head, exhibit a vaguely radial pattern from eye onto middle of jaws, opercle, and occiput. Melanophores on margin of scales near base of ctenii. Some large melanophores concentrated along bases of dorsal and anal fins. Dorsal, anal, pectoral and caudal fins with melanophores arranged in several vertical rows. Pelvic fin with a few large melanophores near base, otherwise without melanophores. Anteriormost upper and lower procurrent rays of caudal fin with a half dozen or more melanophores forming small, diametrically opposed elliptical marks on dorsal and ventral margin of caudal peduncle. A few large, dilated melanophores on dorsal surface

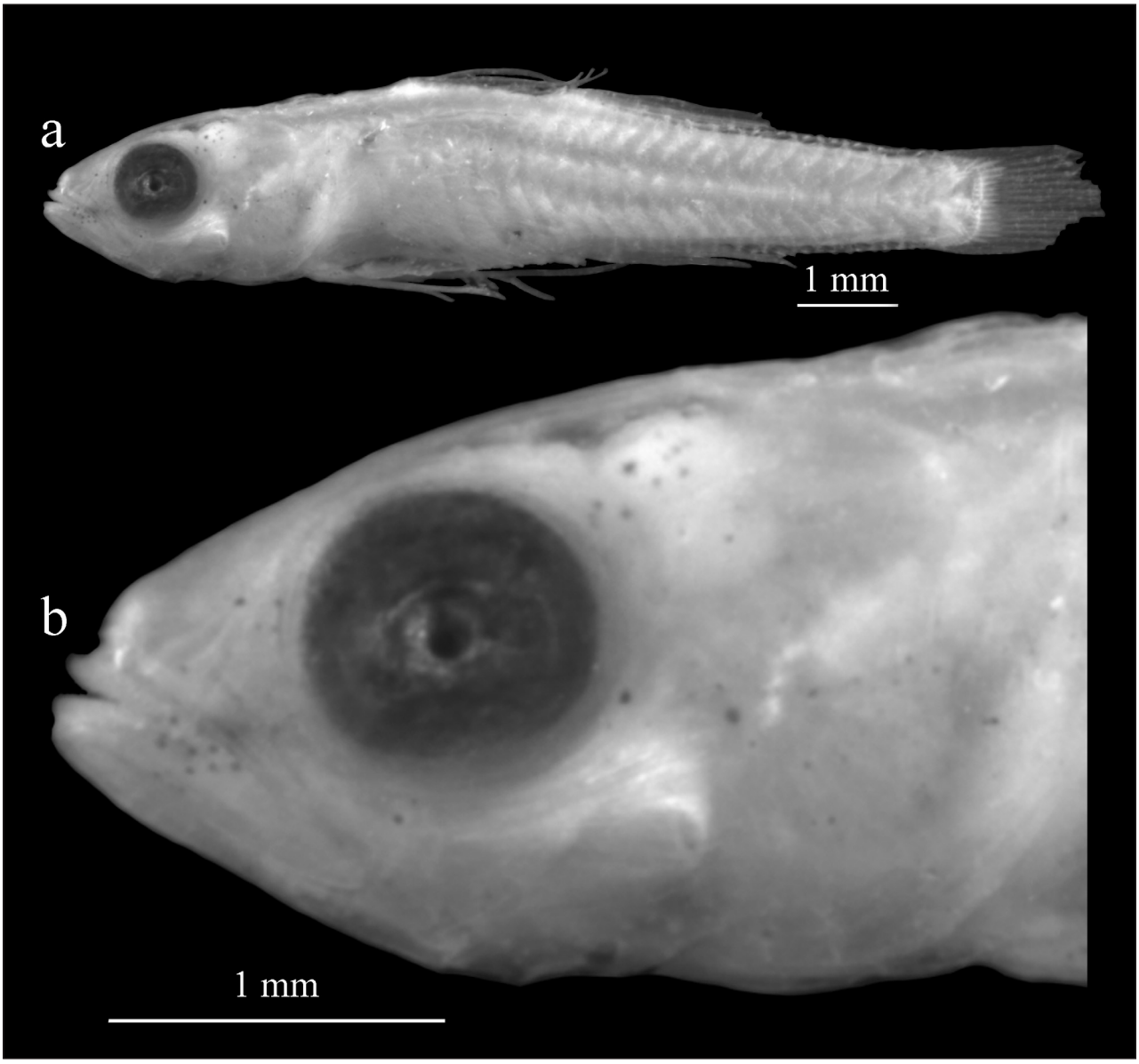

Fig. 1a-b. *Leptophilypnion pusillus*, holotype, 9.1 mm, female. **a**. whole specimen in lateral view; **b**. head in lateral view. Photo by S. O. Kullander.

of head overlying enlarged portion of brain.

