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Chrysiptera caesifrons, a new species of damselfish (Pomacentridae) from the south-western Pacific Ocean

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Abstract

Chrysiptera caesifrons is described on the basis of 23 type specimens, 23.3–48.5 mm SL, from the Raja Ampat Islands (West Papua Province), Indonesia, and 184 additional non-types, 17.8–60.7 mm SL, from Papua New Guinea, Vanuatu, New Caledonia, and the Great Barrier Reef of Australia. Additional records based on photographic evidence and underwater observations include Halmahera and the Solomon Islands. The new species is closely allied to *C. rex* from the Ryukyu Islands, Taiwan, Palau, eastern Indonesia (eastern Kalimantan and Java eastward), Timor Leste, Raja Ampat Islands (off western extremity of New Guinea), and offshore reefs of Western Australia, as well as to *C. chrysocephala* from the South China Sea region and Sulawesi. Although the new species was previously confused with *C. rex*, it is clearly separable from both sibling species on the basis of colour pattern and from *C. rex* by a 6.4% difference (average pairwise distance) in the mitochondrial control region DNA sequence and from *C. chrysocephala* by a 4.3% difference (K2P minimum interspecific distance) in the barcode COI mtDNA sequence.

Key words: coral reef fishes, taxonomy, systematics, Indo-Pacific Ocean, barcode DNA sequence.

Introduction

The genus *Chrysiptera* contains relatively small, often brightly coloured damselfishes, which were formerly included in the genus *Abudefduf* Forsskål 1775. Allen (1991) reviewed the 25 known species, but since then an additional eight species were described (Allen & Adrim 1992, Allen & Rajasuriya 1995, Allen 1999, Allen & Bailey 2002, Manica *et al*. 2002, Allen & Erdmann 2008, Allen *et al*. 2010). In addition, Allen (in *Coastal Fishes of the Western Indian Ocean*, in preparation) recognizes *Chrysiptera leucopoma* (Cuvier 1830) as the Pacific sibling of *C. brownriggiii* (Bennett 1828), which is now restricted to the Indian Ocean. Genetic evidence supports the split between ocean basins, with the barcode COI mtDNA sequences of the two species 3.53% divergent (pairwise minimum interspecific distance) on the barcode database BOLD, including sequences for *C. brownriggii* from Seychelles, Madagascar and Reunion and *C. leucopoma* from French Polynesia. Hubert *et al*. (2012) noted the numerous instances of genetic divergence between Indian and Pacific Ocean populations of many coral reef fishes and reported *C. brownriggiii* as an example of their Pattern 2 (geographic monophyly, deep divergence) with a 3.82% maximum K2P distance between populations from Reunion and French Polynesia. The genus *Chrysiptera*, as presently defined (Allen 1991), contains 34 species (Table 1), including the new species described herein. However, it should be noted that genetic studies (Quenouille *et al*. 2004, Cooper *et al*. 2009) indicate the genus is likely polyphyletic, containing at least three disparate deep genetic lineages.

Pomacentrids generally exhibit a wide range of colour variation, often related to growth stage, sex, or environmental surroundings (Allen 1974, 1991). Some species, particularly those which are relatively wide ranging, also show various degrees of geographical variation. However, there is mounting genetic evidence indicating that many of these so-called geographic "variants" are actually separate species (Allen *et al*. 2008, Drew *et al*. 2008, Drew & Barber 2009, Allen & Drew 2012, Victor 2015). The present study provides yet another example of a wide-ranging taxon, *Chrysiptera rex* (Snyder 1909), that actually represents a multi-species complex, including both sympatric and allopatric sibling species.

This study provides clarification of the species complex containing *Chrysiptera rex*, *C. chrysocephala*, and the new species *Chrysiptera caesifrons*. The taxonomy of these species, formerly considered geographic variants of *C. rex*, has been confused in the recent past. Although Drew *et al*. (2010) provided genetic evidence for the recognition of three separate species, the fish they illustrated as *C. rex* from the Ryukyu Islands (type locality) was incorrectly identified and is the juvenile of a still undetermined species. The identity of the true *C. rex* was further confused by Allen & Erdmann (2012), who illustrated a fish from Palawan, Philippines as this species. However, our subsequent research reveals the Palawan fish was previously described in a relatively obscure publication (*Malayan Nature Journal*) as *Chrysiptera chrysocephala*. The three members of this species complex are closely related, exhibiting no significant morphometric or meristic differences. Fortunately they are readily separable on the basis of consistent colour pattern differences, as well as by different mtDNA sequences. They also appear to have largely allopatric distributions, although the new species and *C. rex* have overlapping ranges at West Papua, Indonesia.

Materials and Methods

Lengths of specimens are given as standard length (SL) measured from the anterior end of the upper lip to the base of the caudal fin (posterior edge of hypural plate); head length (HL) is measured from the same anterior point to the posterior edge of the opercle flap; body depth is the maximum depth taken vertically between the belly and base of the dorsal spines; body width is the maximum width just posterior to the gill opening; snout length is measured from the anterior end of the upper lip to the anterior edge of the eye; orbit diameter is the horizontal fleshy diameter, and interorbital width the least fleshy width; upper-jaw length is taken from the front of the upper lip to the posterior end of the maxilla; caudal-peduncle depth is the least depth, and caudal-peduncle length is the horizontal distance between verticals at the rear base of the anal fin and the caudal fin base; lengths of fin spines and rays are measured to their extreme bases (i.e., not from the point where the ray or spine emerges from the

TABLE 1

Valid species of *Chrysiptera*, in alphabetical order

	species	author & date	species distribution
1	*albata*	Allen & Bailey, 2002	Phoenix Islands
2	*annulatus*	(Peters, 1855)	Red Sea & W. Indian Ocean
3	*arnazae*	Allen *et al*., 2010	E. Indonesia to Papua New Guinea
4	*biocellata*	(Quoy & Gaimard, 1824)	Indo-West Pacific
5	*brownriggiii*	(Bennett, 1828)	Indian Ocean
6	*bleekeri*	(Fowler & Bean, 1928)	Indonesia & Philippines
7	*caeruleolineata*	(Allen, 1973)	Eastern Indian Ocean & W. Pacific
8	*caesifrons*	Allen *et al*., new species	Southwestern Pacific
9	*cyanea*	(Quoy & Gaimard, 1824)	Eastern Indian Ocean & W. Pacific
10	*cymatilis*	Allen, 1999	Eastern PNG and Solomon Is.
11	*chrysocephala*	Manica *et al*., 2002	South China Sea and Gulf of Tomini
12	*flavipinnis*	(Allen & Robertson, 1974)	Southwestern Pacific
13	*galba*	(Allen & Randall, 1974)	Southeastern Pacific
14	*giti*	Allen & Erdmann, 2008	Sulawesi and West Papua
15	*glauca*	(Cuvier, 1830)	Indo-West Pacific
16	*hemicyanea*	(Weber, 1913)	E. Indonesia & Timor Sea
17	*kuiteri*	Allen & Rajasuriya, 1995	Eastern Indian Ocean to W. Papua
18	*leucopoma*	(Cuvier, 1830)	Western Pacific
19	*niger*	(Allen, 1975)	Eastern Papua New Guinea
20	*notialis*	(Allen, 1975)	Southwestern Pacific
21	*oxycephala*	(Bleeker, 1877)	Indo-Melanesian Archipelago
22	*parasema*	(Fowler, 1918)	Western Pacific
23	*pricei*	Allen & Adrim, 1992	Cenderawasih Bay, W. Papua
24	*rapanui*	(Greenfield & Hensley, 1970)	Kermadec Is. & Easter I.
25	*rex*	(Snyder, 1909)	Ryukyu Islands to Indonesia
26	*rollandi*	(Whitley, 1961)	Indo-Australian Archipelago
27	*sinclairi*	Allen, 1987	Bismarck Archipelago
28	*springeri*	(Allen & Lubbock, 1976)	Indonesia & Philippines
29	*starcki*	(Allen, 1973)	Western Pacific
30	*talboti*	(Allen, 1975)	Indo-Australian Archipelago
31	*taupou*	(Jordan & Seale, 1916)	Southwestern Pacific
32	*traceyi*	(Woods & Schultz, 1960)	Marshall & Caroline Islands
33	*tricincta*	(Allen & Randall, 1974)	Western Pacific
34	*unimaculata*	(Cuvier, 1830)	Indo-West Pacific

