

***Halichoeres sanchezi*, a new wrasse from the Revillagigedo Archipelago of Mexico, tropical eastern Pacific Ocean (Teleostei: Labridae) (#92034)**

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I commend the authors for their extensive data set, compiled over many years of detailed fieldwork. In addition, the manuscript is clearly written in professional, unambiguous language. If there is a weakness, it is in the statistical analysis (as I have noted above) which should be improved upon before Acceptance.

***Halichoeres sanchezi*, a new wrasse from the Revillagigedo Archipelago of Mexico, tropical eastern Pacific Ocean (Teleostei: Labridae)**

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A new labrid fish species, *Halichoeres sanchezi* n. sp., is described from specimens collected in the Revillagigedo Archipelago in the tropical eastern Pacific Ocean, off the coast of Mexico. The new species belongs to the *Halichoeres melanotis* species complex that is found throughout the region, and it diverges 2.4% in the mtDNA cytochrome c oxidase I sequence from its nearest relative, *H. melanotis* from Panama, and 2.9% from *Halichoeres salmofasciatus* from Cocos Island, off Costa Rica. The juveniles and initial phase of the new species closely resemble those of *H. salmofasciatus* and *Halichoeres malpelo* from Malpelo Island off Colombia, differing in having a rounded black spot with a yellow dorsal margin on the mid-dorsal fin of initial phase (IP) adults as well as on juveniles. In contrast, the terminal phase male (TP) color pattern is distinct from other relatives, being vermilion with dark scale outlines, a white abdomen becoming yellowish posteriorly, and a prominent black patch on the posterior caudal peduncle and base of the caudal fin. The new species adds to the list of endemic fish species for the isolated archipelago and is an interesting case of island endemism in the region. The discovery was made during the joint 2022 collecting expedition to the archipelago, which featured a pioneering approach where an ichthyofauna is inventoried in a collaboration among researchers and expert photographers, with specimens photographed underwater, hand-collected, photographed fresh, tissue-sampled, and subsequently vouchered in museum collections.

1 ***Halichoeres sanchezi*, a new wrasse from the Revillagigedo**
 2 **Archipelago of Mexico, tropical eastern Pacific Ocean**
 3 **(Teleostei: Labridae)**

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16 **Abstract**

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 18 the Revillagigedo Archipelago in the tropical eastern Pacific Ocean, off the coast of Mexico.

19 The new species belongs to the *Halichoeres melanotis* species complex that is found throughout
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25 (IP) adults as well as on juveniles. In contrast, the terminal phase male (TP) color pattern is

distinct from other relatives, being vermilion with dark scale outlines, a white abdomen becoming yellowish posteriorly, and a prominent black patch on the posterior caudal peduncle and base of the caudal fin. The new species adds to the list of endemic fish species for the isolated archipelago and is an interesting case of island endemism in the region. The discovery was made during the joint 2022 collecting expedition to the archipelago, which featured a pioneering approach where an ichthyofauna is inventoried in a collaboration among researchers and expert photographers, with specimens photographed underwater, hand-collected, photographed fresh, tissue-sampled, and subsequently vouchered in museum collections.

Introduction

The geography of the tropical eastern Pacific Ocean (TEP) provides a useful proving ground for answering questions about marine island biogeography. There is a long and relatively linear coastline with a narrow continental shelf and a series of isolated offshore islands. Unlike most other tropical marine regions, each island group contains a different set of the regional shorefish fauna (Allen & Robertson 1994) and the patterns of occurrence, especially the prevalence of endemism and degrees of connectivity, can be directly assessed and hypotheses can be tested. The Revillagigedo Archipelago of Mexico, forming the Reserva de la Biósfera Archipiélago de Revillagigedo, includes the islands of Clarión, Socorro, San Benedicto, and Roca Partida, and is about 400 km south of the Baja California peninsula and about 700 km west of Manzanillo. It is one of the more remote and least visited of the island groups of the TEP, with a short history of collecting expeditions and a very limited number of museum collections, especially compared to more well-surveyed TEP islands, such as the Galapagos Archipelago.

47 In November 2022, a joint expedition formed by an international team of ichthyologists and a
 48 cadre of experienced underwater photographers, visited the four islands of the archipelago and
 49 compiled a comprehensive inventory of the shallow shorefish fauna, with in situ photographic
 50 documentation of all of the fish species encountered. All species encountered were sampled for
 51 tissues and subsequently DNA-barcoded, i.e. sequenced for the mtDNA cytochrome c oxidase I
 52 gene. One of the more compelling aspects of the expedition was the contemporaneous
 53 underwater documentation, including on videotape, of the discovery and collection of the
 54 holotype of a new species of fish.

55 Among the species discovered by the expedition was a new labrid species of *Halichoeres*
 56 Rüppell, 1835, the local representative of the widespread TEP *Halichoeres melanotis* species
 57 complex, which is made up of *Halichoeres melanotis* (Gilbert, 1890) along the mainland,
 58 *Halichoeres salmofasciatus* Robertson & Allen, 2002, endemic to Cocos Island, and *Halichoeres*
 59 *malpelo* Allen & Robertson, 1992, endemic to Malpelo Island (Robertson & Allen 2015). The
 60 new species had been photographed at least once previously, but had not been identified with any
 61 certainty. It is a particularly elusive wrasse, with adults difficult to approach, accounting for the
 62 dearth of photographs despite many tourist divers photographing fishes in the archipelago. A
 63 series of specimens were collected on San Benedicto, the northernmost island of the
 64 Revillagigedo Archipelago (Fig. 1).

65 **Materials & Methods**

66 Specimens of the new species are presently accessioned in the Colección Nacional de Peces at
 67 the Universidad Nacional Autónoma de México (CNP-IBUNAM), the Natural History Museum
 68 of Los Angeles County (LACM), and the Marine Vertebrate Collection of Scripps Institution of

Oceanography (SIO). Permits for the travel, boats, collection, and export were issued to the Universidad Nacional Autónoma de México by the Secretariat of Agriculture, Livestock, Rural Development, Fisheries and Food (SAGARPA) of the Federal Government of Mexico (permit number PPF/DGOPA-A-099/22). Measurements and counts of congeners used for comparative purposes were from specimens housed at LACM, SIO, and the NMNH or values taken from the literature (Allan & Robertson 1992, Allen & Robertson 1994, Allen & Robertson 2002, Robertson & Allen 2015).

The length of specimens is given as standard length (SL), measured from the median anterior end of the upper lip to the base of the caudal fin (posterior end of the hypural plate); body depth is the greatest depth from the base of the dorsal-fin spines to the ventral edge of the abdomen (correcting for any malformation of preservation); body width is measured just posterior to the gill opening; head length from the front of the upper lip to the posterior end of the opercular flap; orbit diameter is the greatest fleshy diameter of the orbital rim, and interorbital width the least bony width between the orbits; snout length is measured from the median anterior point of the upper lip to the nearest fleshy rim of the orbit; caudal-peduncle depth is the least depth, and caudal-peduncle length the horizontal distance between verticals at the rear base of the anal fin and the caudal-fin base; predorsal, prepelvic and preanal lengths are oblique measurements taken from the median anterior point of the upper lip to the base of the first spine of each respective fin; lengths of spines and rays are measured to their extreme bases; caudal-fin and pectoral-fin lengths are the length of the longest ray; pelvic-fin length is measured from the base of the pelvic spine to the tip of the longest soft ray. The upper rudimentary pectoral-fin ray is included in the count. Lateral-line scale counts list the last pored scale that overlaps the end of the hypural plate as +1. The count of gill rakers is made on the first gill arch and includes all rudiments.