Five or six ovarian eggs observed by transmitted light in posterior part of abdominal cavity of 9.1 mm holotype. A single egg removed and measured, the others left in situ. Egg distinctly oval, 0.50 mm long, with tough chorion, large clear peridermal or perivitteline space, and opaque pale nearly spherical yolk 0.28 mm diameter. The 9.0 mm cleared and stained paratype with a single egg in posterior part of abdominal cavity, only the yolk, 0.24 mm diameter, remaining. Sex of 8.4 mm paratype undetermined; its somewhat larger jaws suggest it may be a male. Each specimen with a small, fleshy genital papilla.

Etymology: The species or trivial name *pusillus* (masc.) is Latin, meaning very small or insignificant.

Notes on type locality: The holotype and two paratypes of *L. pusillus* were collected together from a narrow igarapé or stream 10-30 cm deep where sampled; water clear, slowly flowing, pH 5, surface temperature 29°C on day following collection (Figure 6). Gobies were taken from soft thick debris covering stream bottom a little below a stagnant flooded area with dying trees, water lilies, and water hyacinth; a larger but more slender eleotrid, tentatively *Microphilypnus ternetzi*, was collected at the same time and place. Other fish species taken at the sampling site were the cichlids *Apistogramma agassizii, A. pertensis, Taeniacara candidi, Mesonauta festivus, Mesonauta* sp., *Laetacara curviceps,* and *Crenicara maculatum*; the gymnotoid *Eigenmannia virescens;* the characoids *Charax condei, Copella* sp., *Hemigrammus laevis, H. schmardae,*

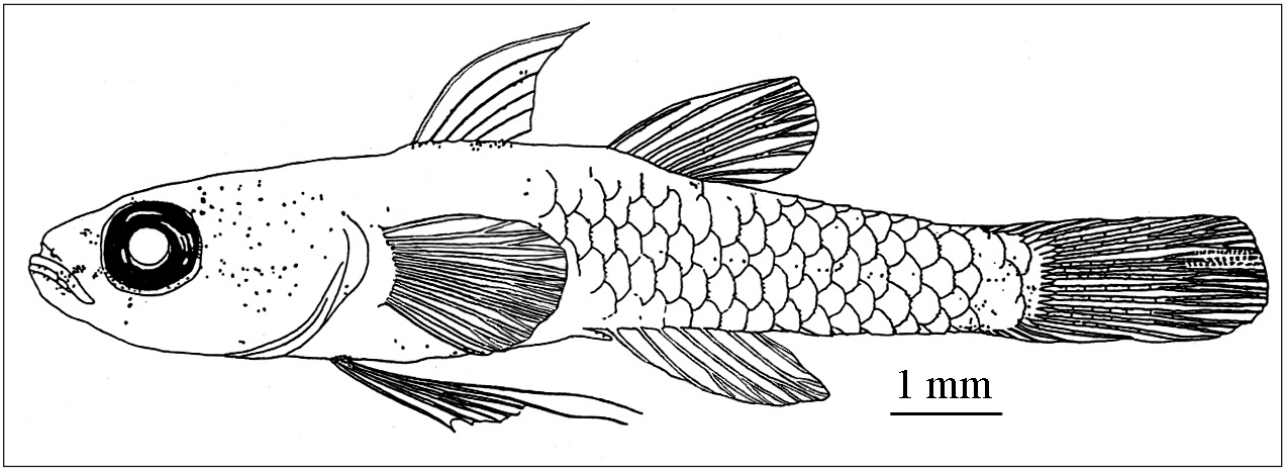

Fig. 2. *Leptophilypnion pusillus*, holotype, 9.1 mm female. Dots on head, body, and fins represent melanophores. Branching of dorsal, anal, and particularly pectoral fin rays may not be reliable. Drawing by T. R. Roberts.