basal scaly sheath); caudal-fin length is the horizontal length from the posterior edge of the hypural plate to a vertical at the tip of the longest ray; caudal concavity is the horizontal distance between verticals at the tips of the shortest and longest rays; pectoral-fin length is the length of the longest ray; pelvic-fin length is measured from the base of the pelvic spine to the filamentous tip of the longest soft ray; pectoral-ray counts include the small splint-like, uppermost rudimentary ray; only the tube-bearing anterior lateral-line scales are counted; a separate count is given for the deeply pitted scales occurring in a continuous series midlaterally on the caudal peduncle; the decimal figure ".5" appearing in the scale row count above the lateral line refers to a small truncated scale at the base of the dorsal fin; gill-raker counts include all rudiments and are presented as separate counts for the upper and lower limbs as well as a combined count; the last fin ray element of the dorsal and anal fins is usually branched near the base and is counted as a single ray.

Counts and proportions appearing in parentheses apply to the paratypes. Proportional measurements for the new taxon, expressed as percentage of the standard length, are provided in Table 2.

Type specimens have been deposited at the Museum Zoologicum Bogoriense, Cibinong, Java, Indonesia (MZB) and the Western Australian Museum, Perth (WAM). Non-types and additional comparative material are housed at the Bishop Museum, Honolulu (BPBM), California Academy of Sciences, San Francisco (CAS), Institut Penyelidikan Marin Borneo, Kota Kinabalu (IPMB), Royal Ontario Museum, Toronto (ROM), and United States National Museum of Natural History, Washington, D.C. (USNM).

Tissue samples of nine individuals of *Chrysiptera caesifrons* and two *C. rex* were collected from the type locality. The specimens were preserved in 95% EtOH. Mitochondrial DNA was extracted using a 10% Chelex solution (Walsh *et al*. 1991). A portion of the mitochondrial control region was amplified via the polymerase chain reaction (PCR) using the primers CRK: 5'-AGC TCA GCG CCA GAG CGC CGG TCT TGT AAA-3' and CRE: 5'-CCT GAA GTA GGA ACC AGA TG-3'. The PCR reaction was carried out in 25 µl volumes, using 1 µl of template. Each reaction included 4 µl 10x PCR buffer (Applied Biosystems), 2.5 µl (10 mM dNTPs), 1.25 µl of each primer at 10 mM, 2 µl (25 mM $MgCl_2$ solution), 0.125 µl AmplyTaq Red (Applied Biosystems) and 14.5 µl ddH_2O. The thermocycling profile for CR included an initial denaturation of 80°C for 10 min, 94°C for 15 min, 38 cycles of 94°C for 15 s, 50°C for 30 s, and 72°C for 45 s, with a final extension of 72°C for 5 min. PCR reactions were checked on 1% agarose gels stained with ethidium bromide. The PCR product was sequenced at the UC Berkeley sequencing facility.

Forward and reverse sequences were proofread in MEGA5 (Tamura *et al*. 2011) and then aligned using MUSCLE (Edgar 2004). Three methods were used to generate tree reconstructions: neighbor joining and maximum likelihood using MEGA5 and Bayesian inference using MRBAYES 3.2 (Ronquist & Hulsenbeck 2003). Neighbor joining was used to calculate relationships between individuals based on genetic distance. The maximum likelihood analysis was used to assess the model of best fit for the nucleotide substitutions. Akaike Information Criterion (AIC) was used to rank the Hasegawa-Kishino-Yano (HKY) model with a discrete Gamma distribution (HKY+G) to derive the best fit to the data. This model assumes different rates of transitions and transversions as well as different nucleotide frequencies, and was chosen as the appropriate model of evolution as determined by JModelTest 0.1.1 (Posada 2008). Bootstrap support was determined using 1000 replicates in MEGA5. For the Bayesian analysis we used a Markov Chain Monte Carlo (MCMC) approach with four chains. Analyses were run for 100,000,000 generations. After 100,000,000 generations, the resulting tree was checked for convergence using Tracer v1.5 (Rambaut & Drummond 2009).

In addition to the mitochondrial control region, the mtDNA COI marker is frequently used for species identification and assessing genetic divergence between populations of coral reef fishes (Ward *et al*. 2009, Victor 2015). Barcode mtDNA sequences for the *C. rex* complex are available as public records on the barcode database BOLD (http://www.boldsystems.org/). Species identifications were confirmed from voucher photographs attached to the records. K2P and pairwise distances were calculated by algorithms used in the distance summary report on the BOLD website.

Figure 1. *Chrysiptera caesifrons*, preserved holotype, MZB 22260, 48.5 mm SL, Ayau Atoll, Raja Ampat Islands, West Papua, Indonesia (G.R. Allen).

Chrysiptera caesifrons, n. sp.

Greyback Damsel

Figures 1–2 & 7a; Tables 2–6.

Chrysiptera rex (non-Snyder 1909) Randall *et al*. 1997: 265 (Great Barrier Reef, Australia).
Chrysiptera species Allen & Erdmann 2012: 595 (photograph of Halmahera fish).

Holotype. MZB 22260, 48.5 mm SL, Ayau Kecil Island, 00° 21.342'S, 130° 58.648'E, Ayau Atoll, Raja Ampat Islands, West Papua Province, Indonesia, 1.5–2.0 m, clove oil, M. Erdmann & G. Allen, 17 October 2014.

Paratypes. MZB 22259, 6 specimens, 33.2–42.8 mm SL, collected with holotype; MZB 22258, 4 specimens, 31.9–47.9 mm SL, Equator Islands, 00° 00.102'S, 130° 10.648'E, Raja Ampat Islands, West Papua Province, Indonesia, 2 m, clove oil, M. Erdmann, 28 August 2010; WAM P.33346-001, 6 specimens, 23.3–47.8 mm SL, collected with MZB 22258; WAM P.34295-001, 6 specimens, 36.1–48.4 mm SL, collected with holotype.