Proportional measurements were taken with digital calipers or from radiograph images using ImageJ (<https://imagej.net/>) and are rounded to the nearest 0.1. Counts were taken from specimens, digital radiographs, and micro-computer tomography (microCT) scans. Counts and measurements for the paratypes are shown in parentheses following data for the holotype (some counts and measurements were not made on the juvenile paratype CNP-IBUNAM XXX).

Proportional morphological measurements as percentages of the standard length are presented in Table 1.

Fluorescent emission was documented using a Canon 5D Mark IV DSLR camera with a Canon EF 100mm f/2.8L Macro IS USM lens. Photographs were taken using two NightSea Xite Fluorescent Flashlights that emitted royal blue light (ca 440–460nm). Each light was held at approximately 45° to the subject and was equipped with a 15° diffuser cap. A 500nm Tiffen #12 Yellow Filter was placed in front of the lens for all photographs. Micro-computed tomography (μCT) scans were used to visualize internal skeletal anatomy. Scans were performed at the NHMLA on a Bruker Skyscan 1273. Scans of the holotype and one of the paratypes were conducted with and without a 1mm aluminum filter, with 70–90kV, 166–214μA, 471–636ms exposure times, with a 10–34μm voxel size. All scans were reconstructed using the software NRecon v2.2.0.6 (Bruker) and visualized using CTVox (Bruker). CT scans were uploaded to Morphosource.org for the holotype

<https://www.morphosource.org/concern/media/000570594?locale=en>

and the largest paratype

<https://www.morphosource.org/concern/media/000570600?locale=en>

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 µl volume including 6.25 µl of 10% trehalose, 2 µl of ultrapure water, 1.25 µl of 10× PCR buffer (10mM KCl, 10mM (NH₄)₂SO₄, 20mM Tris- HCl (pH8.8), 2mM MgSO₄, 0.1% Triton X-100), 0.625 µl of MgCl₂ (50mM), 0.125 µl of each primer (0.01mM), 0.0625 µl of each dNTP (10mM), 0.0625 µl of *Taq* DNA polymerase (New England Biolabs), and 2 µl 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 the Barcode of Life Data Systems (Ratnasingham & Hebert 2007). The sequence data are publicly accessible on BOLD (Process ID LIDMA3845-22 at <https://www.boldsystems.org>) and GenBank (accession number OQ922018). Sequence divergences were 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' sequence divergence. Genetic distances were calculated by the BOLD algorithm, both as uncorrected p-distances and as K2P distances.

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***Halichoeres sanchezi*, n. sp.** urn:lsid:zoobank.org:act:608591F4-2A1B-4271-9BB2-0F0B2BD2DC4E

Tailspot Wrasse, Doncella Colimanchada (Spanish)

mtDNA COI sequence BIN <https://doi.org/10.5883/BOLD:AFA9780>

Figures 2–10, Table 1.

Holotype. LACM 61379, 123.1 mm SL, Mexico, Colima, Revillagigedo Archipelago, San Benedicto Island, canyon rubble plain site, sta. WBL 275, 19°17.740' N, 110°48.466' W (19.2957, -110.8078); 21–22 m depth; spear, W.B. Ludt, 30 November 2022.

Paratypes. (7 specimens 32.3–53.3 mm SL), CNP-IBUNAM XXX, 39.1 mm SL, sta. WBL 274, same locality as holotype, quinaldine and handnet, B.W. Frable, 30 November 2022; CNP-IBUNAM XXX, 2, 19.8–32.3 mm SL, sta. WBL 274, same locality as holotype, quinaldine and handnet, C.A. Sanchez et al., 30 November 2022; LACM 61380, 53.3 mm SL, sta. WBL 274, same locality as holotype; quinaldine and handnet, B.W. Frable, 30 November 2022; SIO 23-48, 40.4 mm SL, sta. WBL 279, 19°17.795'N, 110°48.351'W, quinaldine and handnet, C.A. Sanchez, 30 November 2022; SIO 23-50, 46.2 mm SL, sta. WBL 274, same locality as holotype,

159 quinaldine and handnet, C.A. Sanchez et al., 30 November 2022; SIO 23-51, 36.5 mm SL, sta.
160 WBL 275, same locality as holotype; quinaldine and handnet, B.W. Frable, 30 November 202.

161 **Diagnosis.** Dorsal-fin rays IX,12; anal-fin rays III,12; pectoral-fin rays 13 (counting uppermost
162 rudimentary ray); lateral-line scales 26 (+1 on caudal-fin base); suborbital pores 6; gill rakers
163 14–16; a pair of large, projecting, and slightly recurved canine teeth anteriorly in each jaw, the
164 lowers curving forward and fitting between uppers when mouth closed, a second pair of canines
165 about half size of first, followed by rows of mostly caniniform teeth, no canine posteriorly at
166 corner of mouth; elongate body, body depth 3.6–4.2 in SL; body width 2.2–3.0 in depth; caudal
167 fin slightly rounded; TP vermilion to orangish-brown, yellow-cream ventrally with a dark cross-
168 hatch pattern outlining scales, opercular flap diffusely black and purple; a prominent large black
169 blotch covering rear caudal peduncle and proximal half of caudal fin, a pale patch over mid-
170 abdominal area underlying pectoral fin; a small black spot on first dorsal-fin membrane; IP and
171 juvenile fish grey grading to white ventrally with a broad midlateral red band from snout to
172 caudal peduncle, often breaking up into horizontal block segments, including a distinct black
173 crescent or oval spot on expanded soft flap of upper operculum, and ending in a horizontal
174 oblong black spot at caudal-fin base; a similar narrower band runs along upper body below base
175 of dorsal fin from snout to upper caudal peduncle; fins clear except for a large rounded black
176 spot, edged in yellow dorsally, centered over membranes of last dorsal-fin spine and first three
177 rays and extending partially onto upper body (present in both juvenile and IP adults).

178 **Description.** Dorsal-fin rays IX,12; anal-fin rays III,12, all soft dorsal and anal-fin segmented
179 rays branched, last split to base; pectoral-fin rays 13, first rudimentary, second unbranched;
180 pelvic-fin rays I,5; segmented caudal-fin rays 17, upper two and lower three unbranched, 5 upper

181 and lower procurent rays; pored lateral-line scales 26 (+1 on caudal-fin base); gill rakers 14
182 (14–16); branchiostegal rays 5; vertebrae 25.

183 Body elongate, depth 3.6 (3.8–4.2) in SL, and compressed, body width 8.9 (8.9–11.2) in SL and
184 2.5 (2.2–3.0) in depth; head length 3.3 (3.0–3.2) in SL; snout pointed and short, its length 3.5
185 (3.5–4.1) in HL; eye diameter 5.5 (4.1–4.9) in HL; interorbital space broadly convex, least bony
186 width 5.2 (4.8–8.1) in HL; caudal peduncle short and narrow, least depth 2.2 (2.3–2.5) in HL,
187 caudal-peduncle length 2.7 (3.0–5.0) in HL.

188 Mouth small, terminal, oblique at about 45 degrees, upper-jaw length 4.3 (4.6–5.3) in HL; two
189 pairs of enlarged canine teeth at front of upper jaw (two canines per side, anterior pair larger than
190 posterior pair) and two pairs of enlarged canine teeth at front of lower jaw (two canines per side,
191 anterior pair larger than posterior pair) fitting between upper pair when mouth is closed; teeth
192 behind enlarged canines in a regular row of 10–15 caniniform to conical teeth in each quadrant;
193 no posterior canine at corner of mouth. Upper preopercular margin free and smooth nearly to
194 level of lower edge of orbit; lower margin free anterior to a vertical through anterior nostril. Gill
195 rakers short, longest on first arch (at angle) about one-quarter length of longest gill filament.
196 Nostrils small, in front of anterior edge of orbit. Head pores comprise two over maxilla, then two
197 anterior to orbit, followed by a curving suborbital series of 6 pores in a single row up to rear mid-
198 level of orbit.