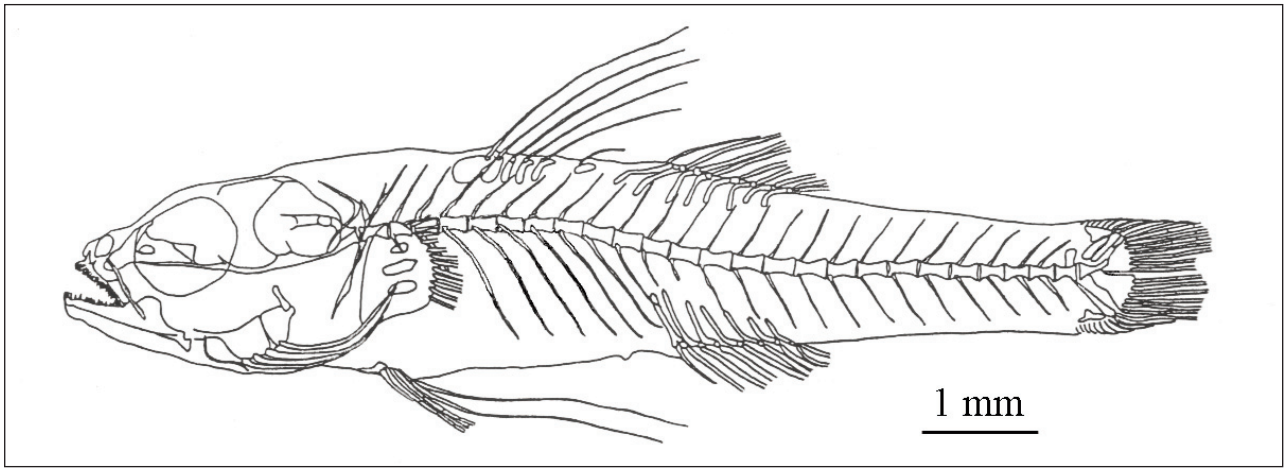

Fig. 3. *Leptophilypnion pusillus*, paratype, 9.0 mm female, skeleton (opercular bones sloughed off during clearing and staining process). Drawing by T. R. Roberts.

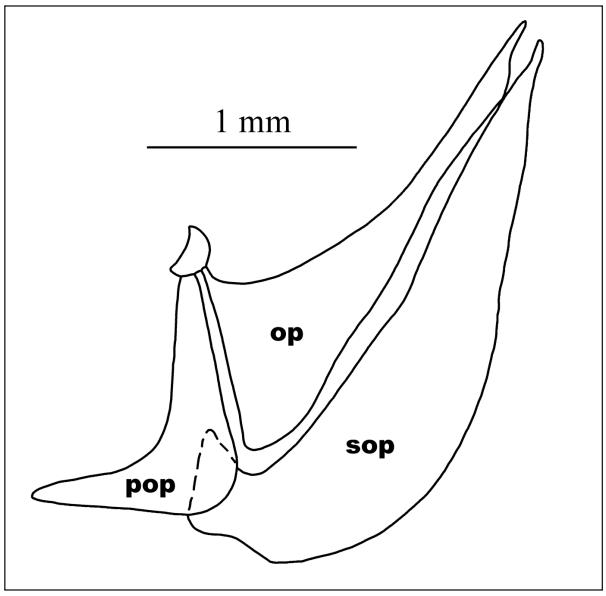

Fig. 4. *Leptophilypnion pusillus*, paratype, 9.0 mm female, opercular bones (interopercle missing). **pop** = preopercle; **op** = opercle; **sop** = subopercle.

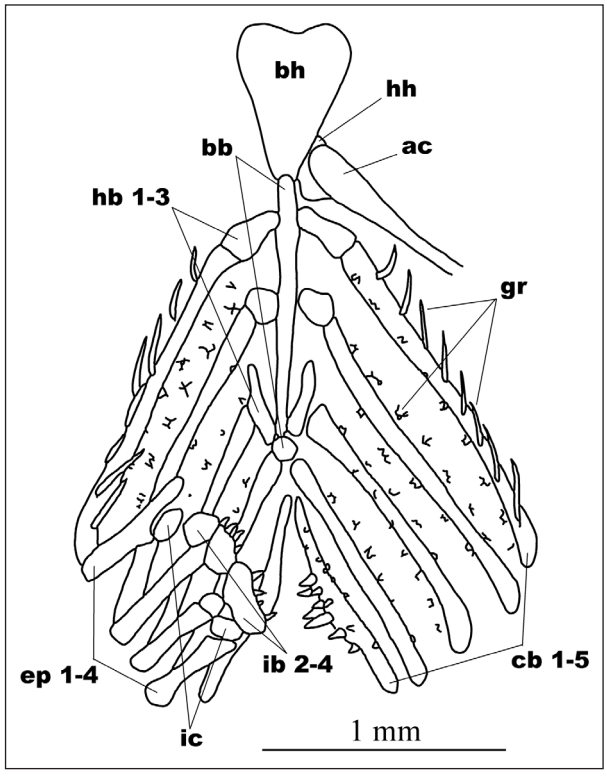

Fig. 6. *Leptophilypnion pusillus,* paratype, 9.0 mm female, branchial arches. **ac** = anterior ceratohyal; **bb** = basibranchials 1-3 (fused), basibranchial 4; **bh** = basihyal; **cb** = ceratobranchials; **eb** = epipharyngobranchials; **gr** = gill rakers; **hh** = hypohyal; **hp** = hypobranchials; **ib** = infraphyngobranchials; **ic** = interarcual cartilages.