Non-type specimens. AMS I.15620–008, 2 specimens, 50.4–56.9 mm SL, One Tree Island, 23° 30.657'S, 152° 05.197'E, Capricorn Group, Queensland, Australia, 1-3 m, explosives, AMS party 26 November 1966; AMS I.17109–017, 2 specimens, 43.0–53.0 mm SL, One Tree Island, Capricorn Group, Queensland, Australia, 2 m, rotenone, G. Allen, 15 May 1973; AMS I.17445–036, 3 specimens, 43.6–55.7 mm SL, One Tree Island, Capricorn Group, Queensland, Australia, 3–4 m, rotenone, F. Talbot, 19 September 1968; AMS I.17512–008, 46.1 mm SL, Passe de Dumbea, 22° 24'S, 166° 16'E, New Caledonia, 3 m, spear, G. Allen, 13 June 1973; AMS I.19473–023, 96 specimens, 17.8–54.6 mm SL, Lizard Island, 14° 40'S, 145° 27'E, Queensland, Australia, 2–7 m, rotenone, AMS party, 24 November 1975; AMS I.19483–006, 27 specimens, 18.1–49.2 mm SL, sand cay north of Lizard

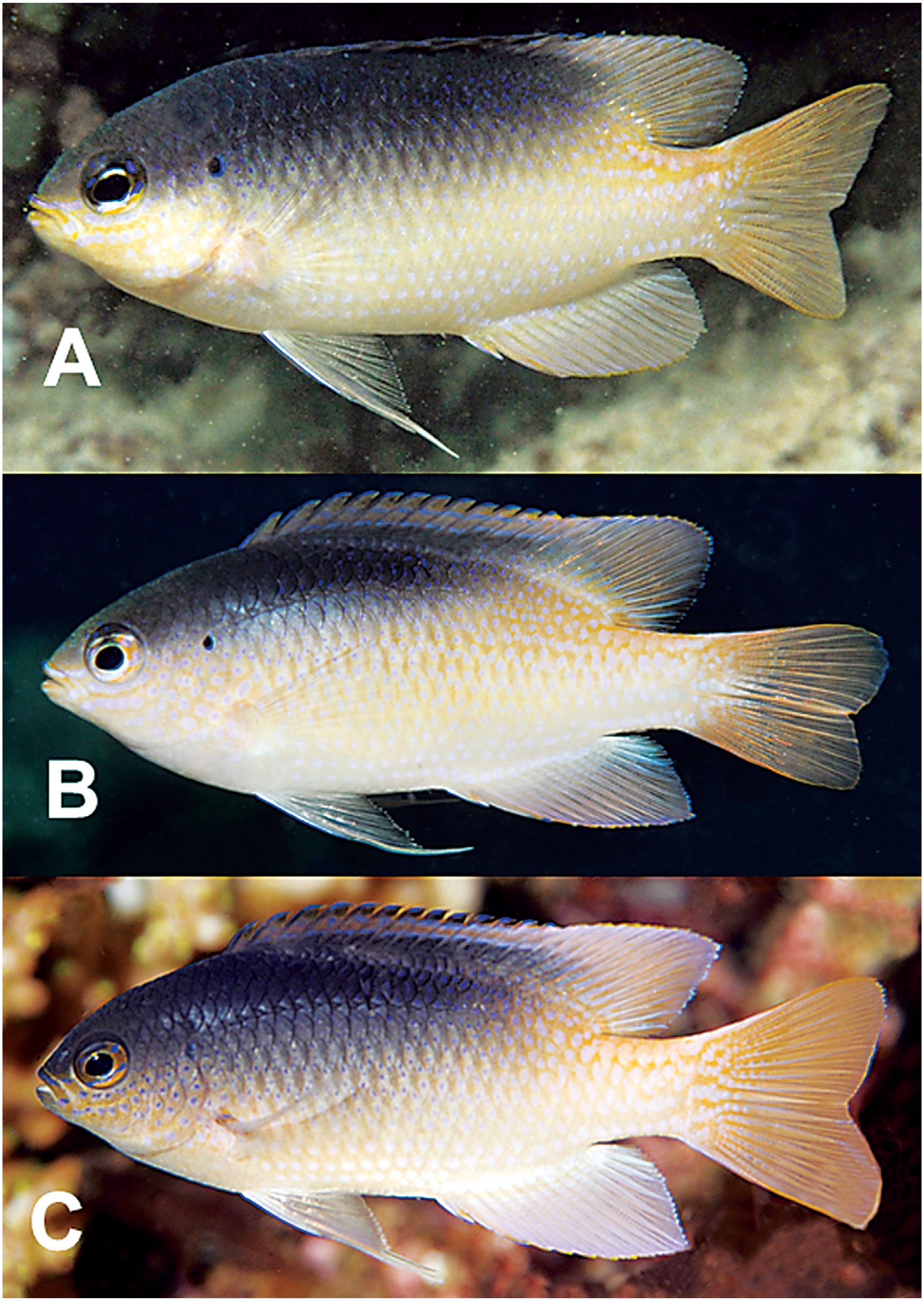

Figure 2. *Chrysiptera caesifrons*, underwater photographs. A. Ayau Atoll, Raja Ampat Islands, West Papua, Indonesia, approximately 45 mm SL; B. Duke of York Islands, Papua New Guinea, approximately 45 mm SL; C. New Georgia Group, Solomon Islands, approximately 50 mm SL (G.R. Allen).

Island, 14° 38'S, 145° 27'E, Queensland, Australia, 1–3 m, rotenone, AMS party, 27 November 1975; AMS I.20774–066, 16 specimens, 36.2–52.2 mm SL, off Cape Melville, 13° 56'S, 144° 36'E, Queensland, Australia, 0–8 m, rotenone, AMS party, 9 February 1979; AMS I.21422-092, 7 specimens, 19.6-48.8 mm SL, Lizard Island, 14° 41'S, 145° 26'E, Queensland, Australia, 1–8 m, rotenone, D. Hoese, 27 January 1975; USNM 281488, 3 specimens, 44.3–60.7 mm SL, One Tree Island, Capricorn Group, Queensland, Australia, 8–13 m, rotenone, V.G. Springer, 7 December 1966; USNM 281531, 2 specimens, 44.0–46.1 mm SL, One Tree Island, Capricorn Group, Queensland, Australia, 8–13 m, rotenone, V.G. Springer, 7 December 1966; USNM 362698, 4 specimens, 35.3–46.1 mm SL, Lakona Bay, 14° 22.333'S, 167° 24.000'E, Santa Maria Island, Banks Islands, Vanuatu, 2–7 m, rotenone, J.T. Williams and party, 14 May 1997; USNM 363217, 5 specimens, 35.1–52.1 mm SL, Vanua Lava, 13° 52.416'S, 167° 32.200'E, Banks Islands, Vanuatu, 1–3 m, rotenone, J.T. Williams and party, 17 May 1997; WAM P.25938–025, 35.0 mm SL, Lizard Island, Queensland, Australia, 1–2 m, Australian Museum party, November 1975; WAM P.27485–003, 7 specimens, 19.9–48.4 mm SL, Escape Reef, approximately 15° 50'S, 145° 50'E, Great Barrier Reef, Queensland, Australia, 1–2 m, rotenone, W.A. Starck, 18 December 1981; WAM P.33808–001, 8 specimens, 36.8–44.9 mm SL, Duke of York Islands, 04° 06.938'S, 152° 26.504'E, New Britain, Papua New Guinea, 1.5–2.0 m, clove oil, G. Allen, 26 November 2012.

Diagnosis. Dorsal-fin rays XIII,12-14 (usually 13); anal-fin rays II,12–14 (usually 13); pectoral rays 16–18 (usually 17); gill rakers on first branchial arch 4–6 + 11–13 total rakers 16–19 (usually 17–18); tubed lateral-line scales 16–18 (usually 17); colour in life overall pale yellow to whitish with dark bluish (nearly black) to grey region encompassing snout, upper head and adjacent anterodorsal portion of body and spinous dorsal fin.

Description. Dorsal-fin rays XIII,13 (12–14); anal-fin rays II,13 (12–14); pectoral rays 17 (16–18); principal-caudal rays 15; branched-caudal rays 13; upper and lower accessory-caudal rays 4–5 (4–6); gill rakers on first branchial arch 5 + 11 (4–6 + 12–13), total rakers 16 (17–19); lateral-line scales with tubes 17 (16–18); vertical scale rows 27 (27–28); scales above lateral-line to base of middle dorsal spines 1.5; scales below lateral line to anus 9; tubed lateral-line scales ending below anterior soft rays of dorsal fin; often 1–2 pitted or pored scales immediately posterior to last lateral-line scale; a series of 7 (7–9) pored or pitted scales mid-laterally on caudal peduncle to caudal base.

