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The genus *Polycera* Cuvier, 1817 (Nudibranchia: Polyceridae) in the eastern
Pacific Ocean, with redescription of *Polycera alabe* Collier & Farmer, 1964
and description of a new species

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ABSTRACT

The nudibranch *Polycera alabe* Collier & Farmer, 1964 has been reported to be extremely variable in coloration. Some of this variation appears to be correlated with geographic range, with four regional colour forms having been recognized. However, the identity of *P. alabe* has been unclear because of its vague original description, based on only two specimens from Baja California, Mexico, without any morphological or molecular study to date. Here we present a complete redescription, based on examination of the holotype and paratype, as well as study of new material from the type locality and vicinity. The description includes colour pictures, drawings of the reproductive system and electron micrographs of the radula and penis. New findings are reported, such as the presence of two well-developed cusps in the outer lateral teeth, a pair of digitiform and rather short salivary glands, and a penis with at least two different types of penial spines. We describe the reproductive system for the first time. Specimens similar to *P. alabe* in the external coloration, but with several external and internal differences such as the type of salivary glands and number of outermost teeth, are described as a new species, *P. anae* n. sp. from Panama and Costa Rica. These two species are compared with five other *Polycera* species known from the eastern Pacific.

INTRODUCTION

The number of described species in the genus *Polycera* Cuvier, 1817 has rapidly increased in recent years. To date, it includes 32 species (Bouchet & Gofas, 2013) with many more still to be described (Valdés *et al.*, 2006; Burn, 2006; Debelius & Kuitert, 2007; Gosliner, Behrens & Valdés, 2008). *Polycera* species are distributed worldwide, with species known from the Indo-Pacific (Debelius & Kuitert, 2007; Coleman, 2008; Gosliner *et al.*, 2008), Western Atlantic (Ortea, Espinosa & Camacho, 1999; Valdés *et al.*, 2006; Debelius & Kuitert, 2007), Eastern Atlantic (Thompson & Brown, 1976; Zsilavec, 2007; Calado & Silva, 2012) and Eastern Pacific (Behrens, 1991; Schrödl, 2003, 2009; Behrens & Hermosillo, 2005). The aposematic colouration of the type species *P. quadrilineata* (O. F. Müller, 1776) has been examined by Tullrot & Sundberg (1991), and the spawning and embryonic development of this species and of *P. aurantiomarginata*

García-Gómez & Bobo, 1984 by Martínez-Pita, Sánchez-España & García (2006). Ongoing research has greatly increased knowledge about the distribution of the potentially invasive *P. hedgbeithi* Er. Marcus, 1964, originally described from California, but also found in the Caribbean, South Africa, South West Africa, the Mediterranean, the Atlantic coast of the Iberian Peninsula, Australia, New Zealand and Japan (e.g. Gosliner, 1982; Cervera *et al.*, 1988, 2010; Keiu, 2000; Miller, 2001; Caballer & Ortea, 2002; Wilson, 2006; Keppel, Sigovini & Tagliapietra, 2012; Giacobbe & De Matteo, 2013).

Palomar, Pola & García-Vázquez (2014) presented the first molecular phylogeny of the subfamily Polycerinae based on two mitochondrial genes (COI and 16S). This study only included seven *Polycera* species and most of the phylogenetic relationships were not resolved. No monographic morphological or molecular study of *Polycera* has verified the number of valid species. Many species were first described between the mid-19th and mid-20th

centuries and the original descriptions were often poor and incomplete. This was the case for *P. capensis* Quoy & Gaimard, 1824 and *P. alabe* Collier & Farmer, 1964, among others. *Polycera alabe* was described using two specimens; the holotype was collected on the Pacific side of Baja California and the paratype in the Gulf of California (Sea of Cortez). The original description of this species included information on the external morphology, yet few details about internal anatomy were presented. The only described aspects of the internal anatomy were the radular formula and the shape of the teeth of the holotype. A general description of the reproductive system was missing, although it was mentioned that the penis was armed. Almost 40 years later, Schrödl (2003) presented some anatomical data on two specimens identified as *P. alabe* from northern Chile. This author briefly described the external anatomy, the shape of the jaws, the radular formula and the shape of the teeth, but the reproductive system remained unknown.

Over the years, specimens with similar colour pattern and external features to *P. alabe* have been photographically documented in several field guides and webpages (Kerstitch, 1989: 62, fig. 142; Schrödl, 2003: 104, fig. 19; Behrens & Hermosillo, 2005: 62; Camacho-García, Gosliner & Valdés, 2005: 76, upper photo; Hermosillo, Behrens, Ríos-Jara, 2006: 68; Debelius & Kuiter, 2007: 37, bottom pictures; <http://slugsite.us/bow/nudwk493.htm>; http://www.medslugs.de/E/Pac-E/Polycera_alabe.htm; <http://www.inbio.ac.cr/papers/babosamarinas/images/alabe.gif>; among others), which has resulted in considerable confusion. Kerstitch (1989: 62) remarked that “there are a number of opisthobranchs in the Gulf that have dark blue (or brownish) bodies covered with yellow orange spots, but most species tend not to co-occur, due to rather different ecological preferences”.