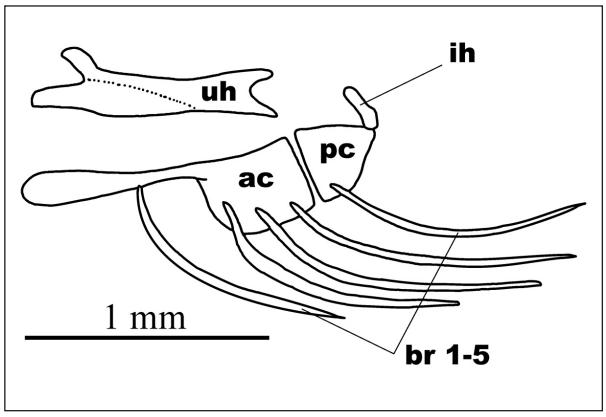

Fig. 5. *Leptophilypnion pusillus*, paratype, 9.0 mm female, urohyal and hyoid bar with five branchiostegal rays. **ac** = anterior ceratohyal; **br** = branchiostegal rays; **ih** = interhyal; **pc** = posterior ceratohyal; **ur** = urohyal.

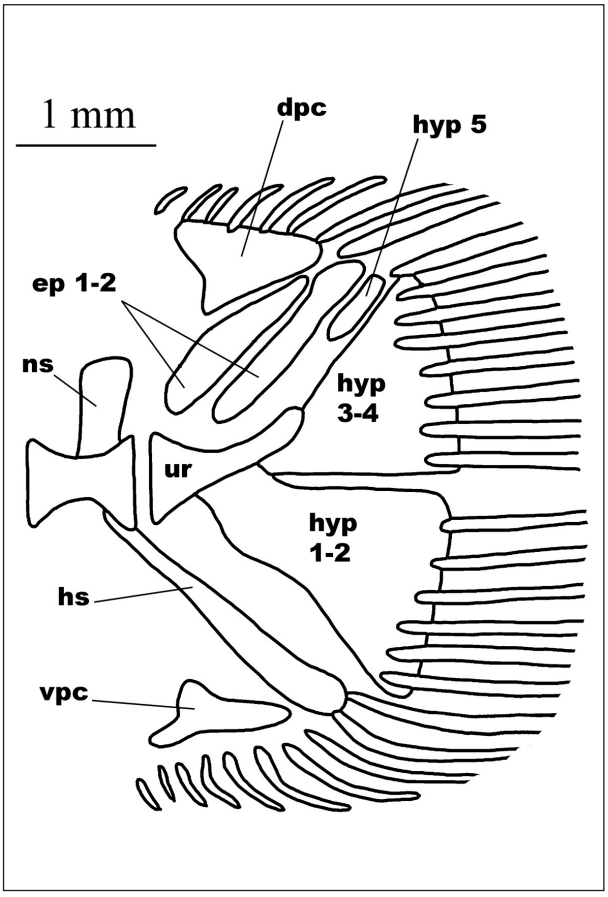

Fig. 7. *Leptophilypnion pusillus*, paratype, 9.0 mm female, caudal fin ekeleton, 9.0 mm. **dpc** = dorsal procurrent cartilage; **ep** = epurals; **hp** = hypurals; **hs** = hemal spine; ns = neural spine; **ur** = urostyle; **vpc** = ventral procurrent cartilage. Caudal fin ray formula = 8, 8/7, 10 (eight upper procurrent rays, eight upper principal rays; seven lower principal rays, ten lower procurrent rays).

Hoplias malabaricus, Moenkhaiusia collettii, Moenkhausia sp., *Nannostomus diagrammus, N. eques, N. marilynae,* and *Pyrrhulina* sp; the cyprinodonts *Poecilia minor* and *Fluviphylax pygmaeus*; and the nandid leaf-fish *Monocirrhus polyacanthus*. All except *Hoplias malabaricus*, represented by a single small juvenile, are relatively small species. By 1989, a shack suburb had grown up around the stream and it had become a polluted swamp but it is likely that *L. pusillus* still occurred in similar sites nearby. At the time collecting at the mouth of the stream yielded some larger specimens of *Microphilypnus* sp and a few other fish species (information in this paragraph provided by S. O. Kullander).

Santarém is a major center of the most active development or destruction of the Amazonian rain forest. The US-based Cargill Corporation completed a port facility for processing soybean there in 2003. This has had widespread negative environmental impacts. When the author flew over the area in January 2008 it was largely deforested for miles in all directions. Soya plantations require large quantities of phosphorus-based fertilizers, and these leach into rivers, cause eutrophication, and kill off most air-breathing fishes. It is doubtful that *Microphilypnus* or *Leptophilypnion* occur now anywhere within many miles of Santarém. While the species of *Microphilypnus* are widely distributed in Amazonas, those of *Leptophilypnion* might be relatively localized.

