



Three new endemic cryptic species revealed by DNA barcoding of the gobies of the Cayman Islands (Teleostei: Gobiidae)

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Abstract

A survey of the mitochondrial DNA marker COI for the gobies of the Cayman Islands reveals two categories of gobies: a set of genera that are no different genetically from other Caribbean populations, including *Coryphopterus*, *Ctenogobius*, *Gnatholepis*, and *Priolepis*, and a set of species that have local mtDNA lineages quite different from populations elsewhere in the region, including *Elacatinus*, *Tigrigobius*, and *Risor*. The cryptic divergent lineages include some that show distinct phenotypic differences and are described here as three new endemic species, as well as others that have divergent local DNA lineages but no apparent phenotypic differences and are treated as genovariants, i.e. populations of the same species with genetic differences. The Cayman Cleaner Goby, *Elacatinus cayman* n. sp., is found to be a close relative of *E. evelynae* (1.5% divergent; minimum interspecific distance, K2P), despite the fact that specimens were originally paratypes of *E. genie* (which are 7.5% divergent): their shared feature of a complete groove above the upper lip is not apparently a synapomorphy with *E. genie* (i.e. not a shared derived character). A neighbor-joining tree of COI mtDNA sequences for all ten cleaner goby species of the tropical W. Atlantic is presented. The Cayman Sponge Goby, *Elacatinus centralis* n. sp., differs from *E. horsti* of the southern Caribbean by the color of the stripe and snout, and is further genetically than is the differently marked Shortstripe Sponge Goby *E. chancei* (the geographically intervening relative from the Lesser Antilles). A neighbor-joining tree of COI mtDNA sequences for all but two of the striped sponge goby species of the tropical W. Atlantic is presented. The Cayman Greenbanded Goby, *Tigrigobius harveyi* n. sp., has many more green bands than the Greenbanded Goby of the Antilles, *T. multifasciatus*, and differs by more than 10% in mtDNA sequence; however, it is only 3% different from the Redcheek Goby of Honduras and the Panamanian Greenbanded Goby. Other species of *Tigrigobius*, as well as *Risor ruber*, have divergent local DNA lineages, but no apparent phenotypic differences from other Caribbean populations. No consistent biogeographical relationships are apparent— the location of the nearest-neighbor lineages within the region differs from species to species. A revised species list for the gobies of the Cayman Islands is presented and the Kuna Goby, *Coryphopterus kuna*, is recorded there for the first time.

Key words: coral reef fishes, *Elacatinus*, *Tigrigobius*, cryptic, cleaner, sponge, taxonomy, systematics, phylogeography, biogeography, endemism, Caribbean.

Introduction

The Cayman Islands consist of three small islands relatively isolated in the central western Caribbean Sea and comprising the tiny peaks of a massive underwater mountain range, known as the Cayman Ridge (Fig. 1) (Jones 1994). The Ridge flanks the great Cayman Trough, an exceptionally deep trench separating the North American Plate from the Caribbean Plate and slicing across the northwestern quadrant of the Caribbean (Cayman Trench in Fig. 1). Oceanographic currents around the Cayman Islands are summarized by Turner *et al.* (2013). The islands sit on the northern edge of the broad westward-flowing Caribbean current that crosses the region and the predominant current at Grand Cayman is a fast flow to the northwest. Little Cayman has a strong SE current looping back to the islands and then toward Jamaica. Nevertheless, drifters off Grand Cayman often follow a southeast trajectory and Turner *et al.* (2013) concluded that the islands are in the focus of “confused crossroads of the mainstream Caribbean current”. The calcareous geology and the adjacent profound dropoff results in exceptional water clarity and a limited reef shelf that characterizes the reef landscapes around the Cayman Islands and reduces the variety of habitat types and faunal diversity compared to other more continental and less isolated locations in the Caribbean (Roberts 1994). The relative isolation of the islands provides an interesting opportunity for assessing potential undetected cryptic diversity and endemism.

The fish fauna of the islands has been relatively well-surveyed and a total of 423 reef fish species have been collected or sighted to date (Burgess *et al.* 1994; Pattengill-Semmens & Semmens 2003). Burgess *et al.* (1994) reviewed the fishes of the Cayman Islands based on collections and museum records, and Pattengill-Semmens



Figure 1. Map of the Caribbean Sea, Cayman Islands in NW quadrant (CIA, US Government, Library of Congress).

& Semmens (2003) reported the underwater observations of fishes based on surveys by the REEF organization (www.reef.org). Burgess *et al.* (1994) list 35 named goby species for the Cayman Islands, while REEF recorded 23 species of gobies. The sets of species are only partially overlapping, since the two studies used very different methods. A review of the combined listing is presented below, documenting 38 species of gobies for the Cayman Islands, three of which are described as new species. The cleaner and sponge gobies of the Greater Caribbean were recently reviewed by Colin (2010), who suggested that both the cleaner goby and the striped sponge goby of the Cayman Islands were likely undescribed species, a prediction that has been borne out.

The recent advent of large-scale DNA sequencing and the assembling of a massive database of sequences of the mitochondrial cytochrome-oxidase I “barcode” gene by the Barcode of Life project has facilitated the application of DNA taxonomy to reef fishes (Ward *et al.* 2009). The initiative for fishes (FISH-BOL) has been particularly successful for the tropical Western Atlantic region, with up to 90% of the known reef fish species sequenced to date (Victor, in press). One of the more intriguing findings from these new data is the marked degree of cryptic diversity (i.e. hidden or undetected) among certain reef fish families, especially the gobies and blennioids (e.g. Tornabene *et al.* 2010; Baldwin *et al.* 2011). These taxa account for a large fraction of the total reef-fish species count, even before the consideration of potential cryptic diversity. As part of a survey of the cryptofauna of unsampled Caribbean locations, the COI mtDNA marker was sequenced for half of the resident species of gobies in the Cayman Islands. Comparisons with sequences from elsewhere in the Caribbean were feasible for most species and the results reveal an interesting pattern of regional endemism that casts new light on the complex biogeography of Caribbean reef fishes.

Materials and Methods

Live specimens were collected by hand on the reefs of SW Grand Cayman and at Bloody Bay on Little Cayman in April and May 2014 and placed directly into 95% ethanol. Type specimens were deposited in the UF collection and the SIO collection. Comparison specimens and sequences for other species were collected from numerous other locations and are listed in Appendices 1–3.

DNA extractions were performed with the NucleoSpin96 (Machery-Nagel) kit according to manufacturer specifications under automation with a Biomek NX liquid-handling station (Beckman-Coulter) equipped with a filtration manifold. A 652-bp segment was amplified from the 5' region of the mitochondrial COI gene using a variety of primers (Ivanova *et al.* 2007). PCR amplifications were performed in 12.5 μ l volume including 6.25 μ l of 10% trehalose, 2 μ l of ultra pure water, 1.25 μ l of 10 \times PCR buffer (10mM KCl, 10mM (NH₄)₂SO₄, 20mM Tris-HCl (pH8.8), 2mM MgSO₄, 0.1% Triton X-100), 0.625 μ l of MgCl₂ (50mM), 0.125 μ l of each primer (0.01mM), 0.0625 μ l of each dNTP (10mM), 0.0625 μ l of Taq DNA polymerase (New England Biolabs), and 2 μ l of template DNA. The PCR conditions consisted of 94°C for 2 min., 35 cycles of 94°C for 30 sec., 52°C for 40 sec., and 72°C for 1 min., with a final extension at 72°C for 10 min. Specimen information and barcode sequence data from this study were compiled using BOLD, i.e. the Barcode of Life Data Systems (Ratnasingham & Hebert 2007). The sequence data is publicly accessible on BOLD and GenBank. Sequence divergence was calculated using BOLD with the Kimura 2-parameter (K2P) model generating a mid-point rooted neighbor-joining (NJ) phenogram to provide a graphic representation of the species divergence (pairwise distances are also calculated for comparisons).

Measurements were made by ocular micrometer or digital calipers and are presented as the range for the paratypes followed by the value for the holotype in parentheses. Only adult specimens over 20 mm SL for *Elacatinus cayman* and *E. centralis* or over 16 mm SL for *Tigrigobius harveyi* were included in morphometrics, while all types were used for counts. The last ray of the dorsal and anal fins are split to the base and counted as one. The first element of the second dorsal fin and anal fin are spines, but included in the count of total elements, following Böhlke & Robins (1968).

Lengths of specimens are mm standard length (mm SL), measured from the front tip of the snout to the base of the caudal fin (posterior end of the hypural plate); body depth is the vertical distance at the base of the first dorsal spine; body width is the maximum width side-to-side just posterior to the gill opening (un squeezed); head length (HL) is the horizontal distance from the tip of the snout to the most posterior end of the

opercular flap; snout length is the horizontal span (not angular distance) from the snout tip to the anterior edge of the bony orbit; orbit diameter is the horizontal distance from edge to edge of the bony orbit; upper-jaw length is the full oblique length; predorsal, prepelvic, and preanal lengths are measured along the oblique line; caudal-peduncle depth is the least depth and caudal-peduncle length is the horizontal distance from the base of the last anal-fin ray to the caudal-fin base; lengths of fin spines and rays are measured to their junction with the body; caudal-fin length is the horizontal distance from the base of the fin to a vertical at the tip of the longest ray; pectoral-fin length is the length of the longest ray; pelvic-fin length is measured from the junction with the body to the tip of the longest soft ray.



Figure 2. *Elacatinus cayman*, Grand Cayman (Everett Turner).

***Elacatinus cayman*, n. sp.**

Cayman Cleaner Goby

Figures 2–7.

Gobiosoma (Elacatinus) genie [in part] Böhlke & Robins 1968: 109.

Gobiosoma genie (non Böhlke & Robins) Colin 1975: 58.

Gobiosoma (Elacatinus) evelynae (non Böhlke & Robins) Burgess *et al.* 1994: 220.

Gobiosoma (Elacatinus) genie (non Böhlke & Robins) Burgess *et al.* 1994: 220.

Gobiosoma evelynae (non Böhlke & Robins) Pattengill-Semmens & Semmens 2003: Appendix A.

Gobiosoma genie (non Böhlke & Robins) Pattengill-Semmens & Semmens 2003: Appendix A.

Elacatinus evelynae (W) (non Böhlke & Robins) Taylor & Hellberg 2003: 108.

Elacatinus evelynae (W) (non Böhlke & Robins) Taylor & Hellberg 2006: 697.

Elacatinus sp. 1 Colin 2010: 7.

Holotype. UF 237057, 23 mm SL, male, SW Grand Cayman Island, 19.286° N, -81.392° W, April 27, 2014.

Paratypes. UF 237058, (9) 12.4–23.3 mm SL, same data as holotype; SIO 14-110, (4) 15.9–22 mm SL, same data as holotype.

Diagnosis. Dorsal rays VII+12; anal rays 11; pectoral rays 16–17; pelvic rays I,5 and fused into a cup; no scales; mouth inferior; complete upper lip groove (no frenum); upper head and body black, pale middorsal streak behind head, lower half of head and ventral aspect of body white; wide iridescent blue lateral band divides black



Figure 3. *Elacatinus cayman*, note pale middorsal streak behind head, Little Cayman (Frank Krasovec).

upper body, band contains a full-length midline yellow stripe which extends forward onto upper orbit and meets on snout in a V or U; notably, yellow stripe is of uniform width from eye to tail— that color pattern is distinctive, shared only with rare individuals of *E. evelynae* YB (YB denotes the yellow-blue striped morph). The absence of the upper-lip frenum distinguishes *E. cayman* from *E. evelynae*, with few exceptions. *E. genie* have a wider pale lateral band that is often bright white and the indistinct yellow stripe widens prominently behind the eye and there is no pale middorsal streak behind the head.

Description. Dorsal rays VII+12 (rare 11); anal rays 11 (rare 10); pectoral rays 16–17 (rare 15, mode 17); most frequent combination is D-VII+12 A-11 Pect-17/17; pelvic rays I,5 and fused into a cup; 11 precaudal and 17 caudal vertebrae (from the revision of the genus *Elacatinus* Jordan 1904 by Böhlke & Robins [1968]). No scales, body covered in a transparent mucus layer visible as a double line on profile in photographs and solidifying into a gray translucent skin on alcohol-preserved specimens.

Body elongate, body depth 14–19 (19)% SL, compressed, body width 12–13 (12)% SL; predorsal distance 33–36 (35)% SL; prepelvic distance short, 28–30 (28)% SL; preanal distance 55–61 (57)% SL; caudal peduncle long and relatively deep, length 21–24 (23)% SL, depth 11–12 (11)% SL. Head short, head length 24–27 (25)% SL; eye large, orbit diameter 24–28 (24)% HL; snout short and blunt, snout span 14–17 (16)% HL; mouth inferior, with a clear protuberance of snout overhanging anteriormost point; mouth relatively large, corner of gape with mouth closed extending beyond vertical through rear of pupil, frequently just past vertical through rear of orbit, upper jaw oblique length 32–40 (34)% HL; lips thick, divided from snout by a complete groove, no frenum eliminating groove at midline. Front teeth upper and lower in a close palisade of tiny peglike teeth with triangular points, up to 25 or more in each quadrant; lower jaw has an additional second and third irregular inner row of



Figure 4. *Elacatinus cayman*, note yellow stripe complete and uniform width, Grand Cayman (Everett Turner).



Figure 5. *Elacatinus cayman*, gravid female with visible eggs, Grand Cayman (Everett Turner).

caniniform teeth, slightly larger and more spaced out, with one or two distinctly larger canines in males. Anterior nostril a short membranous tube overhanging upper lip, posterior nostril well in front of orbit and just anterior to nasal pore (anterior supraorbital of Böhlke & Robins [1968]). Pores on head comprise Birdsong's B' C D E F H' M' N'.

These measurements are mostly concordant with those of Böhlke & Robins (1968), however two are distinctly different: they measure the snout length as almost the same as the eye diameter and the upper jaw length as more than 45% HL. I cannot reconcile the difference other than note that they specifically single out those two measurements as made using their ocular micrometer instead of the calipers they used for all other measurements (Böhlke & Robins [1968: 49]), suggesting it may represent a calibration error.

Two dorsal fins; first spinous and short with 7 slender flexible spines, no extended spines in either sex, membrane of last dorsal spine connecting to body in front of insertion of second dorsal fin, third dorsal spine barely longest 12–15 (14)% SL; second dorsal fin medium length, base 22–25 (24)% SL, spine slender and flexible, shorter than rays 8–9 (10)% SL, first soft ray unbranched, third soft ray usually longest 12–15 (13)% SL, last split to base; anal fin shorter than dorsal, base 15–19 (20)% SL, spine slender and flexible, shorter than rays 6 (7)% SL, first soft ray unbranched, fourth soft ray usually longest 12–13 (14)% SL, last split to base; pectoral fin short, upper and lower two rays unbranched, about 9th ray longest, 20–22 (23)% SL; pelvic fins fused into a cup, all rays deeply branched, short, 18–20 (20)% SL, reaches two-thirds of way to anus; caudal fin truncate, short, length 19–20 (18)% SL, 17 (rare 16) segmented caudal-fin rays, 2–3 uppermost and 2–3 lowermost unbranched, 6–8 upper and 7–9 lower procurrent rays (usually one more lower). No scales. Genital papilla in males a long tapering cone, up to pupil diameter; females with a shorter wider cone with fimbriated rim.

Color in life. Upper head and body black, with black area behind the eye divided into two black bands by a wide central band of iridescent blue containing a full-length midline yellow stripe which extends forward onto upper orbit and then meets on snout in a V; notably, yellow stripe is of uniform width from eye to tail. Upper black band thickest on head, surrounding a pale middorsal streak behind head, narrowing along upper body and trailing into a thin line on caudal fin. Lower black band mostly uniform width until end of caudal fin where it narrows. Lower half of head and ventral aspect of body white. All fins other than caudal fin are transparent.

Color in alcohol. All color is lost and reverts to pale, leaving the black on white pattern intact. The colored stripe can be seen a reflective line containing fine melanophores after preservation.

Etymology. The specific epithet is a noun in apposition.