Body depth 2.5 (2.3–2.6) in SL; maximum body width 2.3 (2.2–2.6) in depth; HL contained 3.5 (3.2–3.6) in SL; snout 4.1 (3.9–5.4), eye 3.0 (2.7–2.9), interorbital width 3.3 (3.2–3.9), length of upper jaw 3.3 (3.1–3.4); least depth of caudal peduncle 1.8 (1.8–2.0), length of caudal peduncle 2.1 (1.9–2.7), all in HL.

Mouth oblique, terminal, the maxillary reaching to a vertical slightly beyond anterior edge of eye; teeth of jaws biserial, those of outer row conical with rounded, slightly incurved tips; about 40 (36–40) teeth in outer row of lower jaw and 42 (36–42) in upper, the largest about one-fourth diameter of pupil in height; a secondary row of slender buttress teeth behind those of outer row in the spaces between them; single nasal opening on each side of snout; nostril with a low fleshy rim; greatest depth of preorbital-suborbital about 1.2 (1.2–1.5) in pupil diameter, the ventral margin smooth; margins of preopercle and opercular series smooth.

Scales of head and body finely ctenoid; anterior part of preorbital, snout tip, lips, chin, and isthmus naked; a single row of small scales beginning on posterior preorbital, extending over suborbital to posterior circumorbitals; preopercle with 2 primary scale rows and row of reduced scales along lower margin, rear margin narrowly naked; dorsal and anal fins with a basal scaly sheath, including scales that cover at least proximal half of inter-radial membranes anteriorly, gradually decreasing towards rear portion of fins; proximal one-half to two-thirds of caudal fin covered by scales (often partially missing in preserved specimens); paired fins covered by scales only at base; length of axillary scale of pelvic fins about 55–73 percent pelvic spine length.

Origin of dorsal fin at level of third tubed scale of lateral line; spines of dorsal fin gradually increasing in length to last spine; membrane between spines moderately incised; first dorsal spine 3.9 (3.8–5.2), seventh dorsal spine 2.0 (2.0–2.3), last dorsal spine 1.8 (1.7–2.2), all in HL; longest (7th or 8th) soft dorsal ray 1.2 (1.2–1.5) in HL; length of dorsal fin base 1.6 (1.6–1.7) in SL; first anal spine 3.8 (3.8–4.5), second anal spine 1.6 (1.7–1.8), both in HL; longest (8th or 9th) soft anal ray 1.1 (1.0–1.6) in HL; base of anal fin 2.3 (2.2–2.6) in base of dorsal fin; caudal fin emarginate, its length 3.2 (3.1–3.5) in SL; pectoral fin relatively short, not reaching a vertical through anal opening, the longest ray 3.8 (3.6–4.2) in SL; first soft ray of pelvic fin with filamentous tip reaching base of second or third soft anal ray if undamaged, pelvic-fin length 3.1 (2.7–3.3) in SL.

TABLE 2

Proportional measurements of selected type specimens of *Chrysiptera caesifrons*, n. sp. as percentages of the standard length

	holotype	paratypes					
	MZB 22260	MZB 22258	WAM P 33346	WAM P 34295	WAM P 33346	WAM P 34295	MZB 22258
Standard length (mm)	48.5	47.9	44.8	43.0	41.9	36.1	31.9
Body depth	40.1	41.1	41.7	42.2	43.0	41.9	41.7
Body width	17.6	16.1	18.8	17.8	17.4	17.7	16.6
Head length	28.4	28.8	30.1	29.2	29.8	31.5	31.7
Snout length	7.0	7.3	7.8	6.9	7.2	5.9	6.3
Orbit diameter	9.4	10.0	10.9	10.2	11.0	11.8	11.6
Interorbital width	8.7	8.1	9.2	8.6	9.1	8.2	9.1
Caudal-peduncle depth	15.9	15.0	15.0	15.4	15.8	15.8	16.3
Caudal-peduncle length	13.5	14.6	12.3	14.4	13.8	15.6	16.9
Upper jaw length	8.7	9.2	9.2	9.1	8.8	9.8	9.4
Predorsal length	34.4	35.5	35.9	34.9	35.6	36.4	36.4
Preanal length	61.4	60.1	66.5	65.5	64.2	63.6	61.1
Prepelvic length	36.0	35.7	37.1	39.6	37.0	36.9	37.3
Length dorsal-fin base	63.1	63.5	62.5	64.3	60.6	60.3	62.7
Length anal-fin base	27.5	26.9	24.3	28.4	26.0	27.6	26.6
Length pectoral fin	26.4	26.1	27.9	25.9	27.0	26.4	27.9
Length pelvic fin	32.8	30.5	33.5	33.3	33.4	32.6	37.6
Length pelvic-fin spine	15.1	15.4	17.4	16.4	17.7	16.8	18.5
Length first dorsal spine	7.2	5.6	6.3	7.3	7.9	6.0	7.2
Length seventh dorsal spine	14.1	13.8	13.6	14.7	13.8	13.9	15.7
Length last dorsal spine	15.5	14.8	14.7	16.7	14.3	14.4	16.9
Length longest dorsal ray	23.8	23.4	21.7	22.1	22.7	20.4	22.6
Length first anal spine	7.6	6.9	7.4	7.7	6.9	7.5	7.5
Length second anal spine	18.1	15.9	17.0	17.6	16.9	18.0	18.2
Length longest anal ray	24.9	21.3	20.3	23.6	19.6	20.1	26.0
Length caudal fin	31.3	28.4	28.6	32.7	29.6	30.6	28.2
Caudal concavity	6.4	5.2	4.5	6.1	3.3	5.8	6.0

Colour in life. (Figs. 2 & 7A) Generally pale yellow to whitish with blue (young fish) or dark blue-grey (adult fish) region encompassing snout, upper head and adjacent anterodorsal portion of body and spinous dorsal fin; small blackish spot at uppermost rear edge of operculum near origin of lateral line; each scale of suborbital, preopercle, and opercle with small blue spot; each body scale with 1–2 small blue spots, generally darker on anterior half of body; young fish with scattered neon blue spots on dark blue forehead and snout; fins (except dark blue or greyish spinous dorsal) semitranslucent with pale yellow to whitish hue except for pair of submarginal blue stripes on spinous dorsal fin.

Colour in alcohol. (Fig. 1) Head and anterodorsal part of body dark grey, including spinous dorsal fin; remainder of body yellowish tan; remaining fins semi-translucent whitish or tan; a small blackish spot at uppermost rear edge of operculum.

Distribution and habitat. The new species is primarily restricted to the southwestern Pacific Ocean (Fig. 3) with confirmed records from Halmahera and West Papua, Indonesia, northeastern Papua New Guinea (including New Britain, Admiralty Islands, and Milne Bay Province), Solomon Islands, Vanuatu, New Caledonia, and Australia. Australian localities include the entire extent of the Great Barrier Reef. The habitat consists of rocky substrates, frequently close to shore, often in gullies just below the surge zone at depths of about 1–6 m. It generally occurs solitarily or in small, loose groups.

Etymology. The species is is named *caesifrons* (Latin: "bluish-grey forehead", *caesius* for blue-grey and *frons* for forehead or brow) with reference to its distinguishing colour pattern. It is a third declension compound adjective in the feminine nominative singular.

Genetic Analyses. We resolved relationships between *Chrysiptera caesifrons* and other *Chrysiptera* species using a 416 base pair segment of the mitochondrial control region gene from nine individuals of *C. caesifrons* and two individuals of *C. rex*, with all sampled individuals occuring sympatrically in the Raja Ampat Islands. One sequence each for *Chrysiptera talboti* and *C. taupou* from GenBank (EU256976.1 and EU256904.1, respectively; http://www.ncbi.nlm.nih.gov) were included in the analyses, originally from Drew *et al*. (2010).