Fig. 8a-b. Type locality of *Leptophilypnion pusillus:* igarapé tributary to Rio Tapajós near Santarém, 27 Sep 1980 (day of collection of type specimens). **a**. general habitat, swampy and shallow. **b**. close-up of muddy bottom. Photos by S. O. Kullander.

Leptophilpnion fittkaui n. sp.

Figs 9-12

Holotype: ZSM 42249, 9.2 mm, Brazil, Amazonas, Rio Negro watershed, Rio Cuieiras, Igarapé Cachoeira at Cachoeira Pedras dos Indios, field station A-294, [approximately 60 km NW of Manaus], E. J. Fittkau, 18-19 Dec 1961.

Paratypes: ZSM 28290, 9.5 mm, same locality, and collector, dates of collection, and field station number as holotype; ZSM 28291, 6: 8.9-9.7 mm, same locality, collector, field station, and date of collection as ZSM 28290; ZSM 28293, 9.5 mm, exact sampling location not recorded, field station 428-5, 2°41'15.00"S, 60°19'0.00"W (same tentative coordinates as for field station A-294, 27 Nov 1962 (cleared and stained with alizarin and alcian blue); ZSM 28288, 9.3 mm, Brazil, Amazonas, Rio Negro watershed, confluence of Rio Cueiras with Igarapé Cachoeira [approx. 60 km linear distance NW of Manaus], 2°41'15.00"S, 60°19'0.00"W (same tentative coordinates as for field station A-294, 17 Dec 1960.

Diagnosis: First dorsal fin pterygiophores insertion pattern 3-121110; vertebrae 11+15=26; dorsal and anal fin pterygiophores 7; and caudal fin with parhypural.

Description: First dorsal fin with 6 spines. Second dorsal fin with a spine and about 7 soft rays, difficult to count without tearing fin membranes. Pectoral fin rays about 13, several of them well branched. Pelvic fin with a spine and 5 rays (preceding counts of fin rays from holotype). Leading edge of lower limb of first gill arch with 9 elongate gill rakers on each side in single cleared and stained specimen. Upper limb of first gill arch without gill rakers.

The specimens designated here as type material of *L. fittkaui* were identified at first as *L. pusillus*. They agree with that species in the robustness of head and body, relatively blunt snout, strongly ctenoid scales restricted to the posterior two-thirds of body (and no cycloid scales on body or head)

much better than with any of the species of the terete, relatively sharp-snouted *Microphilypnus*. *Microphilypnus ternetizi* and *M. macrostoma* are both sharp-snouted; *M. acangaquara* is less sharp-snouted, but still more so than *Leptophilypnion*. Characteristic of *Leptophilypnion*, they also have the elongate two innermost rays of the pelvic fin. On the other hand, the specimens of *L. fittkaui* differ substantially from those of *L. pusillus* in having a shorter body. This is reflected in differences in several proportional measurements and perhaps also in differences in the vertebral counts and numerical relationship of first dorsal fin pterygiophores to neural spines (Table I).

The 9.5-mm paratype was cleared and stained in September 2012 but did not come out well. At the end of the preparation period of five days the jaws, most of the palatoquadratohyomandibular arch, the branchiostegal rays, all fin rays of the dorsal, anal, pelvic and caudal fins, and nearly all of those