Barcode DNA sequence. The 652-nucleotide sequence of the segment of the mitochondrial COI gene used for barcoding by the Barcode of Life informatics database (BOLD)(Ratnasingham & Hebert 2007) was obtained for the holotype and paratypes (Appendix 1). Following the database management recommendation of the BOLD, the sequence of the holotype (GenBank accession number KM987241) is presented here as well:

CCTTTACTTGATTTTTGGTGCATGGGCCGGAATAGTGGGGACTGCTCTTAGCCTCCTTATCCGGGCTG
AGCTAAGCCAACCCGGAGCTCTCTTAGGCGATGACCAAATTTATAATGTAATCGTCACCGCACACGC

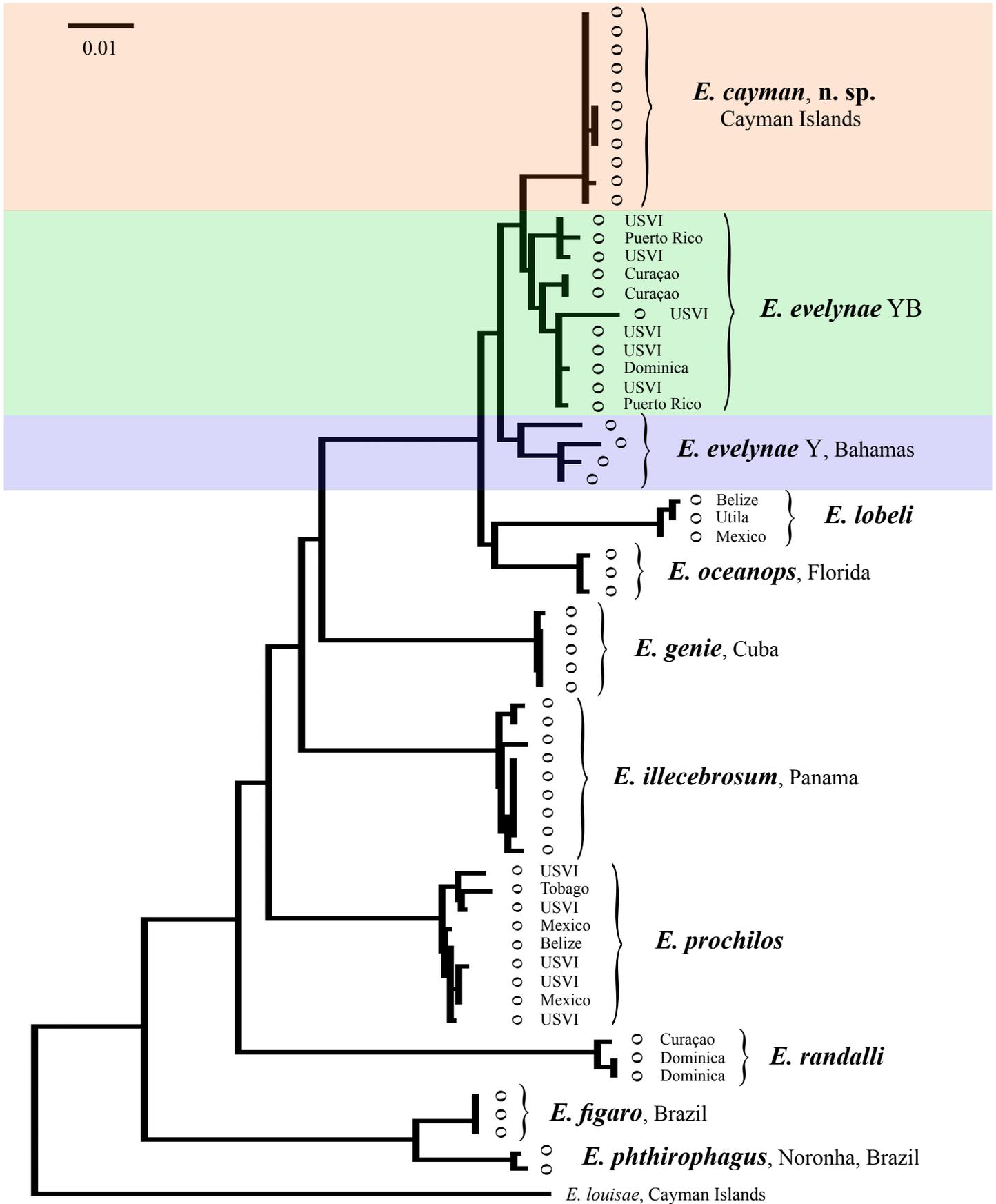


Figure 6. The neighbor-joining phenetic tree of all the tropical W. Atlantic *Elacatinus* cleaner gobies following the Kimura two-parameter model (K2P) generated by BOLD (Barcode of Life Database). The scale bar represents a 1% sequence difference. Collection locations for specimens are indicated, and the sponge goby *Elacatinus louisae* is used as an outgroup. GenBank accession numbers and collection data for the sequences in the tree are listed in Appendix 1 in the same order as the sequences in the tree.

TTTTGTAATAATCTTTTTTCATAGTGATGCCAATTATAATTGGAGGCTTCGGAAACTGATTAATTCCACT
TATGATTGGTGCCCCCGATATAGCATTCCCTCGCATAAATAACATAAGCTTCTGGTTACTCCCTCCCTC
CTTTCTCCTACTTCTTGCCCTCTCCGGGGTTGAGGCCGGTGCCGGTACGGGTTGAACTGTCTACCCC
CCCTTAGCCGGCAACTTAGCCCATGCCGGGGCATCCGTTGACCTAACTATTTTTTCCCTCCATTTAGC
AGGGATTTCTTCAATCTTAGGTGCAATCAACTTTATCACCACCATCATAAACATGAAGCCTCCCGCCG
TATCCCAATATCAAACCCCCCTGTTTGTCTGAGCTGTTTTAATTACAGCAGTCCTCCTCCTTCTTTTAC
TTCCTGTCTTGCAGCTGGTATTACCATACTCCTAACAGACCGAAACCTGAATACCACCTTCTTTGAC
CCTGCAGGTGGAGGAGACCCCATCCTTTACCAACACCTG

Comparisons. Despite being described originally as paratypes of *E. genie* by Böhlke & Robins (1968), and subsequently assigned to *E. genie* (Böhlke & Chaplin 1968; Colin 1975; Burgess *et al.* 1994; and many guidebooks), *E. cayman* is clearly a close relative of *E. evelynae* by mtDNA sequence. A neighbor-joining phenetic tree based on the COI mtDNA sequences (following the Kimura two-parameter model (K2P) generated by BOLD) for the cleaner gobies of *Elacatinus* (Fig. 6) includes all eight species known from the Greater Caribbean, plus two from Brazil, but not the two planktivorous congeners from Bahamas and Veracruz in the Gulf of Mexico. It shows relatively shallow divergences between the *E. cayman* clade and nearby *E. evelynae* color morphs (Fig. 6). The minimum interspecific distance between *E. cayman* and *E. evelynae* YB from the Lesser Antilles is 1.5% (by K2P, 1.5% pairwise) and 2.4% (2.3% pairwise) from Bahamas *E. evelynae* Y specimens. In contrast, the Neon Goby from Florida, *E. oceanops*, is 3% divergent (2.9% pairwise) and true *E. genie* are 7.5% divergent (7% pairwise).

A multigene phylogeny by Rüber *et al.* (2003) also found *E. cayman* (listed as “*E. genie*”) to be a close relative of *E. evelynae*, genetically closer than *E. oceanops*: however, they did not sample true *E. genie*. Taylor & Hellberg (2003; 2006) explored the phylogeography of the cleaner gobies with a different mitochondrial marker and a nuclear marker and found a similar pattern, with the Cayman Cleaner Goby (considered by them as a “white” form of *E. evelynae*) closest to the Puerto Rican *E. evelynae* among regional populations, but also without sampling true *E. genie*.

Unlike most reef-fish species complexes, which have cleanly delineated sets of species with intraspecific variation much less than interspecific variation (often by an order of magnitude)(Victor, in press), the *E. evelynae* species complex is less well delineated and clearly reflects a complex history of isolation and recent limited gene flow (Taylor & Hellberg 2003; 2005; 2006). Indeed, the intraspecific variation, even within a color morph of *E. evelynae* (up to 1.9% maximum intraspecific difference in this study), can be greater than the minimum interspecific distance, i.e. no “barcode gap”. This likely reflects a degree of incomplete lineage sorting (insufficient time since speciation for accumulation of mutually exclusive mitochondrial lineages) or some hybridization between incipient species or subspecies muddying the waters. The cleaner gobies are certainly capable of successful hybridization in captivity (Colin 1975) and hybrids are sold in the aquarium trade. It should be noted, however, that the eleven sequenced type specimens of *E. cayman* sampled here do not exhibit the high genetic variation found in the Bahamas and Lesser Antilles samples of *E. evelynae* (0.31% maximum intraspecific difference for *E. cayman*).



Figure 7. *Elacatinus cayman*, absence of the upper lip frenum (complete groove), Grand Cayman (Everett Turner).



Figure 8. *Elacatinus evelynae* (W), white-striped morph, Dominican Republic (Diego Forero).

The shared feature that caused *E. cayman* to be considered a population of *E. genie* in the original species description is the absence of an upper-lip frenum (Böhlke & Robins 1968). However, the genetic results argue that the character is an independent convergent adaptation and not a synapomorphy shared with *E. genie*. Indeed, two other features were inconsistent with *E. genie* even at the time of the original description: *E. cayman* has a pale middorsal streak (overlooked in the 1968 description) and is much smaller than *E. genie* (maximum among the original paratypes was 24.9 mm SL and the largest of my series is 23.3 mm SL, vs. 36 mm SL for *E. genie*). Furthermore, one of the present paratypes of *E. cayman* has a slight frenum where the upper lip groove is barely discernable. On the other hand, the typical fin-ray counts of *E. cayman*, the combination of D-VII+12 A-11 Pect-17/17, is the same as the typical *E. genie* from the Bahamas: notably not one of the ten type specimens of *E. evelynae* from the Bahamas had that precise combination (Tables 9 & 10 of Böhlke & Robins [1968]).

Elacatinus cayman is genetically very close to various lineages of *E. evelynae* from the Lesser Antilles and resembles the yellow-blue version (YB) of *E. evelynae*; without the complete upper-lip groove they would be sometimes difficult to distinguish and, it could be argued, should represent a local subspecies. *Elacatinus evelynae* is found in the Bahamas and along the Antilles chain down to Venezuela and the Netherlands Antilles and comprises three mostly allopatric color morphs: yellow (Y) in Bahamas and Cuba, white (W) in the W. Caribbean offshore islands, Jamaica, and Hispaniola, and yellow-blue (YB) in the Bahamas and Lesser Antilles (Colin 1975; 2010; Taylor & Hellberg 2003; 2006). It is important to note that the ranges of *E. cayman* and *E. evelynae* (YB) do not overlap nor are they even adjacent—the nearest populations of *E. evelynae*, in Jamaica and Navassa Island, Hispaniola, and Swan Island, are white-striped (Colin 2010)(Fig. 8) and the Cuban population is yellow-striped (Fig. 9).

The color pattern of *E. cayman* does come very close to *E. evelynae* (YB), and rare individuals of *E. evelynae* (YB) have been photographed in the Lesser Antilles and Bahamas that share to some degree the uniform and full-



Figure 9. *Elacatinus evelynae* (Y), yellow-striped morph, no blue, Cuba (Wolfram Sander).

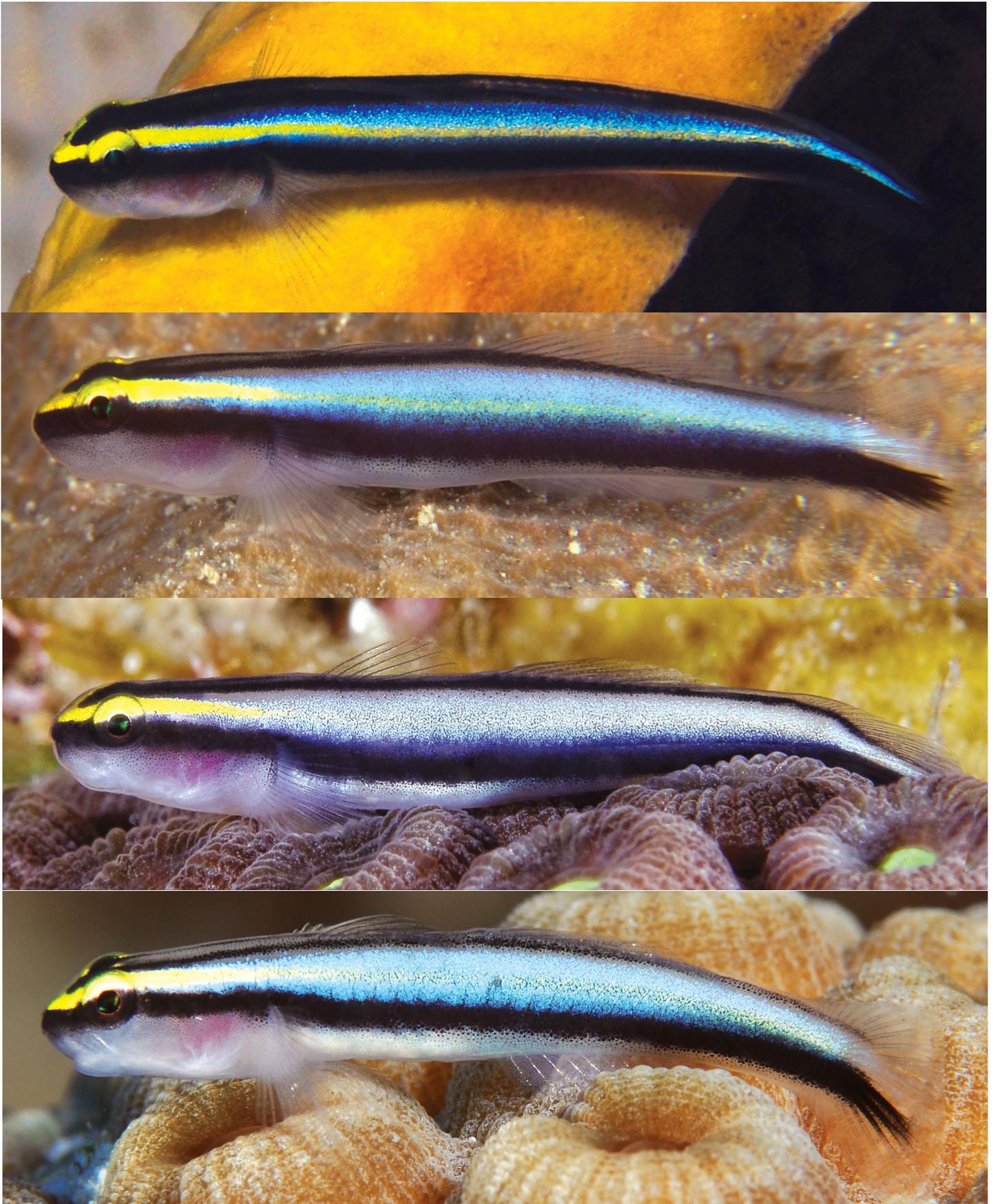


Figure 10. *Elacatinus evelynae* (YB) variations, **top:** yellow stripe complete, but narrowing abruptly behind eye, Nevis, Lesser Antilles (Mark Lessard); **upper middle:** yellow stripe thinning and fading, Nevis, Lesser Antilles (Mark Lessard); **lower middle:** yellow stripe disappearing, Nevis, Lesser Antilles (Jim Garin); **bottom:** yellow stripe disappearing, prominent pale middorsal streak, Turks and Caicos, Bahamas (Frank Krasovec).



Figure 11. *Elacatinus genie*, no pale middorsal streak behind the head and the yellow stripe widens behind the eye, Cuba (Wolfram Sander).

length lateral stripe of yellow. However, my review of hundreds of underwater photographs from the Caribbean shows that almost all *E. evelynae* (YB) show a reduction in the width of the yellow stripe behind the head or the yellow stripe fades and disappears before reaching the mid or posterior body (Fig. 10). *E. genie* has a wider lateral light band, which often appears bright white, and, if they have a yellowish stripe, it widens abruptly behind the head (Fig. 11), note that *E. genie* also lacks the pale middorsal streak (Fig 12). *E. genie* is found only in the Bahamas and Cuba. As noted by Colin (1975; 2010), *E. evelynae* and *E. genie* are most distinct in the northern Bahamas; in the southern part of the Bahamas there are occasional intermediate-appearing individuals. The DNA lineages of *E. genie* and *E. evelynae* are not close, and the blue Neon Goby of Florida, *E. oceanops*, and the blue-striped sibling species on the Mesoamerican Barrier Reef, *E. lobeli*, lie between the two lineages. The status of intermediate appearing individuals in the southern Bahamas needs to be further explored in light of this genetic difference— it may well be that some other characters are more consistent with species identity than color pattern.