199 Scales thin and cycloid; scales on side of thorax less than half as high as largest scales on side of
200 body, becoming still smaller ventroanteriorly; head mostly naked except for small partially
201 embedded scales on nape in irregular rows but sparing midline in front of dorsal fin; fins naked
202 except for several progressively smaller scales on basal region of caudal fin and mid-ventral

scale projecting posteriorly from base of pelvic fins. Scale rows above lateral line 4 and below lateral line 8; circumpeduncular scales 16. Lateral line continuous, nearly following contour of back to about 19th pored scale, below base of about 8th dorsal-fin soft ray, where deflected ventrally to a straight peduncular portion; anteriormost scales in holotype with 3 pores per scale, after a few scales becoming two pores, and straight section with a single pore per scale; on all (smaller) paratypes only a single pore per scale, last pored scale on caudal-fin base.

Origin of dorsal fin above anterior edge of second lateral-line scale; predorsal length 3.2 (3.3–3.9) in SL; dorsal-fin base 1.5 (1.5–1.7) in SL; dorsal-fin spines progressively longer, first 5.9 (5–6) in HL; longest dorsal-fin spine 3.8 (2–3) in HL longest dorsal-fin soft ray 2.5 (2–2) in HL; origin of anal fin below base of last dorsal-fin spine; preanal length 2.1 (1.7–1.9) in SL; anal-fin base 2.6 (2.5–2.9) in SL; first anal-fin spine short, 10.6 (11–12) in HL; second anal-fin spine 6.3 (5–6) in HL; third anal-fin spine 4.5 (4–4) in HL; longest anal-fin soft ray 3.0 (2–3) in HL; caudal-fin length 1.9 (1.3–1.5) in HL; second or third pectoral- fin ray usually longest, 1.6 (1.7–2.0) in HL; pelvic-fin spine short, 3.5 (3.5–4.3) in HL, pelvic-fin length 2.3 (2.2–2.5) in HL.

Color in life. (Figs. 2–7) Fresh TP specimens have a vermilion head and orangish-brown body, grading to yellowish-cream ventrally. Head mostly uniformly colored, jaws paler, yellowish below level of mandible, with a prominently reddish orange iris and a reddish orange opercular flap with irregular indigo lines and and black and purple in diffuse patches. Body with a cross-hatched pattern of rows of scales with dark brown crescents with reddish borders on each scale, grading to yellowish ventrally where spots are less conspicuous; upper abdominal area underlying pectoral fin with opaque white underlying scales forming a conspicuous white patch. Rear body with a large black patch covering end of caudal peduncle and extending over proximal

225 half of caudal fin. Dorsal fin with a small black spot centered on first spinous membrane and
 226 remaining dorsal-fin membranes with irregular yellowish bands and a blue marginal band along
 227 soft-dorsal-fin margin; small bright indigo spots at bases of anterior dorsal-fin spines; caudal-fin
 228 rays brownish orange fading to yellow distally, membranes translucent brown distally; anal fin
 229 yellow with bluish bands midfin and along margin; pectoral fins translucent, orange at base, axil
 230 with a small dark spot; pelvic fins with yellow band anteriorly.

231 Underwater, where red color is absorbed, head and body appear dusky purple to greenish, with
 232 dark cross-hatching from outlined scales; more prominent visible markings are pale lips, a large
 233 white patch over upper abdomen beneath pectoral fin, a pale dorsal-fin base with an expanded
 234 pale area just before black area on caudal peduncle, and a conspicuous black caudal blotch
 235 covering end of caudal peduncle and basal half of caudal fin.

236 IP fish pinkish grey dorsally grading to white on abdomen becoming yellowish posteriorly with a
 237 broad brick-red band running along lateral midline from snout to caudal peduncle, ending in a
 238 horizontal oblong black spot just above midline on caudal-fin base, iris bright red-orange in line
 239 with band, a distinct black crescent with indigo to purplish anterior margin on opercular flap;
 240 upper body with a less-prominent, thinner, reddish band running from upper snout to dorsal
 241 caudal peduncle, bands can form horizontal block segments when fish are disturbed. Dorsal fin
 242 with a large, rounded, black spot on basal two-thirds of fin, extending partially onto body scales,
 243 centered on last spine and first three rays, with most of upper margin of spot edged with yellow
 244 and white, forming a partial ocellus; a small dark spot in the pectoral-fin axil; fin membranes
 245 translucent with brownish or faint purple-red rays. Juveniles (less than 20 mm SL) with similar

color pattern but black spots on mid-dorsal fin and caudal-fin base relatively larger and a more rounded opercular-flap black spot.

Color under fluorescence. (Fig. 8) Entire body of TP male without fluorescent emission except for slight yellow emission on pale patch beneath pectoral fin; dorsal, anal and pelvic-fin spines and rays faint yellow. Entire body of IP fish bright red; eye dark without fluorescence except for upper half to two-thirds of iris red; small, round opercular black spot; round to oblong black spots at midpoint of dorsal fin and slightly above lateral line at base of caudal fin. **Color in alcohol.** (Fig. 9) Head and body of TP male brownish with residual dark-centered scale rows and black markings on first dorsal-fin membrane and rear caudal blotch persisting in preservative; IP and juvenile individuals pale brownish with black marks on opercular flap, rounded spot on mid-dorsal fin, and at end of mid-lateral body band on caudal-fin base.

Etymology. Named for Prof. Carlos Armando Sánchez Ortiz, of the Programa de Investigación para la Conservación de la Fauna Arrecifal (PFA), Biología Marina, Universidad Autónoma de Baja California Sur (UABCS) in La Paz, Baja California Sur, Mexico, in recognition of his contributions to the study of the marine communities of Pacific Mexico and who organized the 2022 expedition and collected the first specimen of the new species.

Distribution. The new species is presently known only from the Revillagigedo Archipelago; it has been documented on Socorro Island by the underwater photographs of Kreg Martin in 2013 (Fig. 4) and from San Benedicto Island by our 2022 expedition. No other records exist in the compendium of underwater photographs we have reviewed or in museum collections we have examined.

Habitat. Specimens were only observed and collected at a specific site off the southern end of San Benedicto, just west of the popular El Canyon dive site at Caletilla Volteadura (Fig. 1). The site was a large, even plain, approximately 21–22 m deep, composed of volcanic gravel-rubble surrounded by lava boulders and with a few boulders dispersed in the plain amongst a dark bottom. Patches of white gravel were interspersed among the dark volcanic gravel-rubble areas. Juveniles and smaller IP fish were found generally just off the bottom around boulders and over lighter patches of gravel while larger IP and TP individuals were observed slightly higher in the water column around the boulders on the eastern edge of the plain.

DNA analysis. (Fig. 10) The neighbor-joining phenetic tree based on the COI mtDNA sequences of a set of TEP labrid species allied to *Halichoeres*, following the Kimura two-parameter model (K2P) generated by BOLD (Barcode of Life Database), shows relatively low divergences between species within complexes and deep divergences between more distant sets of species. The COI sequence of *Halichoeres sanchezi* was recovered in a clade containing the two other members of the complex, i.e. *H. salmofasciatus* and *H. melanotis*; note that there are no sequences available for *H. malpelo*. The members of the complex are less than 3% divergent from each other: 2.4% between *H. sanchezi* and *H. melanotis*, 2.9% between *H. sanchezi* and *H. salmofasciatus*, and 2.1% between *H. melanotis* and *H. salmofasciatus* (all pairwise distances). All other *Halichoeres* species are more than 13.5% different from the *H. melanotis* species complex.