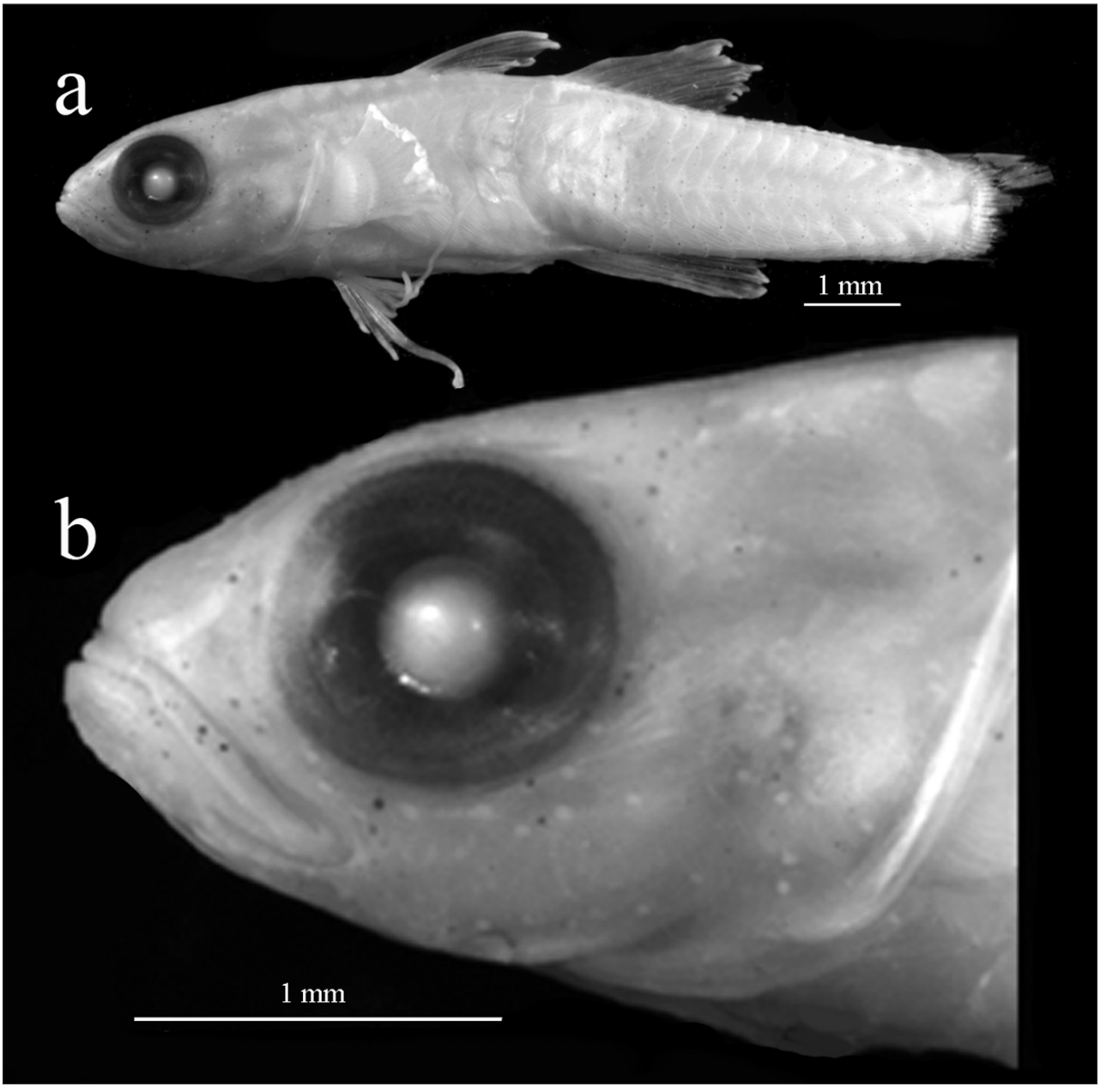

Fig. 9a-b. *Leptophilypnus fittkaui*, holotype, 9.2 mm, sex undetermined. **a.** whole specimen in lateral view; **b.** head in lateral view. Photos by S. O. Kullander.

of the pectoral fins also had been lost. Most of the remaining skeleton, including the branchial arches, vertebrae and neural spines, was well stained with methylene blue, but only lightly stained with alizarin. Observations on the relationship of the dorsal fin pterygiophores to the neural spines and of the gill rakers were made then. Two days later the faint rosy stain of the alizarin had disappeared entirely and the blue stain of the methylene blue was almost gone.

A single specimen (not a type, but also collected from Igarapé Cachoeira at Cachoeira Pedra dos Indios) was cleared and stained by P. J. Miller, who made the drawings of its palatoquadratohyomandibular arch and hyoid bar. He confirmed the presence of 5 branchiostegal rays. He also drew the caudal fin skeleton (without showing the principal and procurrent caudal fin rays) with a parhypural element present.

The external morphology and the evidence that have been gathered from the two cleared and stained specimens indicate fundamental similarities and also striking dissimilarities between the two species. The most striking differences are skele-

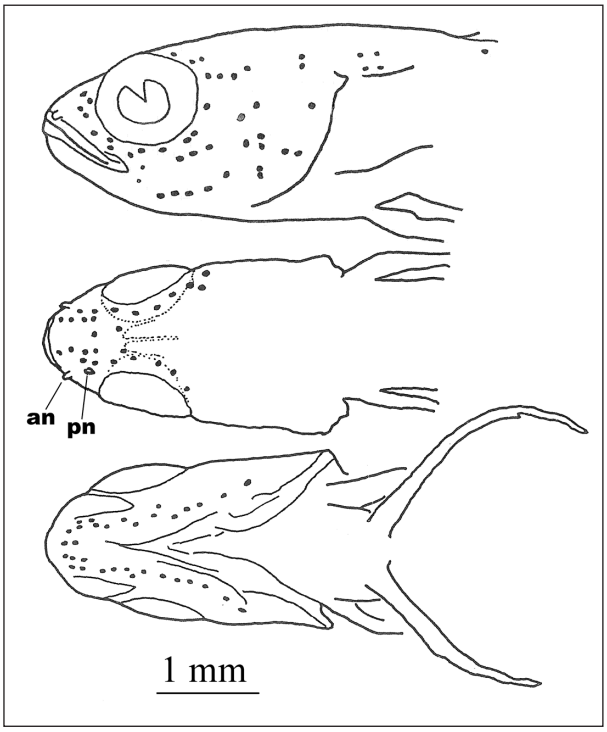

Fig. 10. *Leptophilypnion fittkaui*, distribution of neuromast organs on head. a, dorsal view; b, lateral view; c, ventral view. Composite diagrammatic drawing based on specimens from Igarapé de Cachoeira (by P. J. Miller). **an** = anterior nostril, **pn** = posterior nostril.