Figure 12. *Elacatinus genie*, no pale middorsal streak and the yellow stripe widens behind the eye, Bahamas (Jan Morton)..

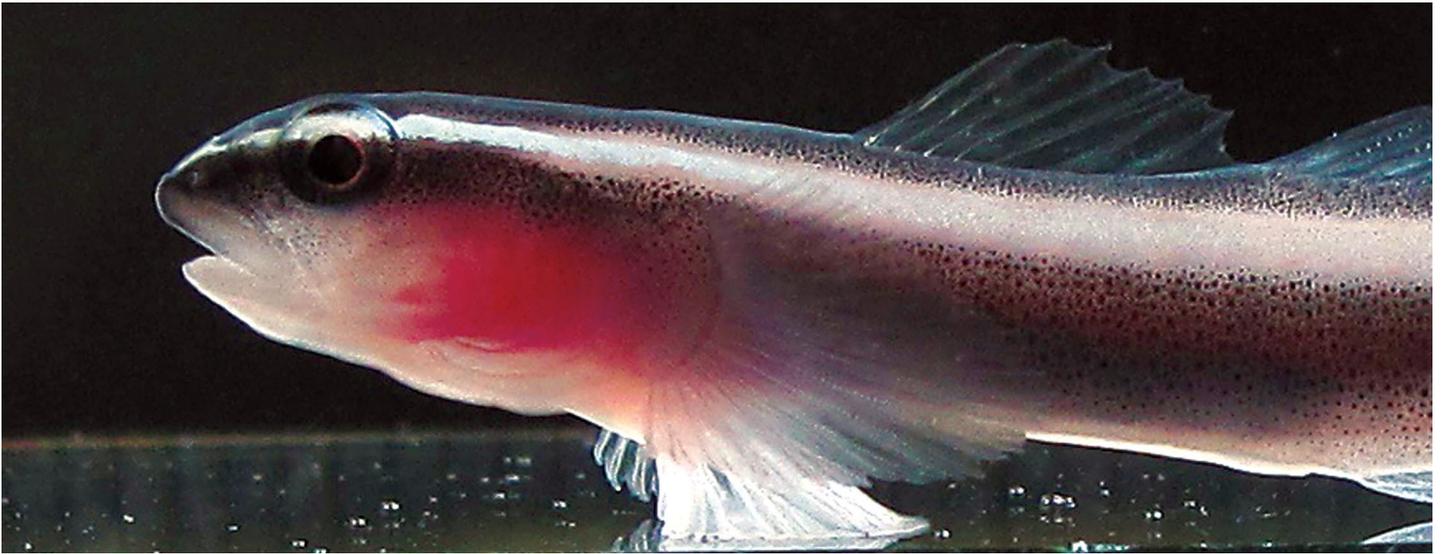


Figure 13. *Elacatinus prochilos*, Xcalak, Quintana Roo, Mexico (Jose A. Cohuo & Lourdes Vasquez-Yeomans).

In fact, Colin states that *E. genie* in the northern Bahamas have a swim bladder that is lacking in other members of the genus.

The placement of the mouth in *E. genie* and *E. cayman* has elicited some confusion in the literature. Although frequently used in keys, the “subterminal” and “inferior” terminology for mouth placement sometimes has been used interchangeably. Technically subterminal includes inferior, but is apparently used *sensu stricto* to mean the upper jaw is terminal and the lower jaw not terminal. However, subterminal is frequently diagrammed in various ichthyological literature with the snout overhanging the upper jaw. In the cleaner goby literature, the terms have been confused since Böhlke & Robins’ (1968) dichotomous key elevated the upper-lip frenum to a higher couplet and *E. genie* was appended to an alternative group with strictly subterminal mouths. Nevertheless, they and Colin (1975) state that both *E. genie* (including *E. cayman* as paratypes) and *E. evelynae* have “distinctly inferior” mouths, yet Colin (2010) includes *E. genie* within the category characterized by a subterminal mouth (Suite 2, his Figure 4) versus *E. evelynae*’s inferior mouth (Suite 1, his Figure 2), and adds that *E. cayman* could be considered to be either a member of Suite 1 (inferior mouth) or Suite 2 (subterminal mouth). Apparently, he follows Böhlke & Robins’ (1968) key conflating the absence of the upper-lip frenum as an alternative state to subterminal mouth placement to join Suite 2, but uses “subterminal mouth” as the title for the category in his Figure 4. It is unclear why *E. genie* or *E. cayman* should be associated in any way with *E. prochilos* and *E. randalli*, which have clearly terminal upper lips (Figs. 13 & 14) and are only distant relatives by mtDNA sequence.



Figure 14. *Elacatinus randalli*, Chichirivichi de la Costa, Venezuela (Mauro Ristorto).

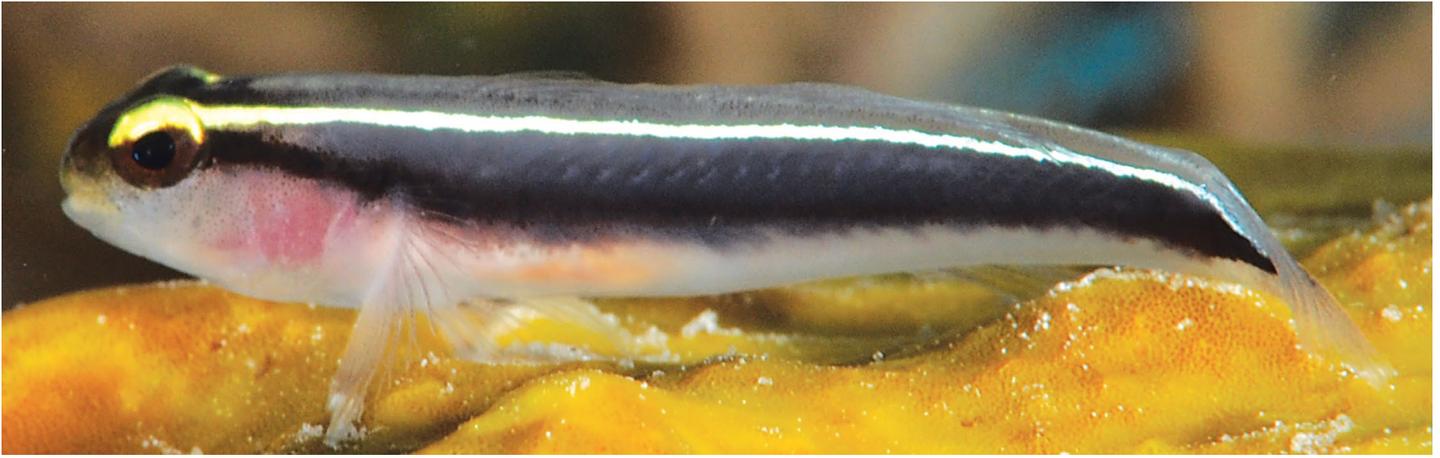


Figure 15. *Elacatinus centralis*, Grand Cayman (Frank Krasovec).

***Elacatinus centralis*, n. sp.**

Cayman Sponge Goby

Figures 15–20.

Gobiosoma (Elacatinus) horsti (non Metzelaar) [in part] Böhlke & Robins 1968: 78.

Gobiosoma horsti (non Metzelaar) Colin 1975: 87.

Gobiosoma (Elacatinus) horsti (non Metzelaar) Burgess *et al.* 1994: 220.

Gobiosoma horsti (non Metzelaar) Pattengill-Semmens & Semmens 2003: Appendix A.

Elacatinus horsti (non Metzelaar) Taylor & Hellberg 2006: 697.

Elacatinus horsti (non Metzelaar) Colin 2010: 9.

Holotype. UF 237059, 37.9 mm SL, male, SW Grand Cayman Island, 19.286° N, -81.393° W, April 28, 2014.

Paratypes. UF 237060, (6) 12.7–32.5 mm SL, same data as holotype; SIO 14-111, (3) 11.2–23.6 mm SL, same data as holotype; SIO 14-113, 26.5 mm SL, Bloody Bay, Little Cayman Islands, 19.687° N, -80.074° W, May 2, 2014.



Figure 16. *Elacatinus centralis*, Grand Cayman (Everett Turner).

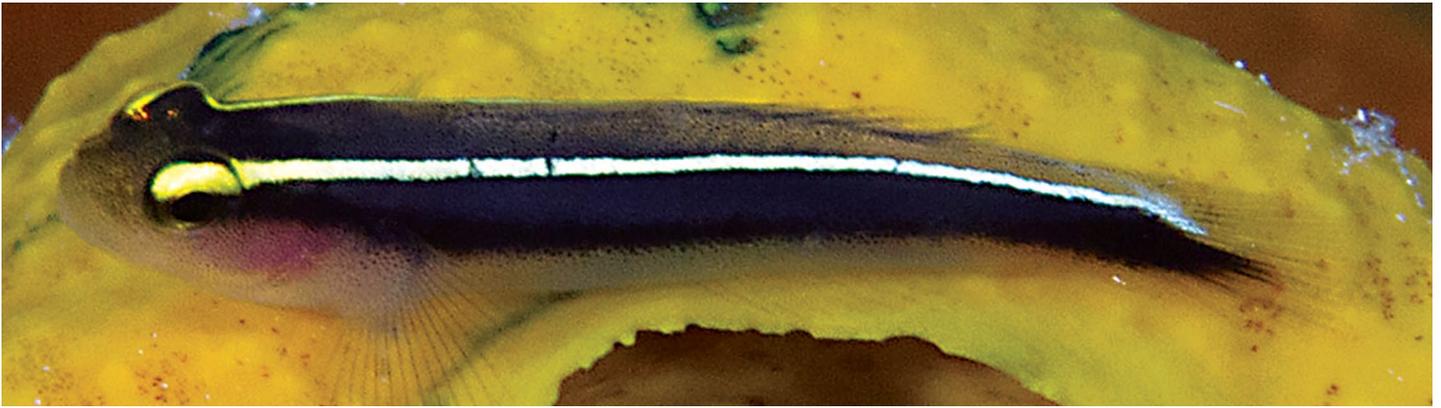


Figure 17. *Elacatinus centralis*, Grand Cayman (Mark Rosenstein).

Diagnosis. Dorsal rays VII+12; anal rays 11; pectoral rays 17–19; pelvic rays I,5 and fused into a cup; no scales; mouth terminal to subterminal (not inferior); complete upper lip groove (no frenum); upper head and body black, lower half of head and lower third of body white; a thin lateral stripe of yellow to bluish white divides black upper body, running from upper orbit onto caudal fin, widest and yellow on orbit, grading into bluish white and narrowing at tail; in smaller fish, adjacent strip above and below unpigmented; snout and front of upper lip distinctly dark with a yellow tinge. New recruits and juveniles have a full-length yellow to white stripe. Large individuals are mostly black with a thin yellow stripe that variably tends to grade into white near tail (Fig. 18). *E. horsti* of the southern Caribbean is distinguished by an all-yellow stripe, closely flanked with black, a pale yellowish snout, often with a centrally located bright yellow spot, and small juveniles with a short stripe (like *E. chancei*), i.e. only on head and over pectoral fin. *E. louisae* (type location Cayman Islands) has a similar stripe to *E. centralis* (yellow on head grading to bluish white), but has a rounded bright yellow spot on snout and black mid-lateral band ends in a rounded expansion on caudal fin blocking off end of light stripe.

Description. Dorsal rays VII+12; anal rays 11 (rare 10); pectoral rays 17–19 (uncommon 17); pelvic rays I,5 and fused into a cup; 11 precaudal and 17 caudal vertebrae from Böhlke & Robins (1968). No scales, body covered in a transparent mucus layer visible as a double line on profile in photographs and solidifying into a gray translucent skin on alcohol-preserved specimens.

Body elongate, body depth 18–22 (21)% SL, compressed, body width 11–13 (13)% SL; predorsal distance 34–37 (34)% SL; prepelvic distance short, 28–31 (31)% SL; preanal distance 56–60 (56)% SL; caudal peduncle long and relatively deep, length 21–25 (22)% SL, depth 12–14 (13)% SL. Head short, head length 25–27 (26)% SL; eye large, orbit diameter 25–31 (26)% HL; snout short and blunt, snout span 13–21 (13)% HL; mouth terminal, upper jaw tip barely in front of lower; mouth large, corner of gape with mouth closed extending well beyond vertical through rear of orbit in largest adults, up to rear edge of pupil in other large adults, upper jaw oblique



Figure 18. *Elacatinus centralis*, Grand Cayman (Everett Turner).

length 35–44 (32)% HL; lips thick, divided from snout by a complete groove, no frenum eliminating groove at midline. Tongue truncate. Teeth in each quadrant a pad of tiny sharp caniniform teeth, mostly anterior, with two spaced large fangs midway back, rear one larger in males; one or two variable relatively sized large canines in each quadrant in females. Anterior nostril in a short membranous tube overhanging upper lip, posterior nostril well in front of orbit and just anterior to nasal pore (anterior supraorbital of Böhlke & Robins [1968]). Pores on head comprise Birdsong's B' C D E F H' N' (B' particularly large and M' is not grossly apparent on large types).

Two dorsal fins; first spinous and short with slender flexible spines, no extended spines in either sex, membrane of last dorsal spine connecting to base of second dorsal fin spine in large males, before dorsal fin in smaller adults, third dorsal spine barely longest 13–16 (16)% SL; second dorsal fin medium length, base 23–29 (28)% SL, spine slender and flexible, shorter than rays 11–14 (11)% SL, first soft ray or two unbranched, third soft ray usually longest 13–19 (17)% SL, last split to base; anal fin shorter than dorsal, base 15–20 (21)% SL, spine slender and flexible, shorter than rays 7–8 (6)% SL, first soft ray or two unbranched, fourth soft ray usually longest 14–16 (14)% SL, last split to base; pectoral fin short, upper and lower two rays unbranched, about 11th ray longest, 23–25 (21)% SL; pelvic fins fused into a cup, all rays deeply branched, short, 18–21 (17)% SL, reaches two-thirds of the way to anus; caudal fin truncate, length 22–29 (26)% SL, 17 segmented caudal-fin rays, 2–3 uppermost and



Figure 19. **top:** *Elacatinus centralis*, new recruit with complete stripe and dark snout, Grand Cayman (Everett Turner); **middle:** *Elacatinus horsti*, new recruit with short stripe and yellow snout, Bonaire (Rick Coleman); **bottom:** *Elacatinus chancei*, new recruit with short stripe and yellow snout, Nevis (Jim Garin).

2–3 lowermost unbranched, 7–9 upper and 6–9 lower procurrent rays (usually one or two more upper or same number). No scales. Genital papilla in males a broad tapering cone, up to pupil diameter; females with a shorter wider cone with fimbriated rim.

Note that counts and morphology cannot be compared with the set reported by Böhlke & Robins (1968) in their description for “*E. horsti*”, since they combined populations from Bahamas, Cayman Islands, and Haiti; examined only two specimens of true *E. horsti* from the southern Caribbean; and included a specimen of *E. xanthiprora* from the Florida Middle Grounds as well.

Color in life. Upper head and body black, lower half of head and lower third of body white (in smaller adults); a thin lateral stripe of color divides black upper body, running from upper orbit onto caudal fin, widest and yellow on orbit, grading into bluish white and narrowing at tail; adjacent strip above and below on body unpigmented in juveniles and smaller adults; snout and front of upper lip distinctly dark with a yellow tinge. New recruits and juveniles have a full-length stripe, clearly yellow grading into white. Large individuals become mostly black with a thin yellow stripe that variably tends to grade into white near tail. All fins other than caudal fin are transparent, becoming dusky in large adults.

Color in alcohol. All color is lost and reverts to pale, leaving the black on white pattern intact. The colored stripe can be seen as a reflective line with melanophores after preservation.

Etymology. The specific epithet is a third declension masculine singular adjective in the nominative case, referring to the central Caribbean location of the Cayman Islands.