Interestingly, another endemic wrasse, *Halichoeres insularis* Allen & Robertson, 1992, was discovered on a prior expedition to the Revillagigedo Archipelago in 1991. That species diverges by 7.6% in mtDNA COI sequence from its widespread mainland sister species, *Halichoeres*

289 *dispilus* Günther, 1864. The wide range in divergences of the putative endemic species in the
 290 Revillagigedo Archipelago, from 0.5% for *Thalassoma virens* Gilbert, 1890 (from *Thalassoma*
 291 *purpureum* Forsskål, 1775) (Victor et al. 2013) up to 7.6% for *H. insularis* makes it difficult to
 292 discern a pattern in phylogenetic age for the splitting of species in the region. Note the
 293 cytochrome-b sequence (GU938863) for “*H. insularis*” used in Wainwright et al. (2019) is
 294 apparently a mislabeled sequence of *H. salmofasciatus*, leading to a mistaken assignment of *H.*
 295 *insularis* to the *H. melanotis* clade in the Bayesian timetree in their Fig. 2 and Rocha et al.
 296 (2010): there were no tissues from Revillagigedo available to them at the time. An additional
 297 correction regarding the phenetic tree is that *Halichoeres burekae* Weaver & Rocha, 2007 as
 298 listed by Victor et al. (2013) (GenBank accession JN313704), is now identified as *Halichoeres*
 299 *caudalis* (Poey, 1860) and newly obtained sequences for *H. burekae* are used in the tree here.

300 **Comparisons.** At the time of prior descriptions, there were few or no underwater photographs
 301 of various phases of the *H. melanotis* species complex to compare with, thus the table of color-
 302 pattern differences in Allen & Robertson (2002, Table 2) is no longer accurate. Notably, Bessudo
 303 & Lefevre (2017), in their guide to the fishes of Malpelo, follow that table describing IP *H.*
 304 *malpelo* as having neither the opercular-flap or caudal-peduncle spots and thus label their
 305 photographs of IP fish with the caudal spot as *H. salmofasciatus*, and those without as *H.*
 306 *malpelo* (and listing both species for Malpelo Island). Recently, a set of additional underwater
 307 photographs have been taken of *H. salmofasciatus* on Cocos Island (Figs. 11 & 12), *H. malpelo*
 308 on Malpelo Island (Figs 13 & 14), and *H. melanotis* from Panama and Baja California (Figs. 15–
 309 18). These photographs greatly expand the known color variations for these species. *Halichoeres*
 310 *sanchezi* is most similar in appearance to its southern island relatives. The IP adults of both *H.*
 311 *salmofasciatus* and *H. malpelo* closely resemble *H. sanchezi*, except they lack the mid-dorsal-fin

spot that is diagnostic of IP adult *H. sanchezi*. Allen & Robertson (2002) reported that the spot is only present on juvenile *H. salmofasciatus* below about 20 mm SL and a diagnostic photograph from Cocos Island shows that loss of the spot as individuals get larger (Fig. 11 top) (juveniles of *H. malpelo* have not been documented). The underwater photographs of the three *H. melanotis* complex species show wide variation in the color patterns of IP fishes with varying intensities of red from candy-stripe red to brown and the opercular-flap spot and the caudal-peduncle spot sometimes faded or absent (Figs. 11, 13 & 17).

The juveniles and IP of mainland *H. melanotis* are especially variable in appearance, with multiple different versions of juvenile color patterns (Fig. 15). One is the classic golden-yellow, black-striped juvenile (leading to the common name Golden Wrasse) as illustrated in Allen & Robertson (1992; 1994): it has been photographed frequently both in Panama and Baja California. The golden juveniles can have a mid-dorsal-fin spot on very small individuals, but it is quickly lost. In contrast, recent photographs by Allison Morgan Estape and Carlos Estape document quite different versions of juveniles and small IP fish in Panama and Baja California, coexisting with golden juveniles. Some are mostly black and white versions of the golden form, but others show dark bars with a variety of colors and a prominent colorful opercular flap with black and yellow and blue, similar to the flap of TP males; some of these small individuals can have a large round mid-dorsal spot, edged on the upper portion with yellow, as in juveniles of *H. sanchezi*. Another juvenile *H. melanotis* from the Sea of Cortez shows a different color pattern but retains the opercular flap markings and has a large caudal-fin base spot but no dorsal-fin spot (Fig. 16). Both juvenile and IP phases can sometimes have a black spot on the membrane of the first dorsal-fin spine, a feature of TP males of the *H. melanotis* complex. The significance of this extreme variation in color patterns in this species remains to be clarified

Larger IP adult *H. melanotis* also have a wide variety of color patterns, from pale salmon with the same black spots on the opercular flap and caudal-fin base of the other members of the complex to shades of grey, yellow, green, or brown with the black opercular-flap spot the only prominent marking (Fig. 17). Others show various degrees of a colorful barred pattern, some with a prominent “abdominal window” marking, a reddish purple patch with conspicuous white streaks of myotomal fascia (this marking occurs in some other labrids, including *Halichoeres nebulosus* (Valenciennes, 1839) and some razorfishes). Larger IP adults can also show some blue lines and spots anterior to the opercular flap, but less prominent than on TP males.

The TP male of *H. sanchezi* shows the most divergence from relatives, most conspicuously the large black blotch covering the posterior caudal peduncle and the proximal half of the caudal fin. In contrast, the TP males of *H. salmofasciatus* (Fig. 12) and *H. malpelo* (Fig. 14) have black covering the posterior end of the caudal fin. They have a cross-hatched scale pattern on a mostly uniform greenish blue background with a pale abdomen, a yellowish wash on the snout, a bright red iris, and most have a pattern of reticulated blue lines and spots on the head behind the eye. The opercular flap ranges from a black oval to an inconspicuous bluish patch. Some photographs of *H. salmofasciatus* show a broad dusky midlateral band, broader than the red stripe of the IP, with only a dusky area on the rear caudal fin and these may include transitional stages from IP females to TP males. When disturbed, all adult members of the complex can apparently develop a barred pattern.

Both the IP and TP phases of *H. salmofasciatus* and *H. malpelo* are not clearly differentiated, and, in view of the variation in markings within these species, the taxonomic status of the two

species needs to be confirmed by additional documentation, as well as by molecular methods, which can assist in evaluating populations for species-level differences.

In sharp contrast to *H. sanchezi*, the TP male of *H. melanotis* shows a wide range of color patterns (Fig. 18). Its most diagnostic feature is the absence of a dark caudal-fin marking as found on island species. The head and body can range from blue-green to yellowish to orange, the body can show cross-hatching with vertical blue lines or rows of spots on each scale or can be uniform. The opercular flap is usually a distinct black vertical oval and a pale band extends from the chin across the lower operculum. All TP males have some degree of a complex pattern of small blue spots and lines on the head behind the eye. Some individuals with mostly IP patterns can also show these head patterns, perhaps including transitional stages. When the dorsal fin is raised, there is a small black spot on the first dorsal-fin membrane, as is found on TP *H. sanchezi*.

Remarks. The joint expedition in November 2022 to the Revillagigedo Archipelago pioneered a comprehensive approach to documenting an inventory of the fish assemblage of an island by combining underwater photographing, targeted collection of the photographed subjects, on-deck photographing of the fresh specimens, tissue sampling for molecular analysis, and preservation of vouchers for museum collections. It brought together an international group of scientists (the authors, Carlos Sanchez, D. Ross Robertson, Michelle Gaither, and Fernando Duarte), along with a cadre of some of the most experienced tropical marine underwater photographers (in alphabetical order): Alasdair Dunlap-Smith, Allison Morgan Estape, Carlos Estape, Jeffrey Haines, Andres Hernandez, Anne Johnson, Keri Kenning, Serenity Mitchell, Ellie Place, Lee Richter, and Sara Richter (Fig. 19). The expedition resulted in the most comprehensive

assessment of the fish fauna of a particular island to date. The results are in preparation for publication and include a species list, a photographic inventory, a DNA-barcode coverage report and documentation of several new species to be described from the islands (Robertson et al. in prep).