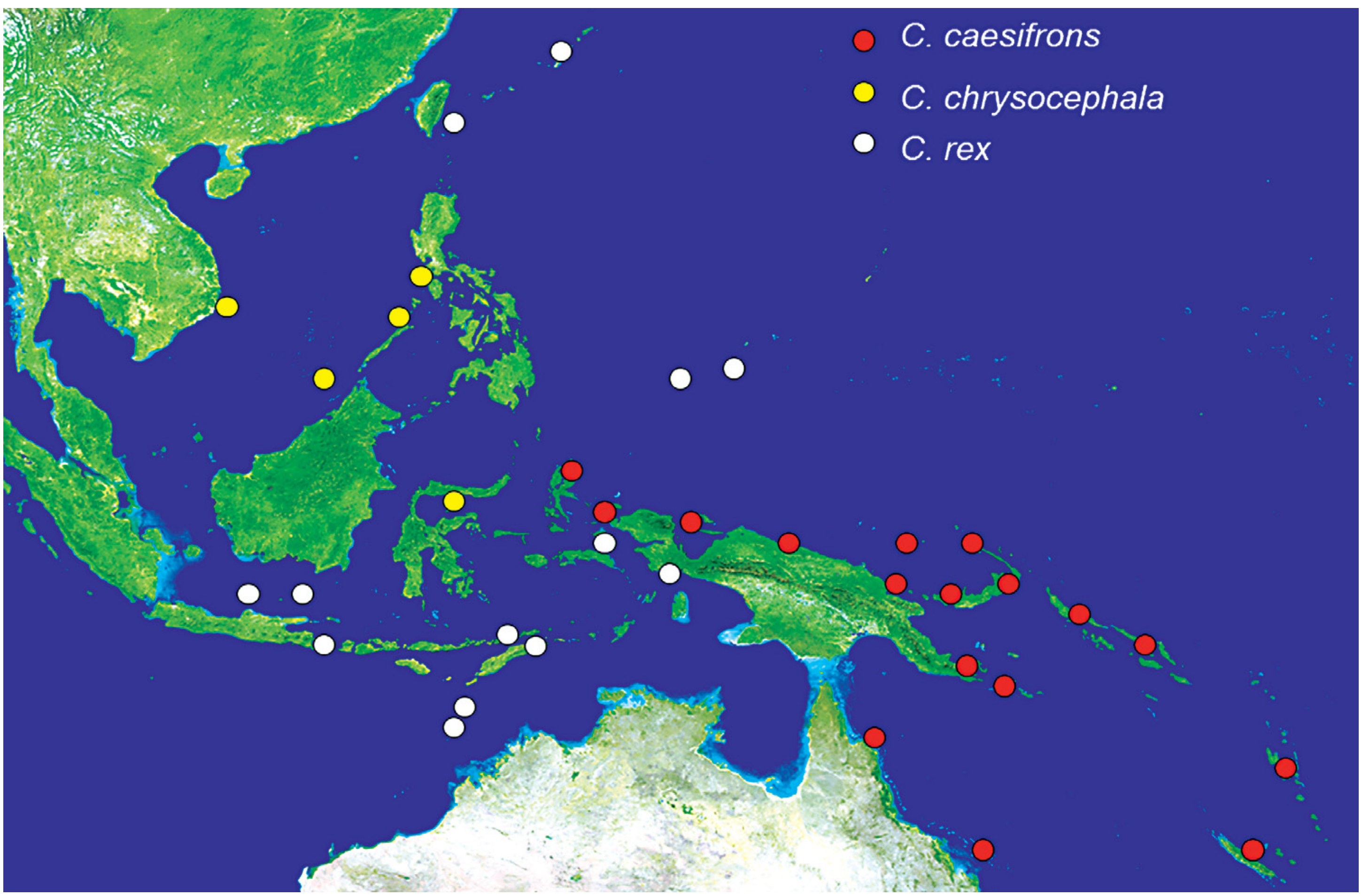

Figure 3. Distribution map showing locality records for members of the *Chrysiptera rex* species complex.

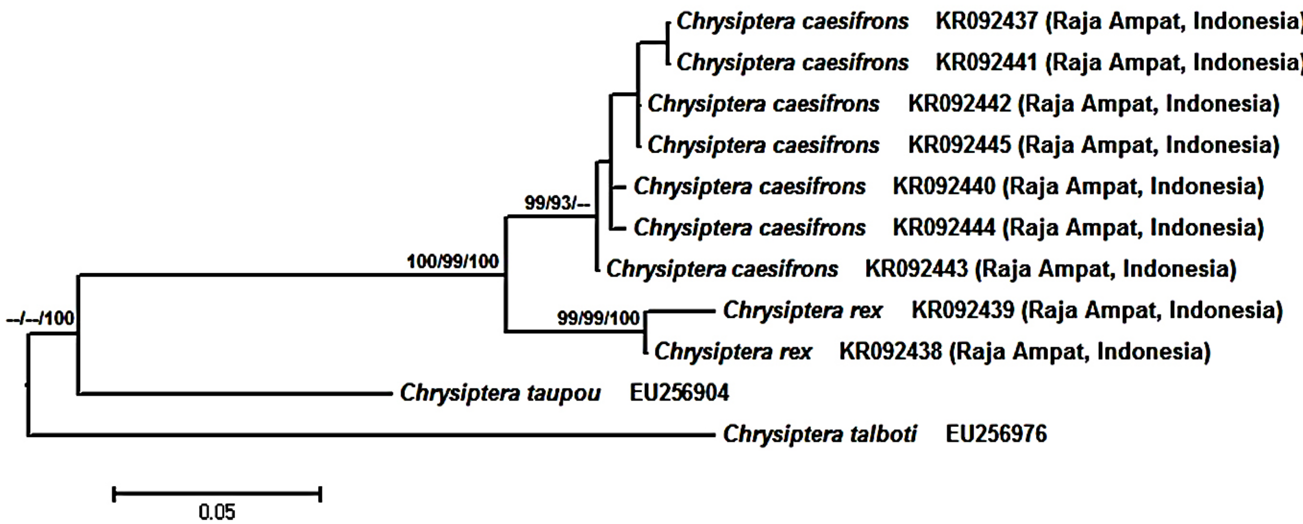

Figure 4. Maximum likelihood tree (HKY+G) of the mitochondrial control region data for four species of *Chrysiptera* (including *C. caesifrons*). Numbers above the major nodes indicate bootstrap support for 1000 replicates using neighbor joining, maximum likelihood, and Bayesian posterior probability, respectively. GenBank accession numbers and collection locality listed for samples.

In the total *Chrysiptera caesifrons* alignment there were 5 parsimony-informative characters. Nucleotide frequencies for the *C. caesifrons* samples were as follows: T = 33.7, C= 17.4, A=36.4, G=12.4.

Our results indicate that *C. rex* and *C. caesifrons* are sister species, forming a clade with 100% bootstrap support using neighbor joining and 99% bootstrap support using maximum likelihood analysis and 100% posterior probability using the Bayesian method (Fig. 4). Pairwise distances between the four species of *Chrysiptera* sequenced in this study ranged from 0.064-0.180, with the least distance between *C. rex* and *C. caesifrons* (Table 3). The average interspecific pairwise genetic distance between *C. caesifrons* and *C. rex* is 0.064, with an average intraspecific genetic distance for *C. caesifrons* of 0.010. Each of the trees generated clearly separate *C. caesifrons* from *C. rex*.