Barcode DNA sequence. The 652-nucleotide sequence of the segment of the mitochondrial COI gene used for barcoding by the Barcode of Life informatics database (BOLD)(Ratnasingham & Hebert 2007) was obtained for the holotype and paratypes (Appendix 2). Following the database management recommendation of the BOLD, the sequence of the holotype (GenBank accession number KM894156) is presented here as well:

```
CCTCTACTTAATTTTTGGTGCATGAGCCGGAATGGTCGGCACAGCTCTTAGCCTTCTAATCCGGGCT
GAACTAAGCCAACCCGGGGCCCTTTTGGGTGATGATCAGATTTATAATGTGATCGTTACTGCACATG
CTTTTGTAATAATTTTTTCATAGTAATACCCATCATAATTGGGGGCTTCGGAAACTGGCTGATCCCA
CTTATGATTGGTGCCCCGACATGGCATTCCCCCGTATAAATAATATAAGCTTTTGATTACTTCCTCCC
TCTTTCCTTTACTTCTTGCCCTCCTCTGGGGTTGAGGCCGGCGCTGGCACAGGTTGAACTGTGTATC
CCCCCTAGCCGGCAACTTAGCCCATGCTGGGGCATCTGTTGACTTAACGATCTTTCCCTTCACCT
GGCCGGAATTTCTTCAATCTTGGGGGCAATCAACTTTATCACTACTATCATGAACATGAAGCCGCCT
GCAGTATCTCAATATCAAACCCCTATTTGTATGAGCCGTCCTAATCACAGCCGTGCTTCTTCTCCT
TTCACCTTCTGTCCTCGCAGCCGGCATTACTATGCTTCTCACAGATCGAAACTTAAACACCACCTTT
TTGACCCTGCAGGCGGAGGAGACCCCATCCTTTATCAACATCTA
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Comparisons. The Cayman Island population of the striped sponge goby has variously been considered a yellow-striped form or a white-striped form (Colin 2010), and, indeed, it is actually a yellow-white (YW) form. In the past it was considered to be conspecific with the yellow-striped *E. horsti*, described originally from Curaçao and native only to the southern Caribbean. However, *E. centralis* is further genetically from *E. horsti* than is the Shortstripe Goby, *E. chancei*, which is found in the intervening range (from the southern Bahamas down through the Puerto-Rican Plateau and the Lesser Antilles to Venezuela, beyond which it is replaced by *E. horsti*). In fact, *E. horsti* and *E. chancei* share the character of a shortened stripe in small juveniles, which elongates into a complete yellow stripe in *E. horsti*, but stays short in *E. chancei*. Interestingly, *E. horsti* and *E. chancei* have overlapping COI mtDNA lineages, despite color differences and fin-ray count differences: one of the more prominent local examples of reef fish species that share mtDNA sequences.

The neighbor-joining phenetic tree based on the COI mtDNA sequences of the sponge gobies of *Elacatinus* (including all species known from the region except *E. tenox* and *E. serranilla* [Randall & Lobel 2009]), following the Kimura two-parameter model (K2P) generated by BOLD (Barcode of Life Database), shows that *E. centralis* is in a distinct clade 3% (K2P minimum interspecific distance, 2.8% pairwise) from the nearest lineage, which contains both *E. horsti* and *E. chancei* intermingled (Fig. 20). *E. aff. horsti* from the Bahamas is more distant, with a minimum interspecific distance of 4.5% (4.3% pairwise). A multigene phylogeny by Rüber *et al.* (2003)

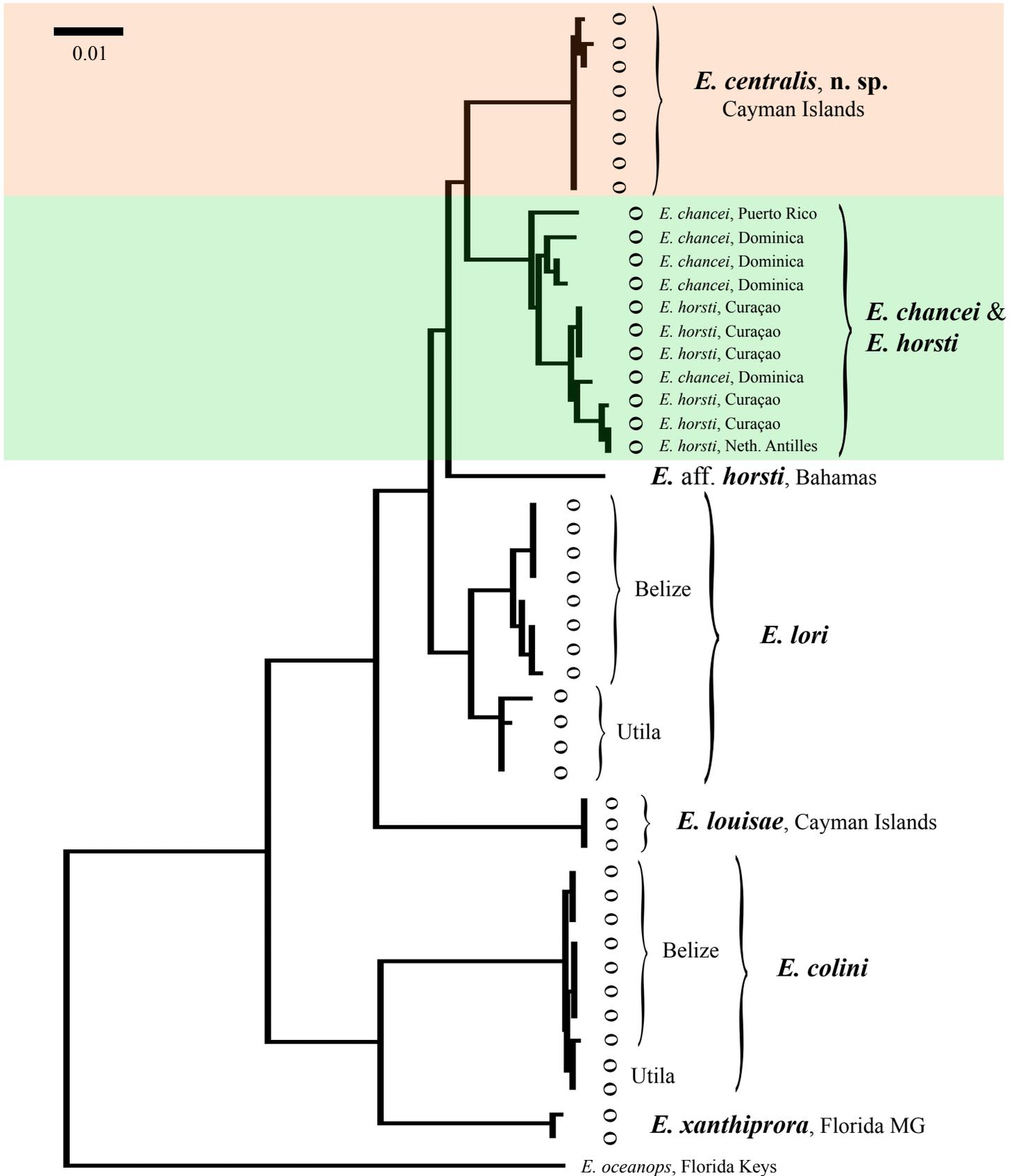


Figure 20. The neighbor-joining phenetic tree of all but two of the W. Atlantic striped sponge gobies of *Elacatinus* following the Kimura two-parameter model (K2P) generated by BOLD (Barcode of Life Database). The scale bar at left represents a 1% sequence difference. Collection locations for specimens are indicated, and the goby *Elacatinus oceanops* is used as an outgroup. GenBank accession numbers and collection data for the sequences in the tree are listed in Appendix 2 in the same order as the sequences in the tree.



Figure 21. *Elacatinus horsti*, yellow snout and all-yellow stripe, Curaçao (Jamie Holdorf).

found *E. centralis* to be further from true *E. horsti* than is *E. chancei*, however, for some unexplained reason the specimen from Cayman Islands is called “*E. lori*”, despite the fact that the range for *E. lori* is then listed as “Gulf of Honduras” (Rüber *et al.* 2003: Appendix). Taylor & Hellberg (2005; 2006) studied the sponge gobies with a different mitochondrial marker (and a nuclear marker) and similarly found *E. centralis* (as “*E. horsti*” from Cayman) to be relatively distant from its nearest relatives, an intermingled grouping of Curaçao *E. horsti* and *E. chancei* from the Lesser Antilles. They also found the Bahamas population of *E. aff. horsti* to be more distant from *E. centralis* than the *E. horsti/E. chancei* clade.

Several features of coloration distinguish *E. centralis* from *E. horsti*. *E. centralis* has the complete stripe from settlement (newly settled fish, about 7.5 mm SL, collected with the types, had the complete stripe), while *E. horsti*

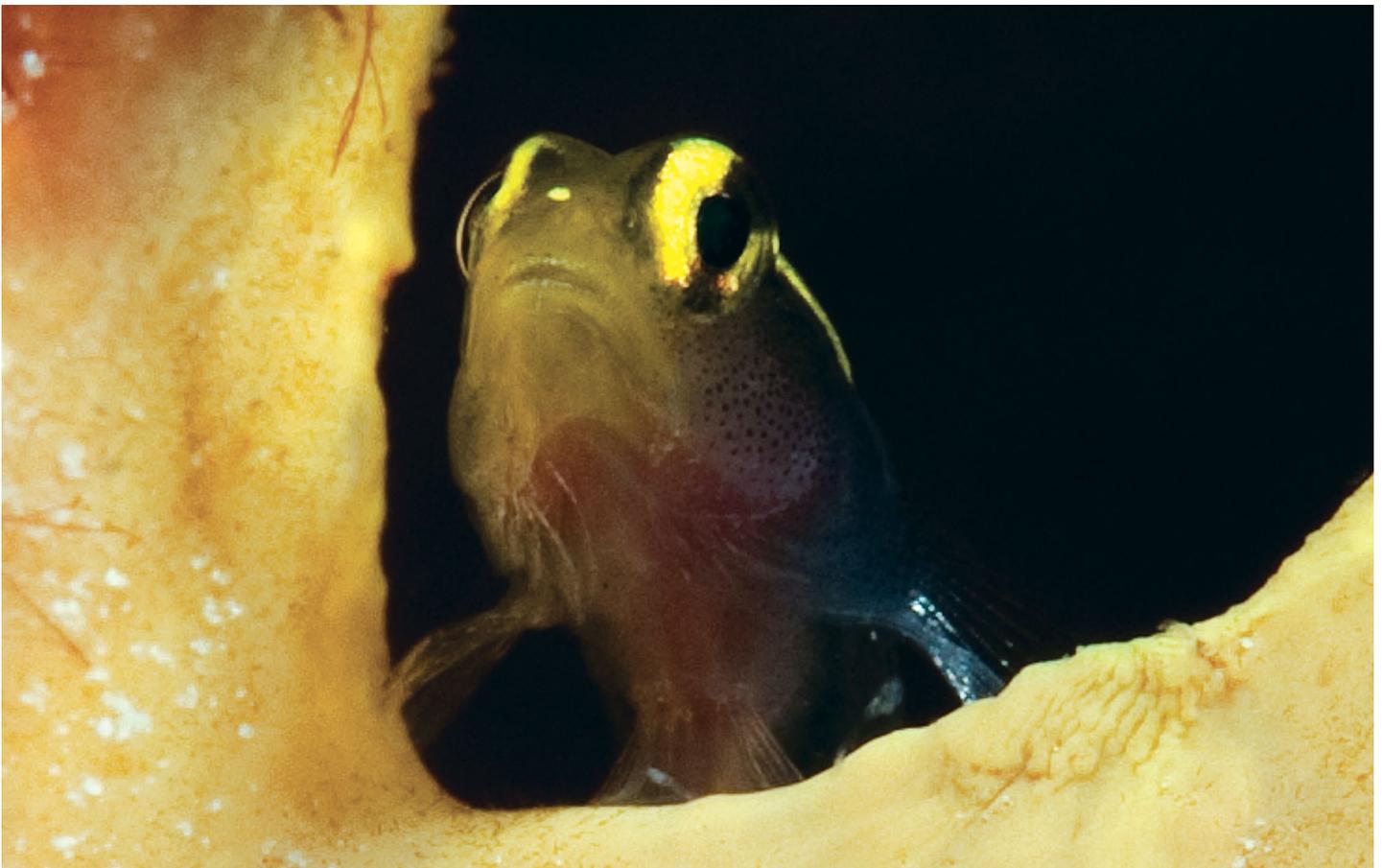


Figure 22. *Elacatinus horsti*, yellow snout with accessory spot, Curaçao (Jamie Holdorf).

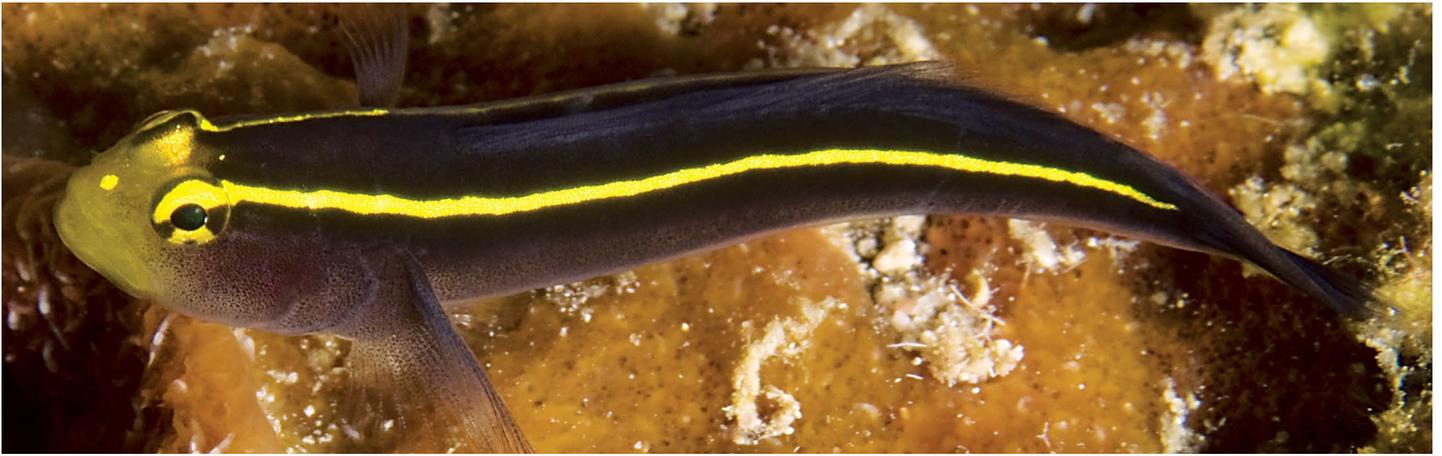


Figure 23. *Elacatinus horsti*, yellow snout with accessory spot and all-yellow stripe, Curaçao (Barry Brown).

has a short stripe in the smallest juveniles (Fig. 19). The stripe of *E. centralis* has a yellow-to-white transition, particularly noticeable in smaller fish, but present to some degree in most large individuals, while *E. horsti* has the stripe fully yellow. *E. centralis* has a dark snout and upper lip vs. a pale yellowish snout with no melanophores on the snout or upper lip in *E. horsti* (Figs. 21–24). Smaller *E. centralis* also have an unpigmented or less pigmented band flanking the yellow stripe (black strip pulls away right after the eye (Figs. 15 & 16) or at least before the pectoral fin), while *E. horsti* have black touching the yellow stripe (e.g. Figs. 21 & 23), or even boldly outlining the yellow stripe (Fig. 24). In addition, *E. horsti* frequently shows an accessory bright yellow spot on the snout, evident on about half of the underwater photographs I have seen (Figs. 22 & 23)(vs. rarely in *E. centralis*, only a single fish observed).

E. chancei is easily distinguished by having a short stripe (Figs. 25 & 26). As expected, perhaps, from the genetic similarity, *E. chancei* shares with *E. horsti* the pale yellow snout, black outlining the stripe, and the occasional accessory snout spot (Fig. 26).

The phenotypic distinction between *E. centralis* and the *E. aff. horsti* population of the Bahamas remains to be resolved; both yellow and yellow-white striped sponge gobies have been photographed in the Bahamas and their affiliations are uncertain without additional sampling. Analogous to the case for the cleaner goby *E. cayman*, the neighboring populations of striped sponge gobies, i.e. from Jamaica, Navassa, and San Andres, are typically white striped (Colin 2010). The shallow striped sponge goby from the Bay of Honduras, *E. lori*, is white striped (Colin 2002), while the cleaner goby there, *E. lobeli*, is blue striped (Randall & Colin 2009).

A second sponge goby, the Spotlight Goby *E. louisae*, is found sympatrically with *E. centralis* (the type location is also Cayman Islands). It has a similar stripe to *E. centralis* (yellow on head grading to bluish white),

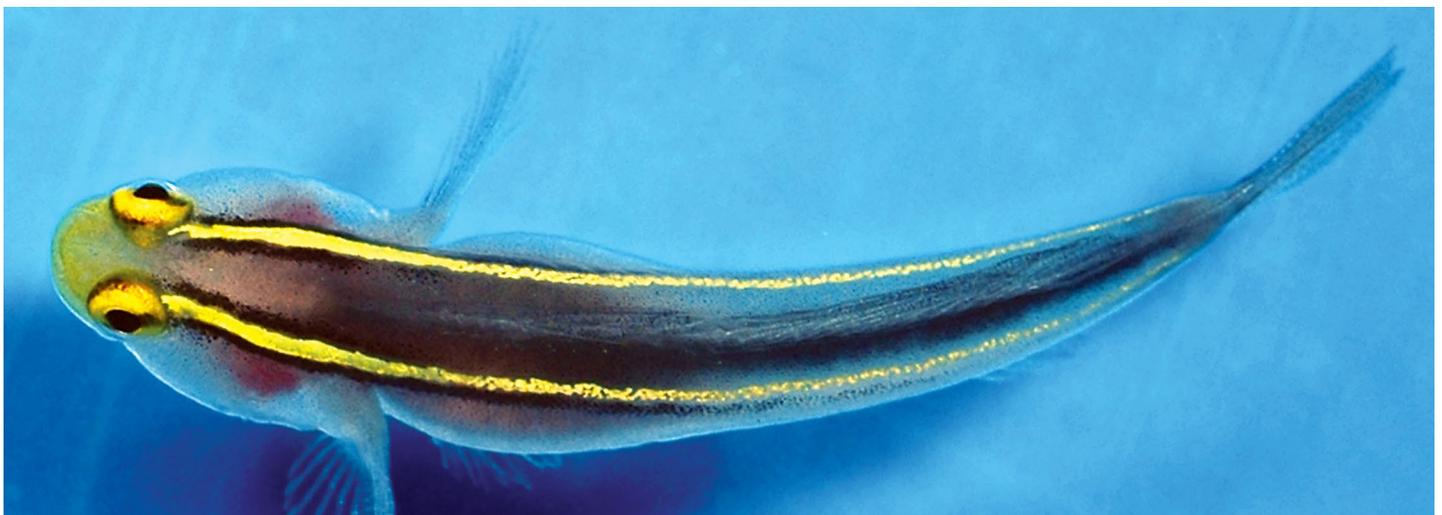


Figure 24. *Elacatinus horsti*, pale snout, outlined yellow stripe, aquarium trade (Dustin Dorton).

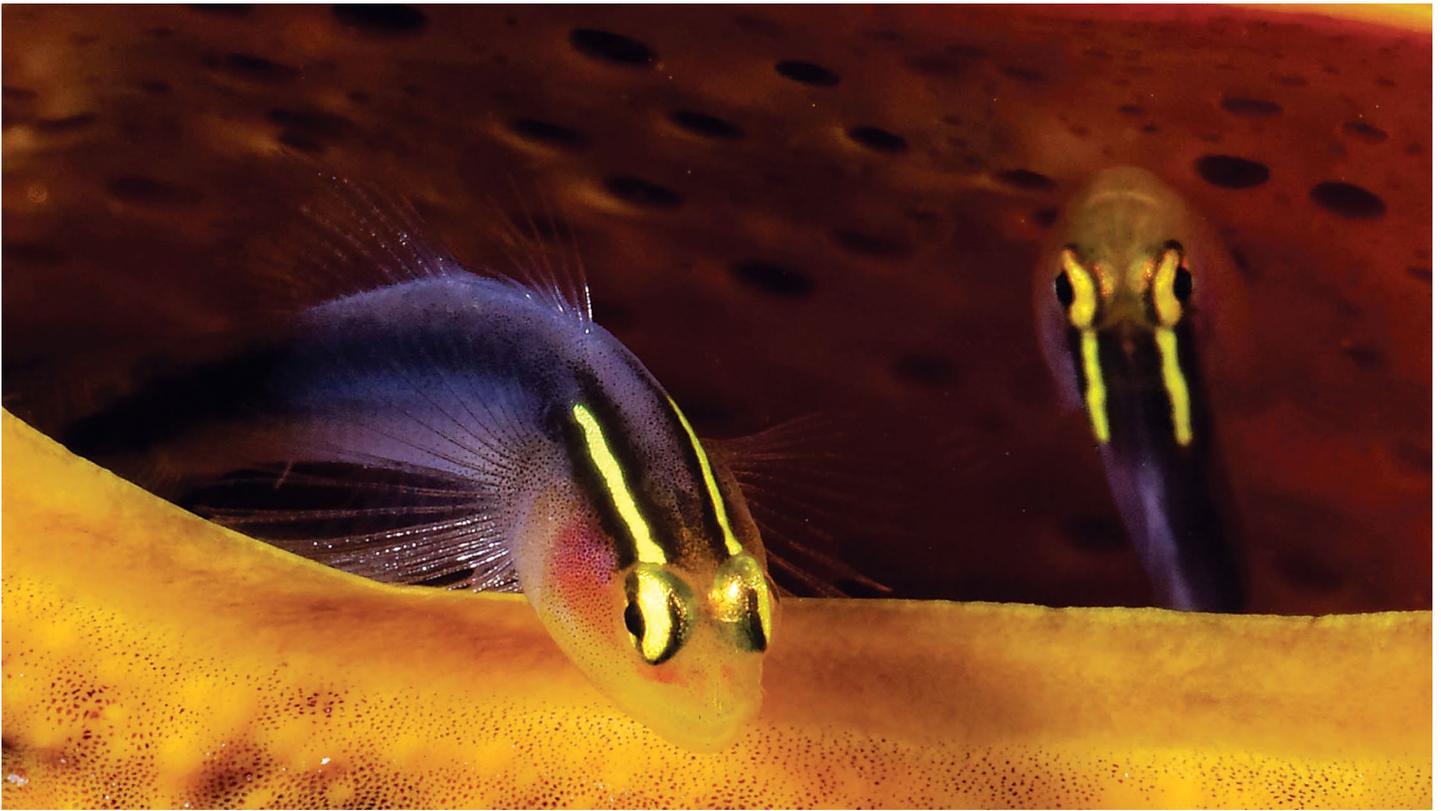


Figure 25. *Elacatinus chancei*, large adults with short stripes, Nevis (Jim Garin).

but has a rounded yellow spot on the snout and the black mid-lateral band ends in a rounded expansion on the caudal fin blocking off the end of the light stripe (Figs. 27 & 28).