A benefit of the comprehensive team approach to inventorying the fish fauna of an island is the opportunity for a detailed documentation of the discovery of a new species of fish. The novelty of this justifies a description of the process. Detailed photographs and videos of the collection dive, the process of locating, subduing and bagging the holotype, and the procedures following on board to process the specimens are available at <https://doi.org/10.5281/zenodo.XXXXX>

In this case, an unknown species of wrasse was fleetingly observed and photographed by Kreg Martin in 2013 (Fig. 4). Both the TP and IP were observed at a single location, Punta Tosca on Socorro, at 20 m depth, on a REEF survey expedition led by Christy Semmens. Kreg Martin notes the fish were evasive and actively avoided divers and were especially difficult to photograph. At the time, the photographs were shown to author BCV and D. Ross Robertson and they concluded that it may represent an endemic species in the *H. melanotis* complex. However, no specimen was forthcoming and until our expedition, the status of the unknown species remained unresolved.

One of the priorities of the joint expedition was finding and documenting lesser-known species, especially those new to science. We did not encounter the unknown wrasse at Socorro or Clarion. However, on the last stop of the trip, at San Benedicto on 30 November 2022, the first dive of the morning was at the “El Canyon” dive site, to visit the shark-cleaning station. On the way back from the deep location, divers passed over a coarse gravel bed interspersed with small

rocks at about 20 m and Dr. Sanchez collected several small fishes, which included a single small IP of the new labrid species. On a subsequent dive targeting the same location, all of the divers focused on collecting, and a number of IP and juvenile wrasses were cornered and captured, not without difficulty since the fish sought refuge under rocks and then buried themselves in sand. Few TP males were observed, and one was cornered and lost. On a final dive, WBL managed to spear the previously lost TP male and WBL and BWF finally maneuvered the specimen into a bag (Fig. 20) and thus the holotype was firmly in hand.

Endemism. The remoteness of the Revillagigedo Archipelago has certainly contributed to the development of endemism and the ratio of endemic species there rivals that of any small oceanic island. Interestingly, the endemic fish species include both chaenopsids on the islands, two of the three known gobies, one of the two local *Thalassoma* species, *T. virens* (with a few records from Clipperton, and vagrants to Baja California in ENSO years (Victor et al. 2001), where they do not persist), and now two of the *Halichoeres* species. In comparison, the Galapagos Islands, Cocos Island, and Malpelo have no endemic *Thalassoma* species and fewer endemic *Halichoeres* species. They do match in having endemic chaenopsids and most of their gobies. A detailed exploration of Revillagigedo endemism will be reviewed in the upcoming inventory report of the fish fauna of the islands (Robertson et al. in prep.).

Conclusions

During an international effort to comprehensively inventory the ichthyofauna of the remote Revillagigedo Archipelago off the Pacific coast of Mexico, a new endemic species of wrasse (Labridae) was collected and documented by a team of underwater photographers. The new species, *Halichoeres sanchezi*, belongs to the *H. melanotis* species complex that is found

throughout the region, and it diverges 2.4% in the mtDNA cytochrome c oxidase I sequence from its nearest relative, *H. melanotis* from Panama, and 2.9% from *H. salmofasciatus* from Cocos Island, off Costa Rica. The juveniles and initial phase of the new species closely resemble those of *H. salmofasciatus* and *H. malpelo* from Malpelo Island off Colombia, while the terminal phase male (TP) has a color pattern distinct from other relatives, most notably having a prominent black patch on the caudal peduncle and base of the caudal fin. The new species adds to the list of endemic fish species for the isolated archipelago and is an interesting case of island endemism in the region.

Acknowledgements. We are especially grateful to Carlos Sanchez (UABCS) for initiating, arranging, facilitating, and leading the 2022 expedition, as well as creating an exceptionally friendly collegial atmosphere. Fernando Duarte, also from UABCS, is appreciated for his able assistance. D. Ross Robertson of the Smithsonian Tropical Research Institute provided unparalleled taxonomic expertise and his guide to SFTEP was the indispensable reference database to which to refer. Allison Morgan Estape was instrumental in organizing the logistics of getting so many people and their equipment in the same place at the same time. We are all indebted to Dora Sierra, the owner of the vessel Quino el Guardian, and the cooperative and patient crew and dive masters who made the trip possible (and safe). We thank William Bensted-Smith, Allison Morgan Estape, Carlos Estape, Jeff Haines, Andres Hernandez, Kreg Martin, Ellie Place, D. Ross Robertson, and Sara Richter for permission to use their photographs and Christy Semmens and Lourdes Vasquez-Yeomans for other assistance. Finally, the group of enthusiastic underwater photographers: Alasdair Dunlap-Smith, Allison Morgan Estape, Carlos Estape, Jeffrey Haines, Andres Hernandez, Anne Johnson, Keri Kenning, Serenity Mitchell, Ellie Place, Lee Richter, and Sara Richter, who provided much of the human energy for the project,

need to be acknowledged for their unparalleled skill in finding fishes, creating superbly composed photographs (usually while upside down and rolling), and being endlessly upbeat, humorous, and passionate about fishes and marine biology.

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Table 1. Proportional measurements of type specimens of *Halichoeres sanchezi*, n. sp.

484 Figures

485 Fig 1. Type location of *Halichoeres sanchezi* n. sp. on south side of San Benedicto Island (El
 486 Canyon dive site in the Caletilla Volteadura in the foreground), Revillagigedo Archipelago,
 487 Colima, Mexico (Raphael Gatti,
 488 https://commons.wikimedia.org/wiki/File:San_Benedicto_from_above.jpg, licensed under a
 489 Creative Commons Attribution 4.0 International (CC BY 4.0)).

490 Fig. 2. *Halichoeres sanchezi* n. sp. TP male holotype, LACM 61379, 123.1 mm SL, San
 491 Benedicto, Revillagigedo Archipelago, Mexico (Alasdair Dunlap-Smith).

492 Fig 3. *Halichoeres sanchezi* n. sp. IP paratype, LACM 61380, 46.2 mm SL, San Benedicto,
 493 Revillagigedo Archipelago (Alasdair Dunlap-Smith).

494 Fig 4. *Halichoeres sanchezi* n. sp. first known photographs, small IP (top), IP adult (middle) and
 495 TP males (below), taken in 2013 at Socorro, Revillagigedo Archipelago (Kreg Martin).

496 Fig 5. *Halichoeres sanchezi* n. sp. TP males underwater San Benedicto, Revillagigedo
 497 Archipelago, Mexico (a) Sara Richter; (b) Andres Hernandez).

498 Fig. 6. (a) *Halichoeres sanchezi* n. sp. IP adult underwater San Benedicto, Revillagigedo
 499 Archipelago, Mexico (Allison Morgan Estape); (b) large juvenile underwater San Benedicto,
 500 Revillagigedo Archipelago, Mexico (Jeff Haines).

501 Fig. 7. *Halichoeres sanchezi* n. sp. small juvenile underwater San Benedicto, Revillagigedo
 502 Archipelago, Mexico (Allison Morgan Estape).

Fig. 8. *Halichoeres sanchezi* n. sp. preserved holotype LACM 61379, 123.1 mm SL (a) and paratype LACM 61380, 53.3 mm SL (b) (William B. Ludt).

Fig. 9. *Halichoeres sanchezi* n. sp. fresh above and fluorescence below: TP male (a & c) holotype LACM 61379, 123.1 mm SL and IP (b & d) paratype LACM 61380, 53.3 mm SL (William B. Ludt).