The barcode COI sequences from BOLD showed a similar pattern of relatedness. In this case, *C. chrysocephala* (Philippines) was sampled and is the nearest relative of *C. caesifrons* (Australia), with a minimum interspecific distance of 4.3% (0.043 K2P distance). *C. rex* (Philippines aquarium trade) is more distant from *C. caesifrons* (Australia), with a minimum interspecific distance of 7.19% (0.0719 K2P distance).

Comparisons. Although essentially morphologically identical to extralimital populations of *Chrysiptera caesifrons*, we have restricted the type series to specimens from the Raja Ampat Islands. Additional documentation of phenotypic differences along with genetic analyses of populations from other regions is required to confirm their taxonomic status. Comparative meristic data for the new species and its closest relatives, *C. chrysocephala* and *C. rex*, are provided in Tables 4 and 5.

TABLE 3

Average interspecific pairwise genetic distance matrix for mtDNA control region sequences

No.	Species	1	2	3	4
1	*Chrysiptera caesifrons*				
2	*Chrysiptera rex*	0.064			
3	*Chrysiptera talboti*	0.173	0.180		
4	*Chrysiptera taupou*	0.139	0.144	0.158	

Drew *et al*. (2010) provided evidence, based on their analysis of the mtDNA control region and cytochrome b, for the separation of *C. rex* into three species-level clades correlated with geographic populations from Japan (Ryukyu Islands, type locality of *C. rex*), Philippines, and Halmahera (Maluku Province, Indonesia). Although they illustrated very different colour patterns for each of the three populations, it was subsequently determined the photograph of *C. rex* from the Ryukyu Islands was erroneously identified and instead represents the juvenile stage of an undetermined species. The Philippines fish, characterised by a bright yellow snout, forehead and anterodorsal body region, was illustrated with an example from the South China Sea, which was subsequently identified as *C. chrysocephala* (Fig. 5), first described from Layang Layang Atoll, South China Sea. The Halmahera fish agree with our present description of *C. caesifrons*.

TABLE 4

Frequency distribution of of soft dorsal and anal fin-ray counts
for members of the *Chrysiptera rex* species complex

Species (location)	Soft dorsal rays				Soft anal rays		
	12	13	14		12	13	14
C. caesifrons (W. Papua)	4	17				20	1
*C. caesifrons (*Papua New Guinea)							
C. caesifrons (Queensland)	4	41	6		1	40	10
C. caesifrons (Vanuatu)	1	8				9	
C. chrysocephala (S. China Sea)	9	29			4	34	
C. rex (Ryukyu Is.)	2	6				8	
C. rex (Taiwan)		4				3	1
C. rex (Indonesia)	1	27	2			27	1
C. rex (W. Australia)	2	6			4	4	

TABLE 5

Frequency distribution of pectoral-fin ray and tubed lateral-line scale counts for members of the *Chrysiptera rex* species complex. Pectoral rays counted on both sides of fish.

Species (location)	Pectoral ray counts					Lateral-line scales			
	15	16	17	18		15	16	17	18
C. caesifrons (W. Papua)			38	4			3	13	6
*C. caesifrons (*Papua New Guinea)		1	15					7	1
C. caesifrons (Queensland)		12	81	13			4	39	8
C. caesifrons (Vanuatu)			11	7				6	3
C. chrysocephala (S. China Sea)		36	41				8	25	5
C. rex (Ryukyu Is.)		2	16	2		2	2	5	1
C. rex (Taiwan)		3	5				1	2	1
C. rex (Indonesia)	2	39	35			2	11	23	2
C. rex (W. Australia)		11	5				4	4	

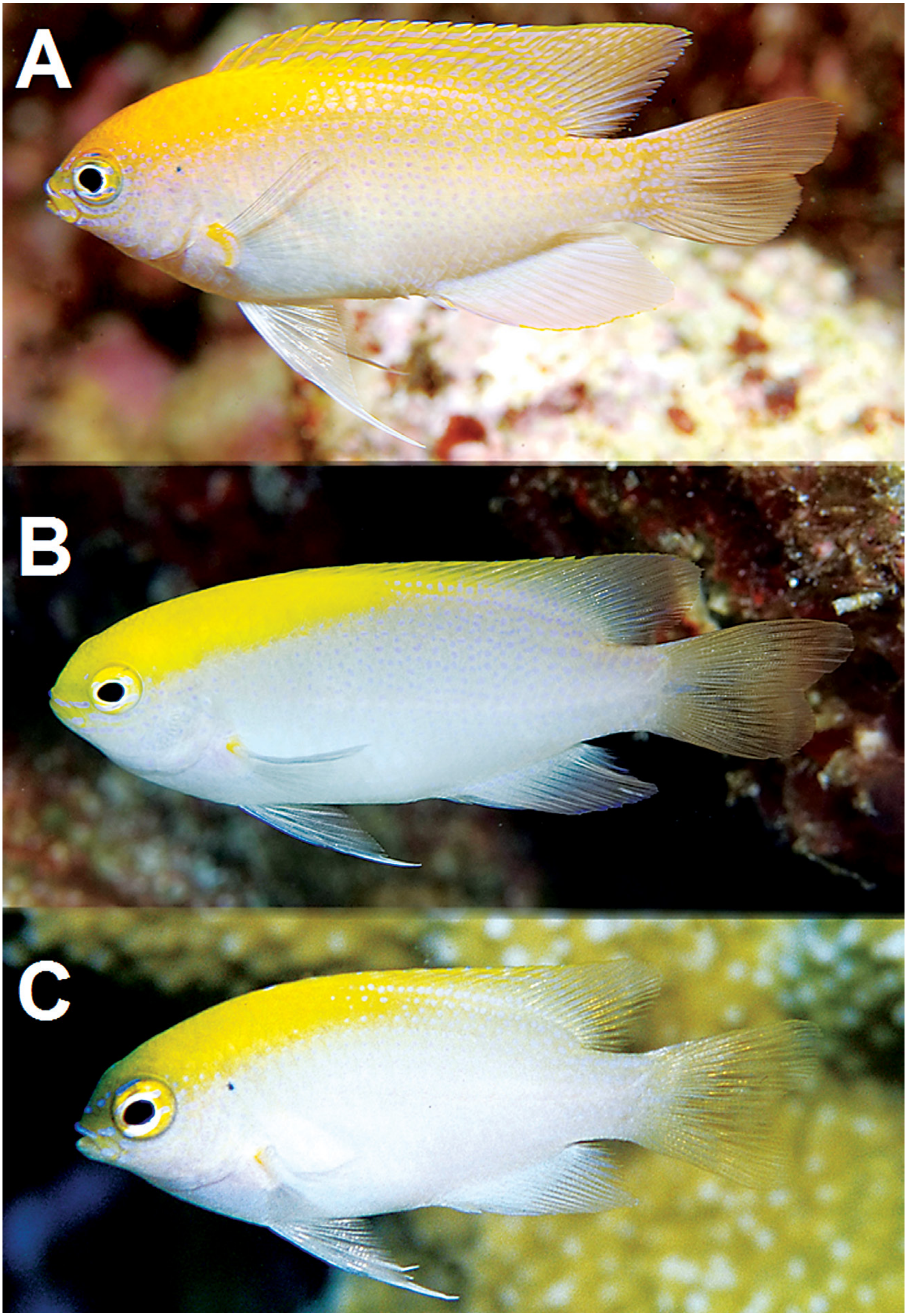

Figure 5. *Chrysiptera chrysocephala*, underwater photographs. A. El Nido, Palawan, Philippines, approximately 50 mm SL; B. Layang Layang Atoll, Spratly Islands, Malaysia, approximately 50 mm SL; C. Togean Islands, Sulawesi, Indonesia, approximately 40 mm SL (G.R. Allen).