Lastly, in a very interesting observation, first noted by Colin (1975), the color pattern of the various cleaner and sponge gobies tend to coincide in any specific location. Indeed, in the Cayman Islands, *E. centralis* and *E. louisae* both have the YW stripe, which is not too different from the yellow stripe over the bluish white band

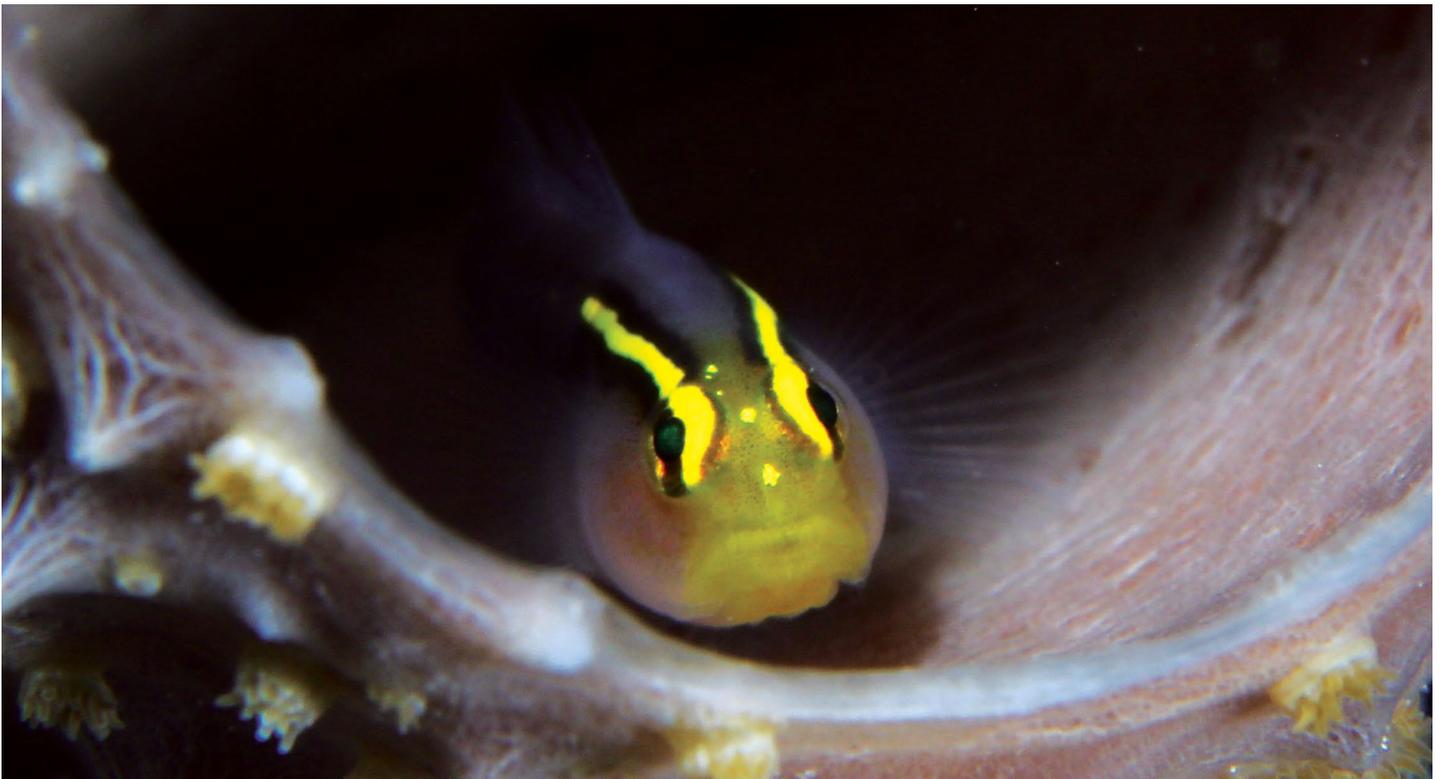


Figure 26. *Elacatinus chancei*, accessory bright yellow spots on snout, Nevis (Mark Lessard).

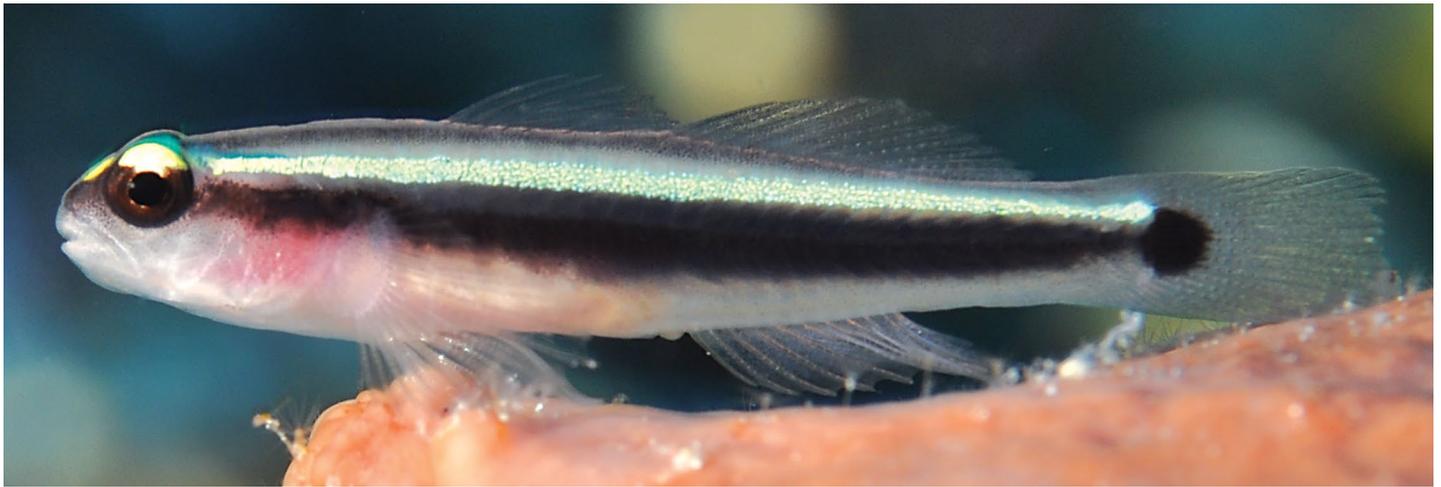


Figure 27. *Elacatinus louisae*, Grand Cayman (Frank Krasovec).

characteristic of the local cleaner goby, *E. cayman*. Whether this is due to protective mimicry by the sponge gobies taking advantage of the relative immunity conferred by predators to the symbiotic cleaner gobies, or the reverse, Batesian mimicry by the cleaner gobies taking advantage of the predator avoidance due to the toxic mucus of the sponge gobies, or both, is presently unknown. The two sponge gobies sharing color patterns would be an example of Müllerian mimicry, if they both have toxic mucus.



Figure 28. *Elacatinus louisae*, Grand Cayman (Cindy Abgarian).



Figure 29. *Tigrigobius harveyi*, Grand Cayman (Everett Turner).

***Tigrigobius harveyi*, n. sp.**

Cayman Greenbanded Goby

Figures 29–33.

Gobiosoma (Tigrigobius) multifasciatum (non Steindachner) [in part] Böhlke & Robins 1968: 66.

Gobiosoma (Tigrigobius) multifasciatum (non Steindachner) Burgess *et al.* 1994: 220.

Holotype. UF 237061, 20.7 mm SL, male, SW Grand Cayman Island, 19.286° N, -81.391° W, April 29, 2014.

Paratypes. UF 237062, (14) 9.8–21.1 mm SL same data as holotype; SIO 14-112, (4) 16.7–18.9 mm SL, same data as holotype.

Diagnosis. Dorsal rays VII+11 (rare 12); anal rays 10; pectoral rays 20–21 (rare 22); pelvic rays I,5 and fused into a cup; no scales; mouth terminal; extended first dorsal spine in large males and females; pale head with a prominent red stripe from tip of snout across eye ending abruptly above pectoral-fin base with a short orange segment, followed by a short black segment; no cheek stripe; a dark green body with 25 (20–28) thin light green bars along full length of body onto caudal fin base (43–55, both sides added together).

Description. Dorsal rays VII+11 (rare 12, in 2 of 19 types); anal rays 10; pectoral rays 20–21 (rare 22, 2/19 types); pelvic rays I,5 and fused into a cup; 11 precaudal and 17 caudal vertebrae from Böhlke & Robins (1968). No scales, body covered in a transparent mucus layer solidifying into a gray translucent skin on alcohol-preserved specimens.

Body elongate, body depth 21–26 (20)% SL, compressed, body width 15–19 (17)% SL; predorsal distance 36–40 (37)% SL; prepelvic distance short, 32–37 (33)% SL; preanal distance 63–68 (62)% SL; caudal peduncle length 17–22 (21)% SL, depth 11–12 (12)% SL. Head short, head length 29–30 (29)% SL; eye large, orbit diameter 25–29 (23)% HL; snout short and blunt, snout span 16–19 (18)% HL; mouth terminal, upper jaw tip



Figure 30. *Tigrigobius harveyi*, Grand Cayman (Everett Turner).

barely in front of lower; mouth large, corner of gape with mouth closed extending well beyond vertical through rear of orbit in largest adults, up to rear edge of pupil in other large adults, upper jaw oblique length 34–44 (47)% HL; lips thick, divided from snout by a complete groove. Tongue truncate. Teeth in each quadrant a pad of small sharp caniniform teeth, largest arrayed in front row, on lower jaw outer row increasingly large rearward with largest at about midpoint. Anterior nostril in a short membranous tube overhanging upper lip, posterior nostril well in front of orbit and just anterior to nasal pore (anterior supraorbital of Böhlke & Robins [1968]). Pores on head comprise Birdsong's B' C D E F H' M' N'.



Figure 31. *Tigrigobius harveyi*, Grand Cayman (Everett Turner).



Figure 32. **top:** *Tigrigobius multifasciatum*, aquarium trade specimen (Jim Burke); **middle:** *Tigrigobius rubrigenis*, Utila, Honduras (Keri Wilk); **bottom:** *Tigrigobius harveyi*, Grand Cayman (Everett Turner).

Two dorsal fins; first spinous and short with slender flexible spines, extended first spines in largest individuals of both males and females, membrane of last dorsal spine connecting to body before origin of second dorsal fin, first spine longest 19–27 (29)% SL, next two or three about equal, third dorsal spine 13–16 (16)% SL; second dorsal fin medium length, base 24–26 (27)% SL, spine slender and flexible, shorter than rays 11–14 (12)% SL, all soft rays branched, fourth soft ray usually longest 15–17 (16)% SL, last split to base; anal fin shorter than dorsal, base 15–17 (19)% SL, spine slender and flexible, shorter than rays 8–10 (8)% SL, all soft rays branched, fourth soft ray usually longest 14–16 (14)% SL, last split to base; pectoral fin short, uppermost ray unbranched, about 12th ray longest, 24–28 (26)% SL; pelvic fins fused into a cup, all rays deeply branched, very short, 15–17 (15)% SL, reaches about half way to anus; caudal fin truncate, length 23–24 (21)% SL, 17 segmented caudal-fin rays, usually only the uppermost and lowermost unbranched, 6–8 upper and 5–7 lower procurent rays (modal 7/6, usually one more upper than lower, or same). No scales. Genital papilla in males a short broad tapering cone, females with a shorter wider cone with fimbriated rim.

Color in life. Juveniles and adults of both sexes similarly marked. Head is pale with a single prominent red stripe running from tip of snout across eye ending abruptly above pectoral-fin base with a short bright-orange segment followed immediately by a short black segment. Background body color is olive green, sometimes dark greenish black, with a mode of 25 (20–28) thin light green bars along full length onto base of caudal fin, added both sides range from 43–55, mean 48.5; narrow bars usually run vertically over full depth of body, but many individuals have unique variations, such as bifurcations, cross-links, or breaks in a pattern reminiscent of fingerprint ridges (bars counted if greater than 1/3 of full depth). All fins transparent, becoming dusky in larger adults.

Color in alcohol. All color is lost, replaced by shades of gray, but characteristic bars remain distinct. Anterior body is darker gray, median fins dusky, darker at distal edge of membranes of anal fin and, to a lesser degree, dorsal fin. Head stripe barely remains as a band of lighter shading.

Etymology. The specific epithet is a noun in the genitive case in recognition of Dr. Guy Harvey's extensive support for research and conservation of sharks and gamefishes in the region. He established the Guy Harvey Research Institute (GHRI) at Nova Southeastern University in Fort Lauderdale, Florida as well as the Guy Harvey Ocean Foundation and has a particular interest in the fishes of the Cayman Islands, where he makes his home.

Barcode DNA sequence. The 652-nucleotide sequence of the segment of the mitochondrial COI gene used for barcoding by the Barcode of Life informatics database (BOLD)(Ratnasingham & Hebert 2007) was obtained for the holotype and paratypes (Appendix 3). Following the database management recommendation of the BOLD, the sequence of the holotype (GenBank accession number KM894141) is presented here as well:

```
CCTTTACCTAGTCTTCGGTGCATGGGCTGGCATAGTCGGCACAGCACTCAGCCTTCTTATCCGGGCC
GAACTAAGTCAACCAGGAGCCTTGCTGGGGGACGACCAGATGTACAATGTAATTGTAACGGCTCAC
GCCTTTGTAATAATCTTTTTTATAGTAATACCAATTATGATTGGAGGATTTGGCAACTGACTAATCCCC
CTAATGATCGGAGCCCCCGACATAGCATTCCC GCGGATAAATAACATAAGCTTCTGACTTTTACCCCC
ATCTTTTCTATTGCTTCTTGCTTCTTCAGGGGTTGAATCTGGGGCGGGCACAGGGTGAACAGTCTAC
CCCCACTTGCAAGTAACCTAGCCCACTCAGGAGCATCCGTCGATTTGACCATCTTCTCACTTCACC
TGGCAGGAATTTCTTCTATCCTAGGAGCAATTAATTCATTACCACCATCATTAATATAAAACCCCTGG
GAACTACGCAGTATCAGACCCCCTGTTTGTATGGGCCGTCCTAATTACAGCAGTCCTCCTGCTCCTA
TCACTTCTGTACTGGCTGCCGGAATCACGATGCTTCTTACTGACCGAAATTTAAATACATCGTTCTT
TGATCCTGCCGGTGGAGGGGACCCAATCTTTATCAACATCTC
```

Comparisons. *Tigrigobius harveyi* is a member of a geographic complex of species originally described as *Gobiosoma multifasciatum* (Steindachner 1876), with the type population from St. Thomas, USVI and nearby St. Barthelemy in the French West Indies (now *Tigrigobius multifasciatus*). This species and its other barred relatives were placed in the subgenus *Tigrigobius* of the genus *Gobiosoma* by Böhlke & Robins (1968). Subsequently, for a time, the subgenus was considered part of a very expansive *Elacatinus*, but lately has been elevated again to the genus *Tigrigobius* (van Tassell 2011; Eschmeyer 2014).

Victor (2010) described the uniquely marked population of Greenbanded Gobies in the Bay of Honduras as the Redcheek Goby, now *Tigrigobius rubrigenis*, and the Panamanian population as *Tigrigobius panamensis*. The

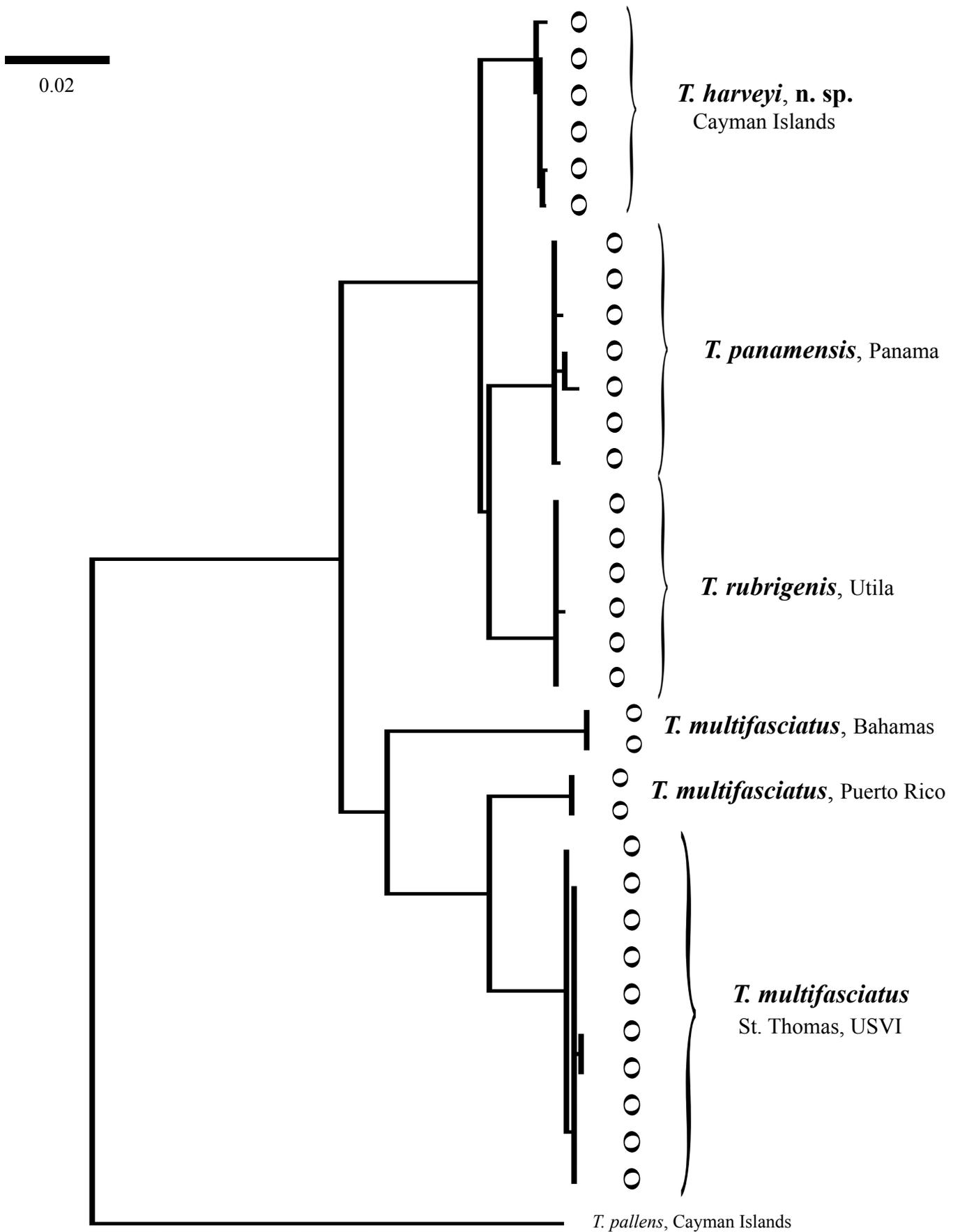


Figure 33. The neighbor-joining phenetic tree of the Greenbanded Goby complex in *Tigrigobius* following the Kimura two-parameter model (K2P) generated by BOLD (Barcode of Life Database). The scale bar at left represents a 2% sequence difference. Collection locations for specimens are indicated, and *Tigrigobius pallens* is used as an outgroup. GenBank accession numbers and collection data for the sequences in the tree are listed in Appendix 3 in the same order as the sequences in the tree.

species in the complex have particularly deep divergences in mtDNA COI. The neighbor-joining phenetic tree based on the COI mtDNA sequences of the Greenbanded Gobies, following the Kimura two-parameter model (K2P) generated by BOLD (Barcode of Life Database), shows that *Tigrigobius harveyi* is relatively close to the Central American species and far from the Antillean species (Fig. 33). *Tigrigobius harveyi* is about equidistant from the Panamanian Greenbanded Goby (3.3%, 3.2% pairwise) and the Redcheek Goby (3.5%, 3.4% pairwise) and 11% divergent from the Antillean Greenbanded Goby lineages.

Phenotypically, *Tigrigobius harveyi* is notable for its high count of green bands along the body, a mean of 25 (20–28). As shown in Fig. 32, *T. multifasciatum* has many fewer bands (15–21), as does *T. panamensis* (16–23), and *T. rubrigenis* has a similarly high number of green bars (23–26), but can be distinguished from all other species in the complex by a prominent red cheek stripe (Victor 2010). In addition, *T. multifasciatum* from the Antilles has about equal numbers of 11 and 12 second-dorsal-fin elements (Ginsburg 1933; Victor 2010), while the three western congeners have almost all with 11.

Phylogeography of Gobies

The present description of three new endemic species for the Cayman Islands confirms the suspicions of Burgess *et al.* (1994) who noted that the single endemic species at the time, *Starksia y-lineata*, would likely not prove to be the only Cayman endemic marine fish. Indeed, that labrisomid blenny is a member of a species complex that just happens to be prominently marked, leading to early recognition of its uniqueness (Gilbert 1965); no doubt, other species complexes of blennioids will prove to have endemic Cayman representatives similar to the pattern for gobies described here.

In addition to those of the new species, the mtDNA lineages of Cayman populations of some other gobies that were collected in the survey proved to be divergent from other populations in the Caribbean. In particular, *Tigrigobius dilepis*, *T. pallens*, and *Risor ruber* populations in Cayman were found to have distinct local lineages. These fishes, however, did not appear to have morphological, meristic, or marking differences and thus were considered to be genetically variant populations, or genovariants (*sensu* Victor, in press). The status of these genovariant populations could change depending on additional specimens, sequences, and collection locations or a more comprehensive analysis of variation in the species. Indeed, the very definition of species is not static and is broadly open to interpretation, depending on the question one is asking (Mallett 2008).

The existence of locally divergent mitochondrial lineages can provide an insight into patterns of connectivity on a genetic scale (relatedness) between populations in the region. This kind of analysis would be especially powerful if there were consistency across taxa, for example as found to some degree for the Mona Passage barrier to gene flow for cleaner and sponge gobies in Taylor & Hellberg (2006); the passage is a well-established faunal break for a number of reef fishes (Dennis *et al.* 2005). Clearly, it is critical to examine more taxa simultaneously to evaluate the reliability of such biogeographic patterns.

The six species that could be assessed (with limitations) in this study proved not to have any consistent pattern of geographic relatedness (Fig. 34). Despite the proximity to the Mesoamerican Barrier Reef in Belize and Honduras, only *E. centralis* had the closest relationship to that region (and that closeness to *E. lori* was not evident for a different mitochondrial marker in Taylor & Hellberg [2006]). For other species, the nearest lineages were from the Lesser Antilles, the Netherlands Antilles, or Panama. As one might expect for a complex phenomenon with many competing drivers, more data and additional species makes interpretation more difficult and often overwhelms facile comparisons made with one or two species.

Notably, a set of other goby genera did not have divergent local mitochondrial DNA lineages and their COI sequences were identical to other Caribbean populations. This pattern of genera varying in their tendency to break up into allopatric, or even sympatric, species complexes has been noted in other reef fish families (Victor, in press). The barcode mtDNA sequences obtained for eight species of *Coryphopterus* (see Table 1), as well as *Ctenogobius saepepallens*, *Gnatholepis thompsoni*, and *Priolepis hipoliti*, were the same as sequences for the same species from multiple other locations throughout the Caribbean Sea. It is likely that this reflects differing dispersal abilities to some degree, since gobies do vary greatly between species in attributes such as the pelagic larval duration, size at settlement, and habitat preferences.

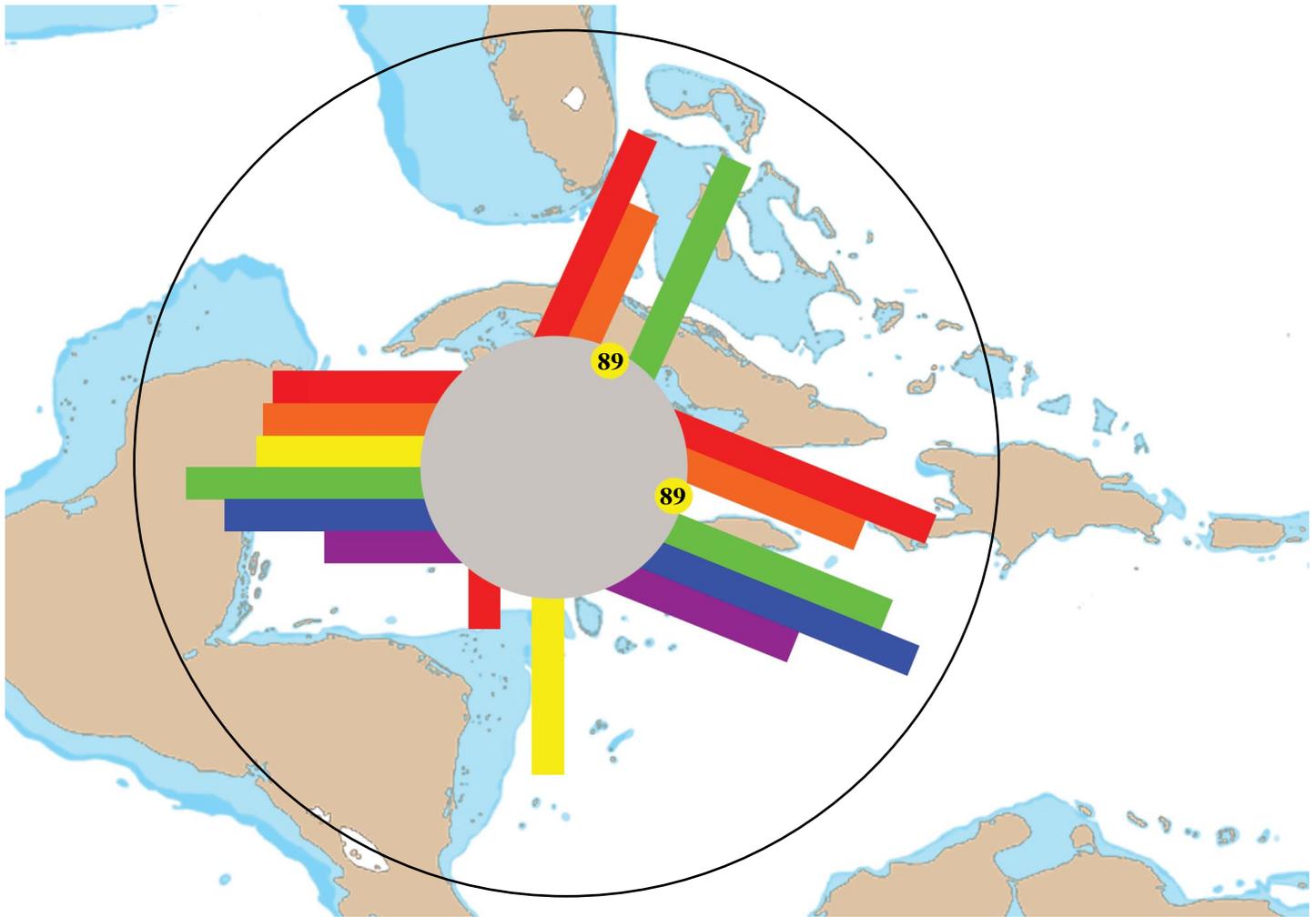


Figure 34. A “similarity bar graph”, i.e. a stylized graphic representation of genetic relatedness (long bars are more similar) of Cayman Island lineages to four other populations/species in the Greater Caribbean region: Bahamas to the NNE, Lesser Antilles to the ESE, Panama to the south and Belize/Honduras to the west. The bars represent the percent similarity above 90% (calculated as maximum percent similarity between two lineages, which equals one minus the minimum interspecific distance, by K2P nearest-neighbor method); bars that are absent means no data. The two yellow circles represent a similarity below 90%, i.e. 89% similarity of the Cayman population to both the Lesser Antilles and Bahamas populations. The longest bar is 99.5% similarity and the thin outer ring would be reached if 100% identical sequences (red is *Elacatinus cayman*, orange is *E. centralis*, yellow is *Tigrigobius harveyi*, green is *Risor ruber*, blue is *T. dilepis*, and violet is *T. pallens*).

Updated Species List of Gobies of the Cayman Islands

The two prior surveys of fishes of the Cayman Islands included 35 named species of gobies (Gobiidae) in Burgess *et al.* (1994) and 23 in Pattengill-Semmens & Semmens (2003). The two lists are somewhat different, but that would be expected since Burgess *et al.* (1994) review museum samples with many species that are collected with poison (e.g. *Lythrypnus*, *Psilotris*, and *Chriolepis*), and include tidepool/inshore/brackish water species (e.g. *Bathygobius*, *Lophogobius*, and true *Gobiosoma*), while the underwater surveys by REEF generally do not sample those difficult-to-observe species, but do pick up more sporadic or sand-based species that scientific collections may miss (e.g. *Microgobius carri*, *Nes longus*, and *Ptereleotris helenae*). Both listings combine the three widespread Caribbean cryptic species of bridled sand gobies (Victor 2008) as “*Coryphopterus glaucofraenum*”, although two, and likely all three, of the widespread species complex of *C. bol*, *C. tortugae*, and *C. glaucofraenum* are present. Both listings include two cleaner gobies, *Elacatinus evelynae* and *E. genie*, although there is no evidence, photographic or genetic, that more than the one Cayman Cleaner Goby, *E. cayman*, is present. The Burgess *et al.* (1994) listing includes *Chriolepis* n. sp. in addition to *C. fisheri* and *Priolepis* n. spp. in addition to *P. hipoliti*, yet the specimens collected and sequenced in this study, as well as for the region, do not support more than one

TABLE 1

Species List of Gobies of the Cayman Islands

Documented (*DNA barcoded)

<i>Bathygobius antilliensis</i>	<i>Ginsburgellus novemlineatus</i>
<i>Bathygobius lacertus</i>	<i>Gnatholepis thompsoni</i> *
<i>Bathygobius mystacium</i>	<i>Gobiosoma spes</i>
<i>Chriolepis fisheri</i> *	<i>Lophogobius cyprinoides</i>
<i>Coryphopterus alloides</i>	<i>Lythrypnus elasson</i>
<i>Coryphopterus bol</i> *	<i>Lythrypnus heterochroma</i>
<i>Coryphopterus dicrus</i> *	<i>Lythrypnus nesioties</i>
<i>Coryphopterus eidolon</i> *	<i>Lythrypnus okapia</i>