Fig. 10. The neighbor-joining phenetic tree of New World species of *Halichoeres*

Fig. 11. *Halichoeres salmofasciatus*, juvenile (top), small IP (middle) and IP adult (bottom), Cocos Island, Costa Rica (top & bottom, Allison Morgan Estape; middle, D. Ross Robertson).

Fig. 12. *Halichoeres salmofasciatus*, TP males, Cocos Island, Costa Rica (top, Allison Morgan Estape; upper middle, William Bensted Smith; lower middle and bottom, D. Ross Robertson).

Fig. 13. *Halichoeres malpelo*, IP, Malpelo Island, Colombia (top, Yves Lefèvre, Biosphoto; upper middle, Hubert Yann, iStock; lower middle, Graham Edgar; bottom, Nippon Hōsō Kyōkai, Tokyo, Japan).

Fig. 14. *Halichoeres malpelo*, TP males, Cocos Island, Costa Rica (top & bottom, Yves Lefèvre, Biosphoto; middle, Graham Edgar).

Fig. 15. *Halichoeres melanotis*, juveniles from Panama, Coiba Island except upper middle from Perlas Islands (Allison Morgan Estape).

Fig. 16. *Halichoeres melanotis*, juvenile from Sea of Cortez, Mexico (Leonardo Gonzalez, depositphotos.com).

522 Fig. 17. *Halichoeres melanotis*, IP from Coiba Island, Panama except upper middle from Baja
523 California and middle from Perlas Islands, Panama (Allison Morgan Estape)

524 Fig. 18. *Halichoeres melanotis*, TP males, top, Baja California (Carlos Estape), upper middle
525 from Perlas Islands, Panama, lower two from Coiba Island, Panama (Allison Morgan Estape).

526 Fig. 19. Members of the joint expedition to Revillagigedo Archipelago in November 2022; left to
527 right, front row: Ellie Place, Serenity Mitchell, Anne Johnson, Michelle Gaither, Ambar Camila
528 Sanchez-Espinoza, Fernando Duarte, Keri Kenning, Benjamin Frable; back row: Andres
529 Hernandez, Alasdair Dunlap-Smith, Jeff Haines, Carlos Sanchez, Benjamin Victor, Ross
530 Robertson, William Ludt, Allison Morgan Estape, Carlos Estape, Sara Richter, and Lee Richter.

531 Fig. 20. Collection of the holotype of *Halichoeres sanchezi* n. sp. at San Benedicto Island,
532 Revillagigedo Archipelago, Mexico; left to right: William Ludt, Alasdair Dunlap-Smith,
533 Benjamin Frable, Keri Kenning (Carlos Estape).

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Figure 1

Type location of *Halichoeres sanchezi* n. sp. on south side of San Benedicto Island (El Canyon dive site in the Caletilla Volteadura in the foreground), Revillagigedo Archipelago, Colima, Mexico

photograph by Raphael Gatti,

https://commons.wikimedia.org/wiki/File:San_Benedicto_from_above.jpg, licensed under a Creative Commons Attribution 4.0 International (CC BY 4.0)



Figure 2

Halichoeres sanchezi n. sp. TP male holotype, LACM 61379, 123.1 mm SL, San Benedicto, Revillagigedo Archipelago, Mexico

photograph by Alasdair Dunlap-Smith



Figure 3

Halichoeres sanchezi n. sp. IP paratype, LACM 61380, 46.2 mm SL, San Benedicto, Revillagigedo Archipelago

photograph by Alasdair Dunlap-Smith

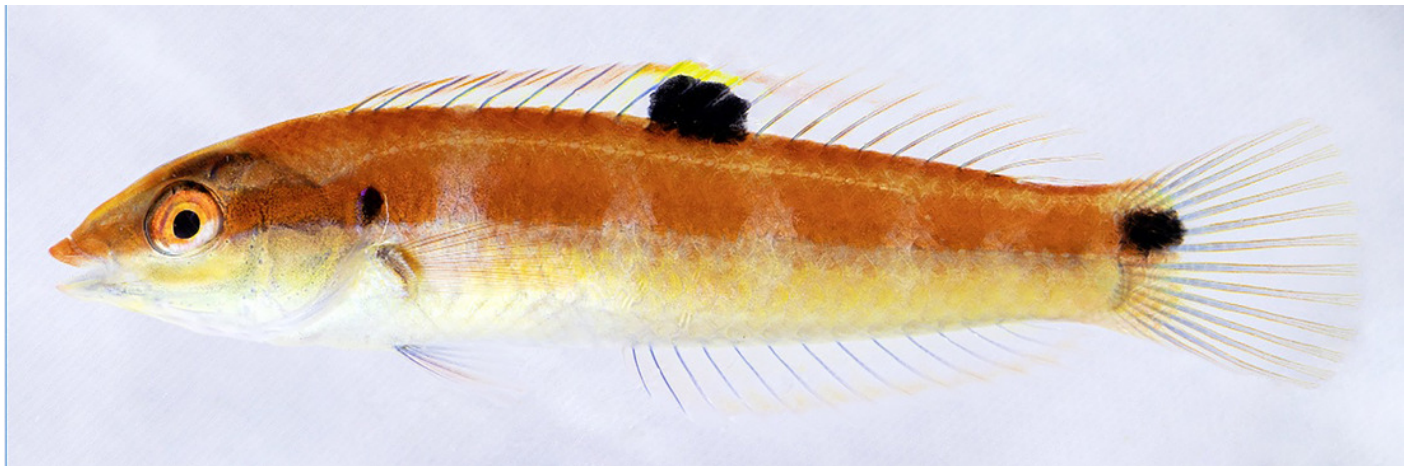


Figure 4

Halichoeres sanchezi n. sp. first known photographs from 2013: small IP (top), IP adult (middle) and TP males (below), at Socorro, Revillagigedo Archipelago

photographs by Kreg Martin



Figure 5

Halichoeres sanchezi n. sp. TP male underwater, San Benedicto, Revillagigedo Archipelago, Mexico

photograph by Sara Richter



Figure 6

Halichoeres sanchezi n. sp. TP male underwater, San Benedicto, Revillagigedo Archipelago, Mexico

photograph by Andres Hernandez

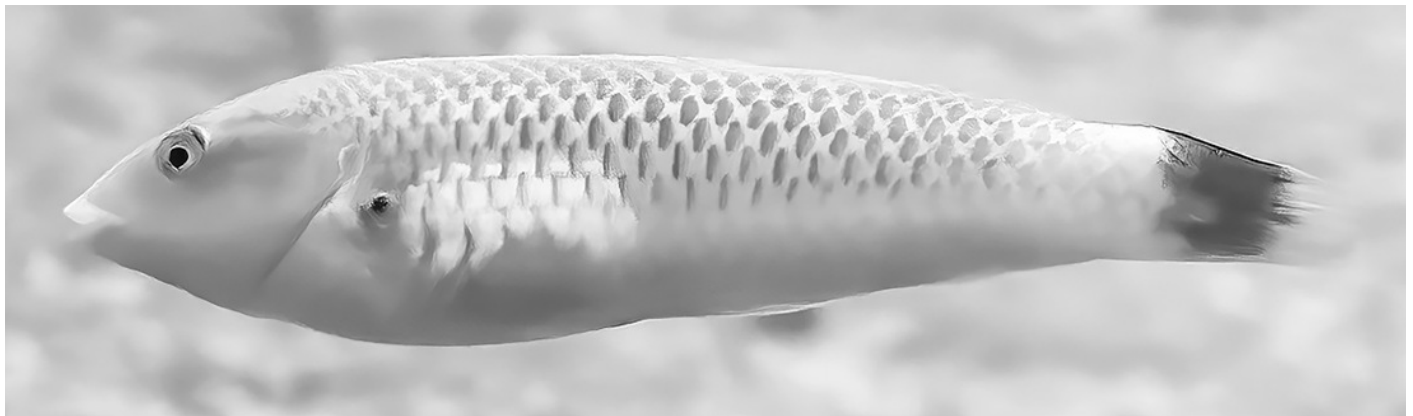


Figure 7

Halichoeres sanchezi n. sp. IP adult underwater, San Benedicto, Revillagigedo Archipelago, Mexico