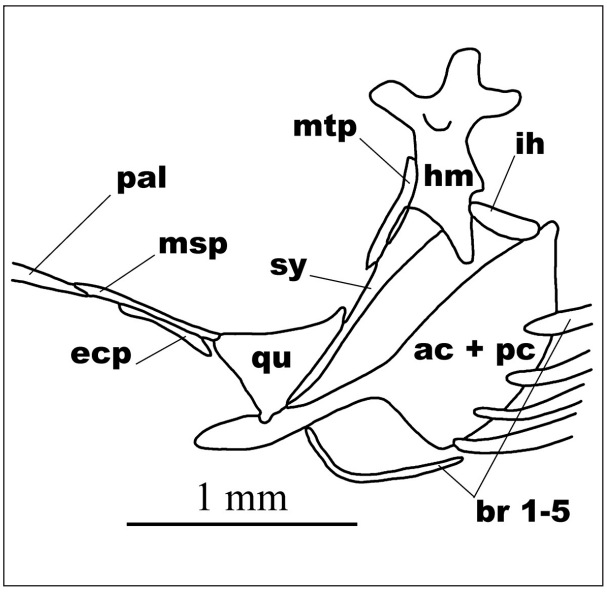

Fig. 11. *Leptophilypnion fittkaui*, non-type specimen from Igarapé de Cachoeira, partial drawing of palatoquadratohyomandibular arch and branchiostegal bar (by P. J. Miller). **ac+pc** = anterior and posterior ceratohyals; **br** = branchiostegal rays; **ecp** = ectopterygoid; **hm** = hyomandibular; **ih** = interhyal; msp = mesopterygoid; **mtp** = metapteygoid; **pal** = palatine; **q** = quadrate; **sy** = symplectic.

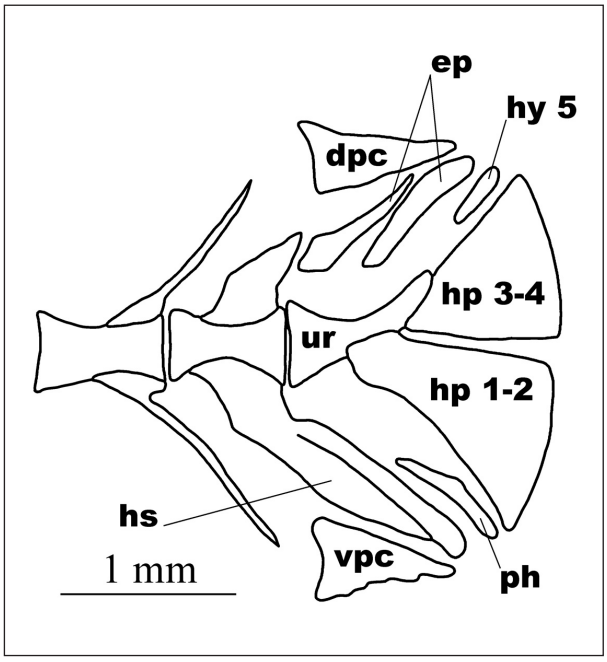

Fig. 12. *Leptophilypnion fittkaui*, non-type specimen from Igarapé de Cachoeira, caudal fin skeleton (drawn by P. J. Miller). **dpc**= dorsal procurrent cartilage; **ep** = epurals; **hp** = hypurals; **hs** = hemal spine; **ph** = parhypural; **ur** = urostyle; **vpc** = ventral procurrent cartilage.

tal: a 3-12201 first dorsal fin pterygiophore insertion pattern, 10+16=26 vertebrae, 7 dorsal and 8 anal pterygiophores, and a caudal fin without the parhypural in *L. pusillus versus* a 3-121110 pattern, 11+15=26 vertebrae, 7 dorsal and anal pterygiophores and a caudal fin with the parhypural in *L. fittkaui*.

Sensory papillae are readily visible in several of the type specimens of *L. fittkaui* including the holotype (Fig. 9b). They were not observed in the three type specimens of *L. pusillus*. The author is inclined to think that this represents a biologically and taxonomically important difference, but it might be due to a difference in the preservation history of the specimens from Santarém.