<i>Coryphopterus hyalinus</i> *	<i>Lythrypnus spilus</i>
<i>Coryphopterus kuna</i> *	<i>Oxyurichthys stigmalocephalus</i> (photo only)
<i>Coryphopterus lipernes</i>	<i>Priolepis hipoliti</i> *
<i>Coryphopterus personatus</i> *	<i>Psilotris alepis</i>
<i>Coryphopterus thrix</i> *	<i>Psilotris batrachoides</i>
<i>Coryphopterus tortugae</i> *	<i>Ptereleotris helenae</i> (photo only)
<i>Ctenogobius boleosoma</i>	<i>Risor ruber</i> *
<i>Ctenogobius saepepallens</i> *	<i>Tigrigobius dilepis</i> *
<i>Elacatinus cayman</i> *	<i>Tigrigobius gemmatum</i>
<i>Elacatinus centralis</i> *	<i>Tigrigobius harveyi</i> *
<i>Elacatinus louisae</i> *	<i>Tigrigobius pallens</i> *

Expected**Visual report only**

<i>Bathygobius soporator</i>	<i>Microgobius carri</i>
<i>Bathygobius curacao</i>	<i>Nes longus</i>
<i>Coryphopterus glaucofraenum</i>	

Doubtful or vagrant

<i>Elacatinus genie</i>
<i>Elacatinus chancei</i>
<i>Elacatinus xanthiprora</i>
<i>Elacatinus evelynae</i>
<i>Tigrigobius saucrum</i>

representative at each locality for these two genera (except the SE Caribbean where two *Priolepis* species are found). Indeed, the museum records show that the “sp.” annotations are from different “dates of modification” of the records and never occur in the same collection with the named species, i.e. likely an artifact. Two species have visual records from REEF, but no photographs or specimens. The REEF observations include rare sightings of species that are not previously recorded and not expected in the Cayman Islands, which may represent either misidentifications or vagrants, and would require specimens or photographs to resolve (i.e. *Elacatinus chancei*, *E. xanthiprora*, and *Tigrigobius saucrum*). Extensive underwater photography, especially by Everett Turner and Cindy Abgarian, has confirmed most species and provides records for two species that have never been collected (Figs. 35 & 36). Lastly, the recently described tiny Kuna Goby, *Coryphopterus kuna* Victor 2007, which has now been found almost everywhere in the Caribbean (when it is carefully looked for), was collected in my Cayman Islands survey and DNA-sequenced for this study.

A revised listing of gobies for the Cayman Islands is presented in Table 1, with 38 documented species and perhaps four more to be expected. The records of Burgess *et al.* (1994) are considered documented, unless the genus has been revised or additional biogeographic information is available. Their records have not been further checked. Species that would be expected, but are not actually documented, are listed as such and visual reports from REEF (without photographs) are separated. The “doubtful” category includes species that have appeared on prior lists but are likely misidentified, or, if correctly identified, were likely rare vagrants. The prior identifications of species of *Bathygobius* by Burgess *et al.* (1994) predated the review of the species by Tornabene *et al.* (2010); after the review, three species are confirmed, but the two remaining widespread Caribbean species cannot be excluded (Luke Tornabene, pers. comm.).

Acknowledgments

The assistance with permits by Phillippe Bush and permission granted by the Marine Conservation Board of the Cayman Islands Department of Environment is greatly appreciated. Mahmood Shivji, Director of the Guy Harvey Research Institute, facilitated the project and Christy Pattengill-Semmens and Dottie Benjamin were instrumental with arrangements. Thanks are due to Bill Smith-Vaniz and Rob Robins of the Division of Ichthyology at the Florida Museum of Natural History and H.J. Walker and Philip Hastings of the Scripps Institution of Oceanography for curatorial help. I am exceptionally grateful for contributions of underwater photographs by Cindy Abgarian, Barry Brown, Jim Burke, Jose Cohuo, Rick Coleman, Dustin Dorton, Diego Forero, Jim Garin, Jamie Holdorf, Frank Krasovec, Mark Lessard, Jan Morton, Mauro Ristorto, Mark Rosenstein, Wolfram Sander, Everett Turner,



Figure 35. *Ptereleotris helenae*, Grand Cayman (Cindy Abgarian).



Figure 36. *Oxyurichthys stigmalophius*, Grand Cayman (Cindy Abgarian).

Lourdes Vasquez-Yeomans, and Keri Wilk. Comparison specimens, tissues, and/or sequences were contributed by Alfred W. Thomson, Curator of Fishes at the Florida Fish and Wildlife Conservation Commission, Fish and Wildlife Research Institute; Carole Baldwin of the Smithsonian; Lourdes Vásquez-Yeomans, Martha Valdez-Moreno, and Jose Angel Cohuo Colli of ECOSUR, Quintana Roo, Mexico; Dustin Dorton; Kenneth Clifton of Lewis & Clark College; Arie de Jong of De Jong Marinelife B.V.; and D. Steinke of the Biodiversity Institute of Ontario, University of Guelph. George Walsh and Walsh Paper Distribution, Inc. of Westminster, CA sponsored preparation and publication of the project. The DNA barcoding was performed at the Biodiversity Institute of Ontario with the assistance of Bob Hanner and the team at BOLD. DNA barcoding was supported by the International Barcode of Life Project (iBOL.org) with funding from the Government of Canada via the Canadian Centre for DNA Barcoding, as well as from the Ontario Genomics Institute (2008-OGI-ICI-03), Genome Canada, the Ontario Ministry of Economic Development and Innovation, and the Natural Sciences and Engineering Research Council of Canada. The manuscript was reviewed by Pat Colin, Helen Randall, and John Randall, whose comments are much appreciated.

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Appendix 1. Specimen data and GenBank accession numbers for the mtDNA COI barcode sequences used to generate the phenogram in Fig. 6. Holotype in bold type.

Genus	species	Collection site	Voucher	GenBank #	BOLD BIN	Collector/Source
<i>Elacatinus</i>	<i>cayman</i>	Grand Cayman, Cayman Islands	c14ec227	KM987246	ACN1576	B. Victor
<i>Elacatinus</i>	<i>cayman</i>	Grand Cayman, Cayman Islands	c14ec233	KM987238	ACN1576	B. Victor
<i>Elacatinus</i>	<i>cayman</i>	Grand Cayman, Cayman Islands	c14ec204	KM987248	ACN1576	B. Victor
<i>Elacatinus</i>	<i>cayman</i>	Grand Cayman, Cayman Islands	c14ec141	KM987247	ACN1576	B. Victor
<i>Elacatinus</i>	<i>cayman</i>	Grand Cayman, Cayman Islands	c14ec181	KM987267	ACN1576	B. Victor
<i>Elacatinus</i>	<i>cayman</i>	Grand Cayman, Cayman Islands	c14ec244	KM987241	ACN1576	B. Victor
<i>Elacatinus</i>	<i>cayman</i>	Grand Cayman, Cayman Islands	c14ec160	KM987258	ACN1576	B. Victor
<i>Elacatinus</i>	<i>cayman</i>	Grand Cayman, Cayman Islands	c14ec159	KM987262	ACN1576	B. Victor
<i>Elacatinus</i>	<i>cayman</i>	Grand Cayman, Cayman Islands	c14ec208	KM987265	ACN1576	B. Victor
<i>Elacatinus</i>	<i>cayman</i>	Grand Cayman, Cayman Islands	c14ec193	KM987243	ACN1576	B. Victor
<i>Elacatinus</i>	<i>cayman</i>	Grand Cayman, Cayman Islands	c14ec210	KM987239	ACN1576	B. Victor
<i>Elacatinus</i>	<i>evelynae</i> YB	St. Thomas, US Virgin Islands	st307eee152	KM987251	AAC5098	B. Victor
<i>Elacatinus</i>	<i>evelynae</i> YB	La Parguera, Puerto Rico, USA	pr784bee120	HQ987838	AAC5098	B. Victor
<i>Elacatinus</i>	<i>evelynae</i> YB	St. Thomas, US Virgin Islands	st80628ee97	HQ987832	AAC5098	B. Victor
<i>Elacatinus</i>	<i>evelynae</i> YB	Curaçao, Netherlands Antilles	CURA8136	JQ842075	AAC5098	C. Baldwin, USNM
<i>Elacatinus</i>	<i>evelynae</i> YB	Curaçao, Netherlands Antilles	CURA8276	JQ842079	AAC5098	C. Baldwin, USNM
<i>Elacatinus</i>	<i>evelynae</i> YB	St. Thomas, US Virgin Islands	st9430ee88	KM987235	AAC5098	B. Victor
<i>Elacatinus</i>	<i>evelynae</i> YB	St. Thomas, US Virgin Islands	st506ee88	HQ987833	AAC5098	B. Victor
<i>Elacatinus</i>	<i>evelynae</i> YB	St. Thomas, US Virgin Islands	st506ee89	HQ987834	AAC5098	B. Victor
<i>Elacatinus</i>	<i>evelynae</i> YB	Soufriere, Dominica	d11719ee87	KM987256	AAC5098	B. Victor
<i>Elacatinus</i>	<i>evelynae</i> YB	St. Thomas, US Virgin Islands	st506ee90	HQ987835	AAC5098	B. Victor

<i>Elacatinus evelynae</i>	YB	La Parguera, Puerto Rico, USA	pr7811ee80	KM987266	AAC5098	B. Victor
<i>Elacatinus evelynae</i>	Y	Little Bahama Bank	bah9ee179	KM987263	ACG7318	B. Victor
<i>Elacatinus evelynae</i>	Y	Little Bahama Bank	bah9ee109	KM987259	ACG6787	B. Victor
<i>Elacatinus evelynae</i>	Y	Little Bahama Bank	bah9ee118	KM987254	ACG6787	B. Victor
<i>Elacatinus evelynae</i>	Y	Little Bahama Bank	bah509ee93	KM987252	ACG6787	B. Victor
<i>Elacatinus lobeli</i>		Belize	BZLW8076	JQ841525	ACF0819	C. Baldwin, USNM
<i>Elacatinus lobeli</i>		Utila, Bay Islands, Honduras	u8630e185	KM987250	ACF0819	B. Victor
<i>Elacatinus lobeli</i>		Quintana Roo, Mexico	MFLV2420	HM389880	ACF0819	L. Vásquez-Yeomans
<i>Elacatinus oceanops</i>		Florida, USA, aquarium trade	HLC-10781	FJ583384	AAB7794	D. Steinke
<i>Elacatinus oceanops</i>		Florida Keys, FL, USA	f9e0177	KM987242	AAB7794	B. Victor
<i>Elacatinus oceanops</i>		Florida, USA, aquarium trade	HLC-15115	FJ583388	AAB7794	D. Steinke
<i>Elacatinus genie</i>		Cuba, aquarium trade	aq10ec320.1	JN313576	AAB7792	A. de Jong
<i>Elacatinus genie</i>		Cuba, aquarium trade	HLC-13154	FJ583390	AAB7792	D. Steinke
<i>Elacatinus genie</i>		Cuba, aquarium trade	HLC-13155	FJ583391	AAB7792	D. Steinke
<i>Elacatinus genie</i>		Cuba, aquarium trade	HLC-13153	FJ583389	AAB7792	D. Steinke
<i>Elacatinus genie</i>		Cuba, aquarium trade	aq10ec310	KM987244	AAB7792	A. de Jong
<i>Elacatinus illecebrosus</i>		San Blas Islands, Panama	sb09ei145	JN313574	AAC1070	K.Clifton
<i>Elacatinus illecebrosus</i>		San Blas Islands, Panama	sb09ei202.1	JN313572	AAC1070	K.Clifton
<i>Elacatinus illecebrosus</i>		San Blas Islands, Panama	n762bgh58	KM987261	AAC1070	B. Victor
<i>Elacatinus illecebrosus</i>		San Blas Islands, Panama	sb09ei190	HQ987827	AAC1070	K.Clifton
<i>Elacatinus illecebrosus</i>		San Blas Islands, Panama	sb09ei141	HQ987828	AAC1070	K.Clifton
<i>Elacatinus illecebrosus</i>		San Blas Islands, Panama	sb09ei156.1	JN313573	AAC1070	K.Clifton
<i>Elacatinus illecebrosus</i>		San Blas Islands, Panama	n7530bei86	JN313607	AAC1070	B. Victor
<i>Elacatinus illecebrosus</i>		San Blas Islands, Panama	sb09ei275.1	JN313571	AAC1070	K.Clifton
<i>Elacatinus illecebrosus</i>		San Blas Islands, Panama	sb09ei89	JN313575	AAC1070	K.Clifton