The current study confirms that the three populations recognized by Drew *et al*. (2010) are indeed three valid species in the *C. rex* complex. Moreover, our mtDNA comparisons of sympatric populations of *C. caesifrons* and *C. rex* from the Raja Ampat Islands of West Papua, Indonesia adds additional confirmation of the genetic separation of *C. caesifrons* and *C. rex* (Fig. 4). It has been noted that finding genetic differences between distant populations may not establish that gene flow is restricted, since intermediate populations may show some genetic continuity– thus it is critical to compare sympatric populations when possible (Victor 2015). Although wider sampling and more genetic analyses are required to confirm the geographic boundaries of the members of the *C. rex* complex, our present understanding of their ranges is presented in Fig. 3. The distribution of *C. rex* includes the Ryukyu Islands, Taiwan, Palau, eastern Indonesia (eastern Kalimantan and Java eastward), Timor Leste, Raja Ampat Islands (off western extremity of New Guinea), and offshore reefs of Western Australia including Scott, Cartier, and Ashmore. The range of *C. chrysocephala* is mainly restricted to the South China Sea with records from the Philippines, Vietnam, and Malaysia (Spratly Islands). There is also a single photographic record (Fig. 5C) from outside this region at the Togean Islands in Tomini Bay (Central Sulawesi, Indonesia). This significant disjunction warrants future genetic investigation of the Sulawesi population. *Chrysiptera caesifrons* is known from Indonesia (Halmahera and Raja Ampat Islands), Papua New Guinea, Solomon Islands, Vanuatu, New Caledonia, and Great Barrier Reef (Queensland Australia). Illustrations of this species from the Great Barrier Reef and Halmahera appeared in Randall *et al*. (1997; as *C. rex*) and Allen & Erdmann (2012; as *Chrysiptera* species), respectively.

Chrysiptera chrysocephala (Fig. 5) is readily distinguished from the other species by an extensive yellow zone on the head and upper anterior body. It also has a yellow/orange pectoral-fin base and axil, a feature shared with *C. rex* (Fig. 6), but absent in *C. caesifrons* (Fig. 2). The salient colour pattern differences of the three closely related species are presented in Table 6. The juvenile pattern of *C. caesifrons* is also distinctive and clearly separates this species from similar sized *C. chrysocephala* and *C. rex* (Fig. 7). Most noticeably, the upper head, anterodorsal body, and adjacent anterior half of the spinous dorsal fin are dark blue in contrast to the yellow dorsal fin of the other species, which also lack the pronounced dark blue on the head and anterior body.

TABLE 6

Live colour pattern differences of *Chrysiptera* species

Pectoral base includes axil of fin and head marks refers to the blue markings

Species	**Forehead**	**Upper back**	**Pectoral base**	**D. spine tips**	**Head marks**
C. caesifrons	nearly black	nearly black	whitish	dark blue	inconspicuous
C. chrysocephala	yellow	bright yellow	yellow/orange	yellowish	inconspicuous
C. rex	blue-grey	pale yellow	yellow/orange	yellowish	vivid

Other material examined. *Chrysiptera chrysocephala*: BPBM 40164, 4 specimens, 20.7–45.2 mm SL, Hon Mot Island, 12° 10' 30.8" N, 109° 16' 40.2"E, Nha Trang Bay, Vietnam; CAS 222286, 5 specimens, 24.4–38.0 mm SL, same data as BPBM 40164; IPMB 33.17.01, 41.6 mm SL, Layang Layang Atoll, 7° 21'N, 117° 52'E, Malaysia, South China Sea; IPMB 33.17.02, 49.6 mm SL, collected with previous IPMB specimen; ROM 77193, 3 specimens, 23.2–39.2 mm SL, Hon Mun Island, 12° 10' 12.1"N, 109° 18' 27.1"E, Nha Trang Bay, Vietnam; ROM 73803, 5 specimens, 26.3–45.8 mm SL, Hon Noc Island, 12° 11' 29.3"N, 109° 20' 31.8"E, Nha Trang Bay, Vietnam; ROM 77194, 2 specimens, 33.6–36.9 mm SL, Lon Island, 12° 11' 16.2"N, 109° 17' 32.4"E, Nha Trang Bay, Vietnam; ROM 77195, 6 specimens, 28.1–49.7 mm SL, Hon Mun Island, 12° 10' 5.8"N, 108° 18' 43.8"E, Nha Trang Bay, Vietnam; ROM 77196, 26.2 mm SL, Mua Island, 12° 10' 15.5"N, 109° 18' 42.8"E, Nha Trang Bay, Vietnam; ROM 77197, 2 specimens, 21.4–27.6 mm SL, Hon Mot Island, 12° 10' 33.3"N, 109° 16' 45.4"E, Nha Trang Bay, Vietnam; ROM 77198, 2 specimens, 40.8–47.1 mm SL, Hon Mot Island, 12° 10' 37.4"N, 109° 16' 43.5"E, Nha Trang Bay, Vietnam; USNM 383522, 4 specimens, 36.0–46.8 mm SL, Hon Mun