Etymology: *Leptophilypnion fittkaui* is named in honour of Ernst Josef Fittkau (1927- 2012), advocate of Amerindian rights, conservationist, entomologist (Diptera, especially Chironomidae), and collector of the type specimens of this species.

Notes on type locality: The holotype and one paratype from field station A-294 flushed out of aquatic plant *Tonina fluviatilis* (Eriocaulaceae) together with a tadpole, *Hydracarina* and algae. Paratypes from field station 428-5 from leaf litter together with Dytiscidae, small Corixidae, Copepoda, Tubificidae, numerous Ceratopogonidae, Mysidacea and *Macrobrachium* (from field notes of E. J. Fittkau).

DISCUSSION

Leptophilypnion seemingly is most closely related, not to the Amazonian species of *Microphilypnus*, but to the Central American estuarine and freshwater eleotrids of the genus *Leptophilypnus* Meek and Hildebrand 1916 (type species *Leptophilpynus fluviatilis* Meek and Hildebrand 1916). The closest similarity appears to be with *L. guatemalensis*, a freshwater species recently described from the Usumacinta River system of the Atlantic coast of Mexico and Guatemala. Characters shared by *Leptophilypnion* and *L. guatemalensis* but not *Microphilypnus* include a far more robust head and body, details of the insertion of first dorsal fin pterygiophores with the neural spines, number of vertebrae, and number of principal caudal fin rays.

The single cleared and stained specimen of *L. pusillus* has a neural spine to dorsal fin pterygiophore configuration of 3-12201. Among eleotrids this is otherwise known only in *L. guatemalensis*, which may also have 3-12210, 3-22110, or 3-22101. The first two of these, 3-12210, 3-22110. are widespread among gobioids including *Microphilypnus*. The last, 3-22101, apparently is unique to *L. guatemalensis*. The configurations 3-12201 and 3-22101 both indicate that the last spine of the first dorsal fin is widely separated from the preceding five spines, a character shared by *L. pusillus* and at least some specimens of *L. guatemalensis*, but not with *Microphilypnus* or other species of *Leptophilypnus*. The configuration in the single cleared and stained specimen of *L. fittkaui*, 3-121110, has been reported for the gobiid genus *Evermannichthys* (Birdsong et al., 1988) and *Microphilypnus* (Caires & Figueiredo, 2011). It suggests the gap between the sixth or last dorsal fin spine and the preceding five dorsal fin spines is less marked in *L. fittkaui* than in *L. pusillus*.

Vertebral counts of 10+16=26 occur in *L. pusillus* and *L. guatemalensis* (which can also have 10+17=27). *Microphilypnus* have at least 11 abdominal vertebrae (precaudal vertebrae of some authors). A vertebral count of 11+15=26, found in the single cleared and stained *L. fittkaui*, has not been found in *Microphilypnus* or in *Leptophilypnus*.

The three known species of *Leptophilypnus* grow to a maximum of less than 55 mm standard length, small but still relatively large compared to *Microphilypnus* and *Leptophilypnion*. *Leptophilypnus fluviatilis* attains nearly 55 mm, *L. guatemalensis* at least 38 mm.

As pointed out by reviewers, a proper reappraisal of eleotrid phylogenetic relationships might not substantiate the hypothesis that *Leptophilynion* is most closely related to *Leptophilypnus*. Although reported pterygiophore counts are similar between these taxa, examination of more cleared and stained specimens is likely to reveal considerably more intraspecific variation in this character.

The same anonymous reviewer also feels that the present discussion of relationship "ignores the presumably great skeletal differences between the new genus and other neotropical eleotrids. For example, Fig. 11 of *L. fittkaui* depicts a large endopterygoid, which reaches the posterior border of the dermopalatine, and a small ectopterygoid that does not reach this point. These features are very unusual not only in eleotrids, but in other gobioids as well. Gobies generally have a large ectopterygoid connecting with palatine by a shaft, and a small endopterygoid. *Leptophilypnion* appears to be a remarkably miniaturized taxon, so it will be difficult to ascertain its phylogenetic place inside Eleotridae. Therefore, it is premature to suppose

close relationships among this taxa and other eleotrids based on the similarities indicated". The unusual condition of the ectopterygoid and endopterygoid of *Leptophilypnion* may well be uniquely derived or apomorphic characters, and thus unhelpful in determining its phyletic relationships.

It is incumbent upon those describing a distinctive new taxon to try to identify its phylogenetic relationships. Even the most advanced phylogenetic analysis, by whatever means, and based upon unlimited material, may result in a faulty phylogenetic hypothesis. The present hypothesis of relationship of *Leptophilypnion* to *Leptophilypnus* is supported by the (admittedly limited) available evidence from a very small number of specimens, not in the best of condition. Anyone can replace it with an alternative hypothesis supported by better evidence at any time.

ACKNOWLEDGEMENTS

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