<i>Elacatinus prochilos</i>		St. Thomas, US Virgin Islands	st952ep95	HQ987830	AAE8190	B. Victor
<i>Elacatinus prochilos</i>		Tobago	TOB9340	JQ842832	AAE8190	C. Baldwin, USNM
<i>Elacatinus prochilos</i>		St. Thomas, US Virgin Islands	st952ep195	KM987264	AAE8190	B. Victor
<i>Elacatinus prochilos</i>		Quintana Roo, Mexico	MFLV2419	HM389879	AAE8190	L. Vásquez-Yeomans
<i>Elacatinus prochilos</i>		Carrie Bow Cay, Belize	bz86ep155	KM987249	AAE8190	B. Victor
<i>Elacatinus prochilos</i>		St. Thomas, US Virgin Islands	st952ep1301	KM987255	AAE8190	B. Victor
<i>Elacatinus prochilos</i>		St. Thomas, US Virgin Islands	st951ep150	KM987257	AAE8190	B. Victor
<i>Elacatinus prochilos</i>		Quintana Roo, Mexico	MFLV2390	HM389854	AAE8190	L. Vásquez-Yeomans
<i>Elacatinus prochilos</i>		St. Thomas, US Virgin Islands	st952ep171	KM987269	AAE8190	B. Victor
<i>Elacatinus randalli</i>		Curaçao, Netherlands Antilles	CURA8275	JQ842080	AAF6345	C. Baldwin, USNM
<i>Elacatinus randalli</i>		Soufriere, Dominica	d11712er225	KM987240	AAF6345	B. Victor
<i>Elacatinus randalli</i>		Soufriere, Dominica	d11712er185	KM987260	AAF6345	B. Victor
<i>Elacatinus figaro</i>		Brazil, aquarium trade	aq10ef250	KM987265	AAO2001	B. Victor
<i>Elacatinus figaro</i>		Brazil, aquarium trade	aq10ef280	KM987253	AAO2001	B. Victor
<i>Elacatinus figaro</i>		Brazil, aquarium trade	aq10ef270	KM987237	AAO2001	B. Victor
<i>Elacatinus phthirophagus</i>		Noronha, Brazil	fn01er165	KM987236	AAF6344	B. Victor
<i>Elacatinus phthirophagus</i>		Noronha, Brazil	fn01er240	KM987268	AAF6344	B. Victor
<i>Elacatinus louisae</i>		Grand Cayman, Cayman Islands	c141280	KM894160	ACN1545	B. Victor

Appendix 2 (right). Specimen data and GenBank accession numbers for the mtDNA COI barcode sequences used to generate the phenogram in Fig. 20. Holotype in bold type.

Genus	species	Collection site	Voucher	GenBank #	BOLD BIN	Collector/Source
<i>Elacatinus</i>	<i>centralis</i>	Grand Cayman, Cayman Islands	c14eh325	KM894149	ACN0273	B. Victor
<i>Elacatinus</i>	<i>centralis</i>	Grand Cayman, Cayman Islands	c14eh260	KM894157	ACN0273	B. Victor
<i>Elacatinus</i>	<i>centralis</i>	Grand Cayman, Cayman Islands	c14eh160	KM894166	ACN0273	B. Victor
<i>Elacatinus</i>	<i>centralis</i>	Grand Cayman, Cayman Islands	c14eh235	KM894164	ACN0273	B. Victor
<i>Elacatinus</i>	<i>centralis</i>	Little Cayman, Cayman Islands	c14eh265	KM894172	ACN0273	B. Victor
<i>Elacatinus</i>	<i>centralis</i>	Grand Cayman, Cayman Islands	c14eh372	KM894156	ACN0273	B. Victor
<i>Elacatinus</i>	<i>centralis</i>	Grand Cayman, Cayman Islands	c14eh243	KM894150	ACN0273	B. Victor
<i>Elacatinus</i>	<i>centralis</i>	Grand Cayman, Cayman Islands	c14eh240	KM894169	ACN0273	B. Victor
<i>Elacatinus</i>	<i>chancei</i>	La Parguera, Puerto Rico, USA	pr785aec180	KM894151	AAE0159	B. Victor
<i>Elacatinus</i>	<i>chancei</i>	Soufriere, Dominica	d11718ec182	KM894162	AAE0159	B. Victor
<i>Elacatinus</i>	<i>chancei</i>	Soufriere, Dominica	d11718ec320	KM894170	AAE0159	B. Victor
<i>Elacatinus</i>	<i>chancei</i>	Soufriere, Dominica	d11718ec330	KM894153	AAE0159	B. Victor
<i>Elacatinus</i>	<i>horsti</i>	Curaçao, Netherlands Antilles	CURA8447	JQ842069	AAE0159	C. Baldwin, USNM
<i>Elacatinus</i>	<i>horsti</i>	Curaçao, Netherlands Antilles	CURA8092	JQ842072	AAE0159	C. Baldwin, USNM
<i>Elacatinus</i>	<i>horsti</i>	Curaçao, Netherlands Antilles	CURA8094	JQ842074	AAE0159	C. Baldwin, USNM
<i>Elacatinus</i>	<i>chancei</i>	Soufriere, Dominica	d11721ec144	KM894158	AAE0159	B. Victor
<i>Elacatinus</i>	<i>horsti</i>	Curaçao, Netherlands Antilles	CURA8093	JQ842073	AAE0159	C. Baldwin, USNM
<i>Elacatinus</i>	<i>horsti</i>	Curaçao, Netherlands Antilles	CURA8434	JQ842070	AAE0159	C. Baldwin, USNM
<i>Elacatinus</i>	<i>horsti</i>	Netherlands Antilles, aquarium trade	ddaqeh180	KM894173	AAE0159	D. Dorton
<i>Elacatinus</i>	aff. <i>horsti</i>	Little Bahama Bank	bah9eh99	KM894159	AAI6221	B. Victor
<i>Elacatinus</i>	<i>lori</i>	Belize	BZLWD7802	JQ841120	AAD0747	C. Baldwin, USNM
<i>Elacatinus</i>	<i>lori</i>	Belize	BZLW5399	JQ840477	AAD0747	C. Baldwin, USNM
<i>Elacatinus</i>	<i>lori</i>	Belize	BZLW5086	JQ840474	AAD0747	C. Baldwin, USNM
<i>Elacatinus</i>	<i>lori</i>	Belize	BZLW4090	JQ840032	AAD0747	C. Baldwin, USNM
<i>Elacatinus</i>	<i>lori</i>	Belize	BZLWD7723	JQ841126	AAD0747	C. Baldwin, USNM
<i>Elacatinus</i>	<i>lori</i>	Belize	BZLW5401	JQ840479	AAD0747	C. Baldwin, USNM
<i>Elacatinus</i>	<i>lori</i>	Belize	BZLW5334	JQ840483	AAD0747	C. Baldwin, USNM
<i>Elacatinus</i>	<i>lori</i>	Belize	BZLW5400	JQ840478	AAD0747	C. Baldwin, USNM
<i>Elacatinus</i>	<i>lori</i>	Utila, Bay Islands, Honduras	u872e117	KM894154	ACF2986	B. Victor
<i>Elacatinus</i>	<i>lori</i>	Utila, Bay Islands, Honduras	u872e82	KM894165	ACF2986	B. Victor
<i>Elacatinus</i>	<i>lori</i>	Utila, Bay Islands, Honduras	u871e87	KM894168	ACF2986	B. Victor
<i>Elacatinus</i>	<i>lori</i>	Utila, Bay Islands, Honduras	u872e86	KM894155	ACF2986	B. Victor
<i>Elacatinus</i>	<i>louisae</i>	Grand Cayman, Cayman Islands	c14el279	KM894152	ACN1545	B. Victor
<i>Elacatinus</i>	<i>louisae</i>	Grand Cayman, Cayman Islands	c14el175	KM894161	ACN1545	B. Victor
<i>Elacatinus</i>	<i>louisae</i>	Grand Cayman, Cayman Islands	c14el280	KM894160	ACN1545	B. Victor
<i>Elacatinus</i>	<i>colini</i>	Belize	BZLW4019	JQ840028	AAC1069	C. Baldwin, USNM
<i>Elacatinus</i>	<i>colini</i>	Belize	BZLW4043	JQ840030	AAC1069	C. Baldwin, USNM
<i>Elacatinus</i>	<i>colini</i>	Belize	BZLW4018	JQ840034	AAC1069	C. Baldwin, USNM
<i>Elacatinus</i>	<i>colini</i>	Belize	BZLW6172	JQ840816	AAC1069	C. Baldwin, USNM
<i>Elacatinus</i>	<i>colini</i>	Belize	BZLW6170	JQ840817	AAC1069	C. Baldwin, USNM
<i>Elacatinus</i>	<i>colini</i>	Belize	BZLW6266	JQ840818	AAC1069	C. Baldwin, USNM
<i>Elacatinus</i>	<i>colini</i>	Belize	BZLW6171	JQ840815	AAC1069	C. Baldwin, USNM
<i>Elacatinus</i>	<i>colini</i>	Belize	BZLW4020	JQ840031	AAC1069	C. Baldwin, USNM
<i>Elacatinus</i>	<i>colini</i>	Utila, Bay Islands, Honduras	u8630el199	KM894167	AAC1069	B. Victor
<i>Elacatinus</i>	<i>colini</i>	Utila, Bay Islands, Honduras	u871e106	KM894174	AAC1069	B. Victor
<i>Elacatinus</i>	<i>xanthiprora</i>	Florida Middle Grounds, USA	fwri1341	KM894163	ACL3709	A. Thomson, FWRI
<i>Elacatinus</i>	<i>xanthiprora</i>	Florida Middle Grounds, USA	fwri1342	KM894171	ACL3709	A. Thomson, FWRI
<i>Elacatinus</i>	<i>oceanops</i>	Florida Keys, FL, USA	f9eo177	KJ591651	AAB7794	B. Victor

Appendix 3. Specimen data and GenBank accession numbers for the mtDNA COI barcode sequences used to generate the phenogram in Fig. 33. Holotype in bold type.

Genus	species	Collection site	Voucher	GenBank #	BOLD BIN	Collector/Source
<i>Tigrigobius</i>	<i>harveyi</i>	Grand Cayman, Cayman Islands	c14th196	KM894139	ACM9875	B. Victor
<i>Tigrigobius</i>	<i>harveyi</i>	Grand Cayman, Cayman Islands	c14th163	KM894140	ACM9875	B. Victor
<i>Tigrigobius</i>	<i>harveyi</i>	Grand Cayman, Cayman Islands	c14th200	KM894141	ACM9875	B. Victor
<i>Tigrigobius</i>	<i>harveyi</i>	Grand Cayman, Cayman Islands	c14th174	KM894142	ACM9875	B. Victor
<i>Tigrigobius</i>	<i>harveyi</i>	Grand Cayman, Cayman Islands	c14th188	KM894146	ACM9875	B. Victor
<i>Tigrigobius</i>	<i>harveyi</i>	Grand Cayman, Cayman Islands	c14th191	KM894145	ACM9875	B. Victor
<i>Tigrigobius</i>	<i>panamensis</i>	Portobelo, Panama	n7529bem88	GU908153	AAB2884	B. Victor
<i>Tigrigobius</i>	<i>panamensis</i>	Portobelo, Panama	n7527aem142	GU908147	AAB2884	B. Victor
<i>Tigrigobius</i>	<i>panamensis</i>	Portobelo, Panama	n7527aem120	GU908155	AAB2884	B. Victor
<i>Tigrigobius</i>	<i>panamensis</i>	San Blas Islands, Panama	sb81046em230	GU908156	AAB2884	B. Victor
<i>Tigrigobius</i>	<i>panamensis</i>	San Blas Islands, Panama	sb83116em195	GU908157	AAB2884	B. Victor
<i>Tigrigobius</i>	<i>panamensis</i>	Portobelo, Panama	n7527aem205	GU908144	AAB2884	B. Victor
<i>Tigrigobius</i>	<i>panamensis</i>	Portobelo, Panama	n7527aem140	GU908145	AAB2884	B. Victor
<i>Tigrigobius</i>	<i>rubrigenis</i>	Utila, Bay Islands, Honduras	u873em166	GU908149	AAB2885	B. Victor
<i>Tigrigobius</i>	<i>rubrigenis</i>	Utila, Bay Islands, Honduras	u873em169	GU908150	AAB2885	B. Victor
<i>Tigrigobius</i>	<i>rubrigenis</i>	Utila, Bay Islands, Honduras	u873em174	GU908148	AAB2885	B. Victor
<i>Tigrigobius</i>	<i>rubrigenis</i>	Utila, Bay Islands, Honduras	u873em180	GU908146	AAB2885	B. Victor
<i>Tigrigobius</i>	<i>rubrigenis</i>	Utila, Bay Islands, Honduras	u873em193	GU908152	AAB2885	B. Victor
<i>Tigrigobius</i>	<i>rubrigenis</i>	Utila, Bay Islands, Honduras	u873em185	GU908154	AAB2885	B. Victor
<i>Tigrigobius</i>	<i>multifasciatus</i>	Berry Islands, Bahamas	BAHA8239	JQ839751	AAB2887	C. Baldwin, USNM
<i>Tigrigobius</i>	<i>multifasciatus</i>	Berry Islands, Bahamas	BAHA8238	JQ839748	AAB2887	C. Baldwin, USNM
<i>Tigrigobius</i>	<i>multifasciatus</i>	NW Puerto Rico, USA	pr786em161	GU908158	AAB2886	B. Victor
<i>Tigrigobius</i>	<i>multifasciatus</i>	NW Puerto Rico, USA	pr786em162	GU908151	AAB2886	B. Victor
<i>Tigrigobius</i>	<i>multifasciatus</i>	St. Thomas, US Virgin Islands	st9429em199	GU908134	AAB2888	B. Victor
<i>Tigrigobius</i>	<i>multifasciatus</i>	St. Thomas, US Virgin Islands	st9430em117	GU908139	AAB2888	B. Victor
<i>Tigrigobius</i>	<i>multifasciatus</i>	St. Thomas, US Virgin Islands	st953em192	GU908142	AAB2888	B. Victor
<i>Tigrigobius</i>	<i>multifasciatus</i>	St. Thomas, US Virgin Islands	st9430em159	GU908138	AAB2888	B. Victor
<i>Tigrigobius</i>	<i>multifasciatus</i>	St. Thomas, US Virgin Islands	st953em202	GU908143	AAB2888	B. Victor
<i>Tigrigobius</i>	<i>multifasciatus</i>	St. Thomas, US Virgin Islands	st9429em193	GU908136	AAB2888	B. Victor
<i>Tigrigobius</i>	<i>multifasciatus</i>	St. Thomas, US Virgin Islands	st9429em183	GU908137	AAB2888	B. Victor
<i>Tigrigobius</i>	<i>multifasciatus</i>	St. Thomas, US Virgin Islands	st953em204	GU908141	AAB2888	B. Victor
<i>Tigrigobius</i>	<i>multifasciatus</i>	St. Thomas, US Virgin Islands	st953em230	GU908140	AAB2888	B. Victor
<i>Tigrigobius</i>	<i>multifasciatus</i>	St. Thomas, US Virgin Islands	st9429em185	GU908135	AAB2888	B. Victor
<i>Tigrigobius</i>	<i>pallens</i>	Grand Cayman, Cayman Islands	c14tp98	KM894143	ACN0211	B. Victor