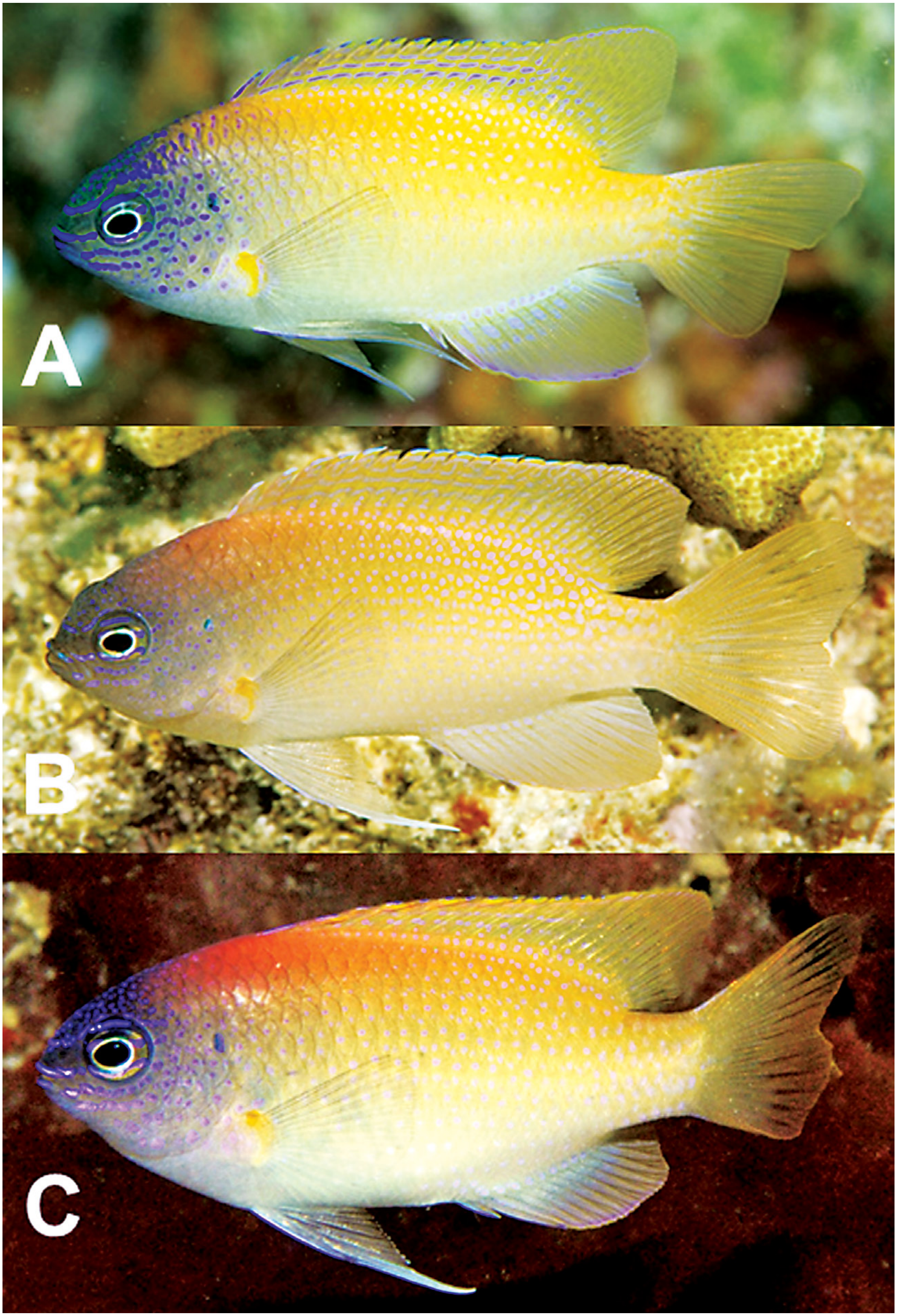

Figure 6. *Chrysiptera rex*, underwater photographs. A. Ishigaki Island, Ryukyu Islands, approximately 40 mm SL (K. Sodo); B. East Timor, 45 mm SL (G.R. Allen); C. Misool, Raja Ampat Islands, West Papua Province, Indonesia (G.R. Allen).

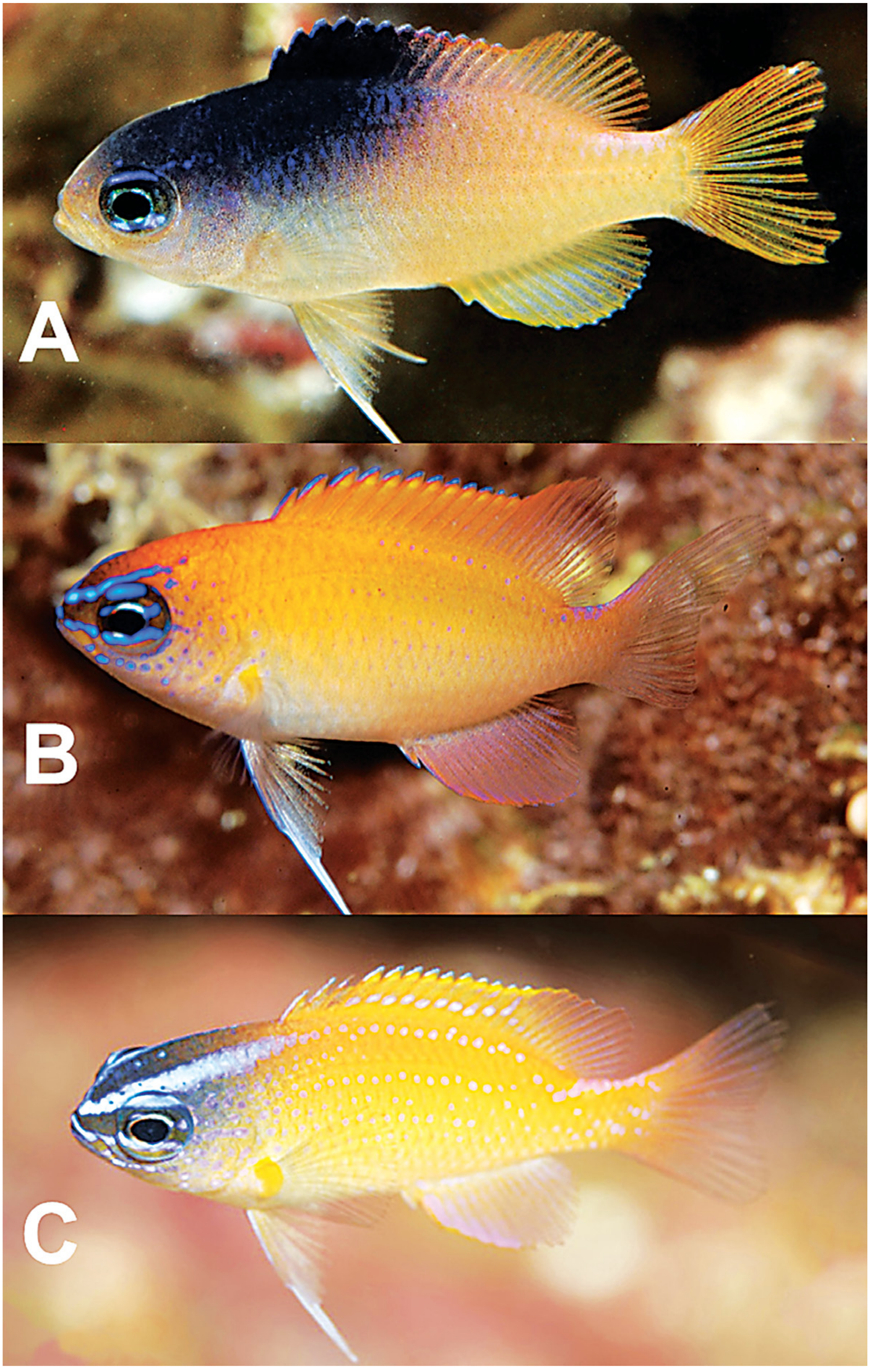

Figure 7. Underwater photographs of small juveniles, approximately 15-20 mm SL: A. *C. caesifrons*, Ayau Atoll, Raja Ampat Islands, West Papua, Indonesia (G.R. Allen); B. *C. chrysocephala*, Palawan, Philippines (G.R. Allen); C. *C. rex*, Ishigaki, Ryukyu Islands, Japan (K. Sodo).

Island, 12° 10' 18.3"N, 109° 18' 02.6"E, Nha Trang Bay, Vietnam; WAM P. 32756–001, 3 specimens, 40.7–44.0 mm SL, collected with IPBM specimens; WAM P.33153–002, 2 specimens, 38.0–40.2 mm SL, Basura Beach, approximately 13° 45'N, 120° 55'E, Anilao, Luzon, Philippines.
Chrysiptera rex: AMS I.21315–033, 5 specimens, 17.4–40.2 mm SL, Scott Reef, 14° 06'S, 123° 01'E, Western Australia; AMS I.26749–005, 33.4 mm SL, Ashmore Reef, 12° 17'S, 121° 56'E, Timor Sea; USNM 62949 (holotype), 46.0 mm SL, Naha, Okinawa; USNM 74535, 11 specimens (paratypes), 26.0–41.0 mm SL, Naha, Okinawa; USNM 215891, 27 specimens, 43.9–29.7 mm SL, Karimundjawa, Java Sea, Indonesia; USNM 275033, 28.8 mm SL, Okinawa, Ryukyu Islands; USNM 275032, 2 specimens, 28.5–36.2 mm SL, Okinawa, Ryukyu Islands; USNM 352638, 8 specimens, 40.0–49.9 mm SL, Ishigaki-Shima, Ryukyu Islands; WAM P.29040–007, 29.9 mm SL, Ashmore Reef, Timor Sea; WAM P.29053-004, 45.3mm SL, Ashmore Reef, Timor Sea; WAM P.30293–011, 2 specimens, 34.4–40.3 mm SL, Scott Reef, Western Australia; WAM P.30330–001, 3 specimens, 26.1–43.6 mm SL, Scott Reef, Western Australia; WAM P.30333–022, 35.0 mm SL, Scott Reef, Western Australia; WAM P.32807–001, 36.0 mm SL, Pulau Lauzaro, Triton Bay, West Papua, Indonesia; WAM P. 33743–003, 17 specimens, 29.4–43.2 mm SL, Com, 08° 21.301'S, 127° 03.631'E, Timor Leste.

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