photograph by Allison Morgan Estape



Figure 8

Halichoeres sanchezi n. sp. large juvenile underwater, San Benedicto, Revillagigedo Archipelago, Mexico

photograph by Jeff Haines



Figure 9

Halichoeres sanchezi n. sp. small juvenile underwater, San Benedicto, Revillagigedo Archipelago, Mexico

photograph by Allison Morgan Estape



Figure 10

Halichoeres sanchezi n. sp. a) preserved holotype, 123.1 mm SL, LACM 61379; b) preserved paratype, 53.3 mm SL, LACM 61380

photograph by William B. Ludt

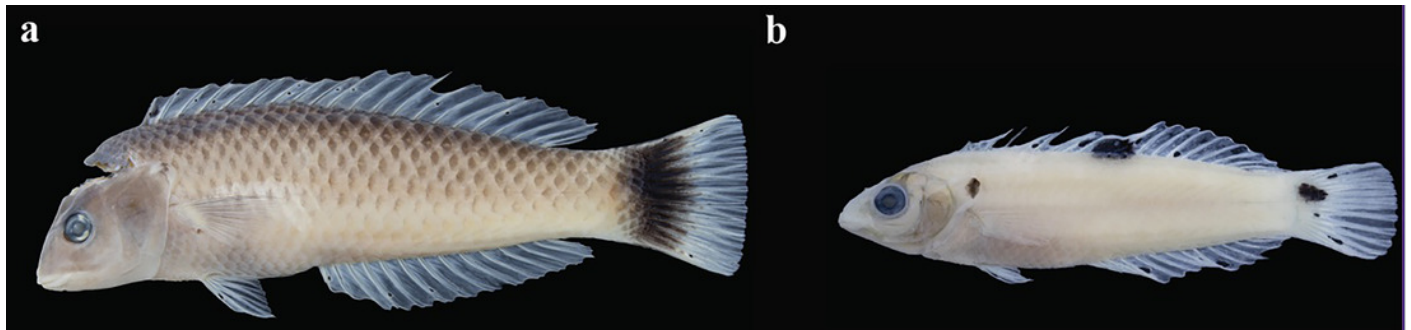


Figure 11

Halichoeres sanchezi n. sp. fluorescence pattern: (a & c) TP male holotype LACM 61379; (b & d) IP paratype LACM 61380

photographs by William B. Ludt

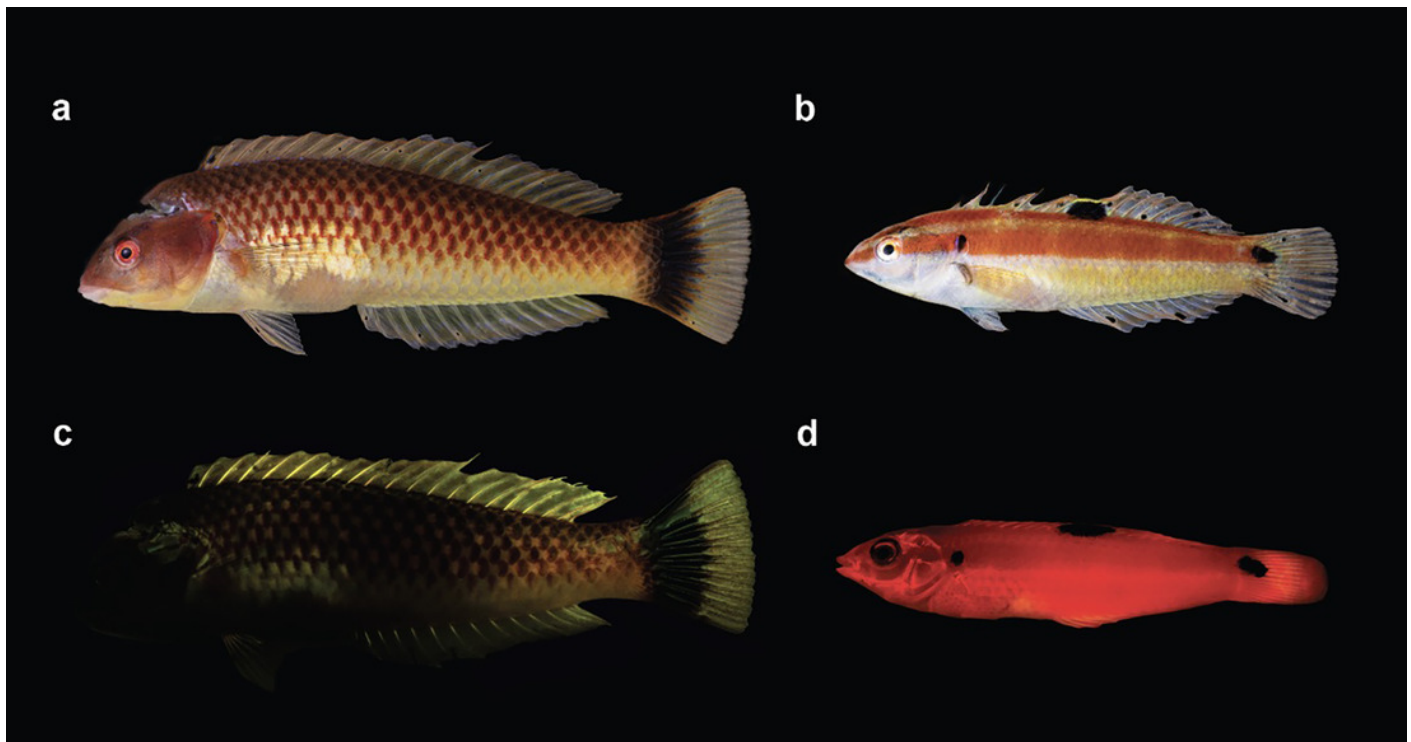


Figure 12

Neighbor-joining tree of mtDNA COI sequences generated by BOLD (www.boldsystems.org) for the *Halichoeres* species of the New World

Each sequence has the species identification, location of collection and GenBank number indicated. The sequence of *Halichoeres argus* is used as an outgroup and the scale bar at left represents a 2% sequence difference

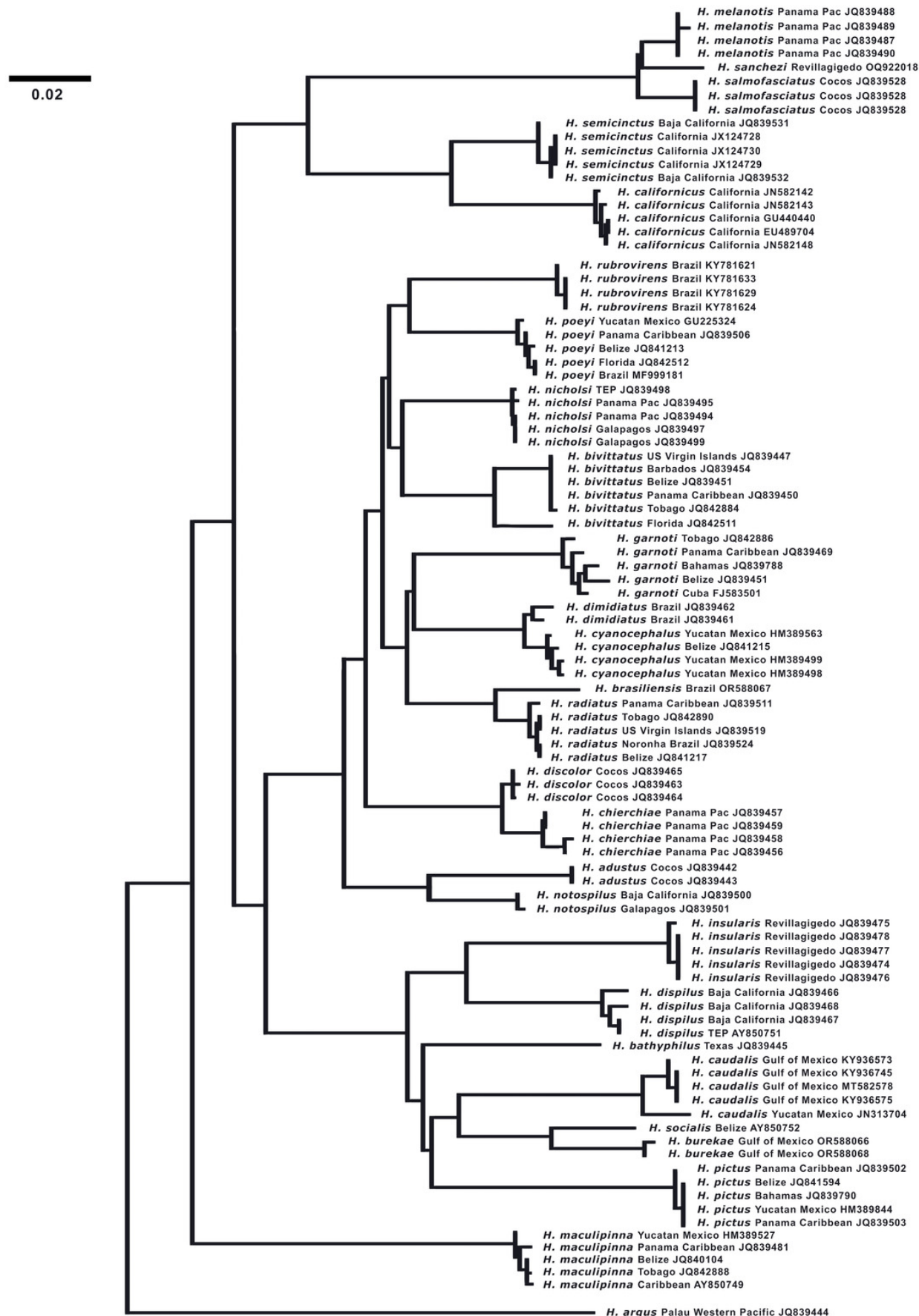


Figure 13

Halichoeres salmofasciatus, juvenile (top), small IP (middle) and IP adult (bottom), Cocos Island, Costa Rica

photographs top & bottom by Allison Morgan Estape; middle by D. Ross Robertson



Figure 14

Halichoeres salmofasciatus, TP males, Cocos Island, Costa Rica

photographs: top by Allison Morgan Estape; upper middle by William Bensted Smith; lower middle and bottom by D. Ross Robertson



Figure 15

Halichoeres malpelo, IP, Malpelo Island, Colombia

photographs: top by Yves Lefèvre, Biosphoto; upper middle by Hubert Yann, iStock; lower middle by Graham Edgar; bottom, by Nippon Hōsō Kyōkai, Tokyo, Japan



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Figure 16

Halichoeres malpelo, TP males, Cocos Island, Costa Rica

photographs top & bottom by Yves Lefèvre, Biosphoto; middle by Graham Edgar



Figure 17

Halichoeres melanotis, juveniles from Panama at Coiba Island, except upper middle from Perlas Islands

photographs by Allison Morgan Estape



Figure 18

Halichoeres melanotis, juvenile from Sea of Cortez, Mexico

photograph by Leonardo Gonzalez, depositphotos.com



Figure 19

Halichoeres melanotis, IP from Coiba Island, Panama, except upper middle from Baja California and middle from Perlas Islands, Panama

photographs by Allison Morgan Estape



Figure 20

Halichoeres melanotis, TP males, top Baja California, upper middle Perlas Islands, Panama, and lower two from Coiba Island, Panama

photographs: top by Carlos Estape, remainder by Allison Morgan Estape



Figure 21

Members of the joint international expedition to the Revillagigedo Archipelago, Colima, Mexico in November 2022

left to right, front row: Ellie Place, Serenity Mitchell, Anne Johnson, Michelle Gaither, Ambar Camila Sanchez-Espinoza, Fernando Duarte, Keri Kenning, Benjamin Frable; back row: Andres Hernandez, Alasdair Dunlap-Smith, Jeff Haines, Carlos Sanchez, Benjamin Victor, Ross Robertson, William Ludt, Allison Morgan Estape, Carlos Estape, Sara Richter, and Lee Richter



Figure 22

Collection of the holotype of *Halichoeres sanchezi* n. sp. at San Benedicto Island, Revillagigedo Archipelago, Mexico

left to right: William Ludt, Alasdair Dunlap-Smith, Benjamin Frable, Keri Kenning (photograph by Carlos Estape



Table 1 (on next page)

Proportional measurements of type specimens of *Halichoeres sanchezi*, n. sp., as percentages of the standard length

TABLE 1
Proportional measurements of type specimens of *Halichoeres sanchezi*, n. sp.
as percentages of the standard length

	holotype	paratypes					
	LACM 61379	LACM 61380	SIO 23-50	CNP IBUNAM XXXXX	SIO 23-51	SIO 23-48	CNP IBUNAM XXXXX
Standard length (mm)	123.1	53.3	46.2	40.4	39.1	36.5	32.3
Body depth	27.8	25	24.5	23.8	24.2	24.9	26.4
Body width	11.3	9.4	11.3	9.8	10.7	10.6	9.3
Head length	30.8	30.9	32	32.7	31.6	31.5	32.5
Snout length	8.8	8.5	9.1	8.2	8.1	8.5	8.2
Orbit diameter	5.6	6.8	6.5	7.6	7.5	7.8	7.9
Interorbital width	5.9	5.5	5.7	5.5	4	4.1	4.2
Upper-jaw length	7.2	5.8	6.1	6.4	6.9	6.5	6.6
Caudal-peduncle depth	14.2	13.7	13.2	13.4	13.5	13.2	13.2
Caudal-peduncle length	12.7	7.7	9.0	8.4	8.5	6.6	8.9
Predorsal length	31.5	29.8	29.9	29.4	29.9	29.7	29.6
Preanal length	47.4	53.5	55.6	58	57.1	55.3	56.5
Prepelvic length	27.5	32.1	33.3	32.9	30.9	32.7	30.9
Dorsal-fin base	66.8	60.2	62.9	62.4	64.6	66.7	66.8
First dorsal-fin spine	5.3	-	4.1	5.0	3.9	3.7	3.8
Longest dorsal-fin spine	8.2	8.2	8.2	9.2	8.4	8.5	8.9
Longest dorsal-fin ray	12.5	10.5	10.8	10.5	10.5	10.6	11.2
Anal-fin base	39.0	39.3	35.5	37.3	37.9	35.4	33.9
First anal-fin spine	2.9	2.3	1.9	2.2	2.4	2.2	2.1
Second anal-fin spine	4.9	5.6	5.4	6.1	6.3	6.2	5.9

Third anal-fin spine	6.8	7.3	7.6	7.9	7.6	7.6	7.2
Longest anal-fin ray	10.4	9.6	9.3	9.8	9.9	9.9	9.6
Caudal-fin length	16.4	19.9	21.7	23.3	24.9	24.5	25.8
Pectoral-fin length	17.9	17.2	16.2	19.1	17.6	17.8	18.1
Pelvic-fin spine length	8.8	8.8	8.6	9.3	9.2	8.1	7.8
Pelvic-fin length	13.3	13.1	13.1	13.0	13.4	13.2	13.4

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