Behrens & Hermosillo (2005) recognized four distinct colour forms of *P. alabe*. These authors suggested the possible existence of several species based on the external appearance and geographical range. Most recently, Santander & Valdés (2013) published a molecular study of the phylogenetic relationships and genetic structure of *P. alabe* and *P. atra*, based on nuclear (H3) and mitochondrial (16S) genes. However, the authors made no detailed morphological study of the original specimens of Collier & Farmer (1964) in order to confirm the identity of *P. alabe*. Therefore, although Santander & Valdés (2013) found several different morphological and molecular forms within *P. alabe*, they were unable to resolve their taxonomy.

In addition to *P. alabe*, five other species of *Polycera* are known to be present in the eastern Pacific Ocean: *P. atra* MacFarland, 1905, *P. hedgpethi* Marcus, 1964, *P. tricolor* Robilliard, 1971, *P. priva* Er. Marcus, 1959 and *P. kaiseriae* Hermosillo & Valdés, 2007. In addition a small undescribed species has been reported from Isla Revillagigedo (Mexico), Costa Rica, Panama and the Galápagos (*Polycera* sp. of Camacho-García et al., 2005).

In the present contribution *P. alabe* is redescribed based on the holotype, paratype and five additional specimens from the type locality and vicinity. The ‘*Polycera* sp.’ of Camacho-García et al. (2005) is described as *P. anae* n. sp., based upon external, radular and genital differences. Finally, a comparative table based on the available literature summarizes the most important features of the six species known from the eastern Pacific.

MATERIAL AND METHODS

The material examined for this study (Table 1 Fig. 1) is deposited in the Department of Invertebrate Zoology and Geology of the California Academy of Sciences, San Francisco (CASIZ) and the Museo de Zoología de la Universidad de Costa Rica (MZUCR). The anatomical study was performed by first making a dorsal incision during dissection. Internal features were examined using a dissecting microscope with a camera lucida for drawing anatomical details. The buccal mass was

removed and dissolved in 10% sodium hydroxide until the labial cuticle and radula could be isolated from the surrounding tissue. The labial cuticle and radula were then rinsed in water, dried and mounted for examination by a Hitachi S-3000N scanning electron microscope (SEM). Special attention was paid to the morphology of the reproductive system, including the penial spines. The external morphology of the holotype, and its radula and jaws (which were extracted and mounted by Collier & Farmer, 1964), as well as serial sections of the entire paratype, were examined under a dissecting microscope.

Nomenclatural acts

This published work and the nomenclatural acts it contains have been registered in ZooBank (<http://zoobank.org/>). The Life Science Identifier (LSID) for this publication is: urn:lsid:zoobank.org:pub:7CD6704F-8CF5-4F1A-9B17-009061DAE953.

SYSTEMATIC DESCRIPTIONS

Family Polyceridae Alder & Hancock, 1845 Genus *Polycera* Cuvier, 1817

Type species: *Doris quadrilineata* Müller, 1776 by subsequent designation (Gray, 1847).

Diagnosis: Body limaciform, highest at middle; notum smooth, partially or entirely papillate or tuberculate. Anterior margin of head expanded, forming frontal veil bearing velar processes. Oral tentacles short, lobate; perfoliate rhinophores nonretractile; extra-branchial appendages present or absent. Pinnate gill in semicircle surrounding anus; gill not retractable into gill pocket. Paired jaws with wing-like processes. Radular formula n.2.0.2.n; inner laterals hamate, second lateral larger than first; outer laterals small, simple plates. Large prostate gland; penis armed with spines. (Compiled from Miller, 1996; Hermosillo & Valdés, 2007).

Polycera alabe Collier & Farmer, 1964 (Figs 2–5)

Material examined: Holotype: 25 mm long alive, SE side Isla de Cedros, Baja California, Mexico, coll. J. Sloan, 27 Jan. 1963, slide of radula and jaws (CASIZ 18190). Paratype: Isla Angel de la Guarda, Puerto Refugio, Gulf of California, Mexico (29°33'00"N, 113°34'60"W), coll. J. Sloan, 30 March 1963; serial sections on 11 slides (CASIZ 18373). Additional material: 2 adults (11, 12 mm preserved), NE corner of Isla de Cedros, Baja California, Mexico, 19 Aug. 1987, 12 m, dissected (CASIZ 72119); 1 adult (12 mm preserved), Bahía de los Angeles, Isla Coronado, Baja California Sur, Mexico, 4 Oct. 1984, 6–9 m, dissected (CASIZ 57308); 1 adult (19 mm preserved), Punta Gringa, Baja California, Gulf of California, Mexico (29°02'21"N, 113°32'15"W), 27 Feb. 1989 (CASIZ 66930); 1 adult (20 mm preserved), Los Morros, Bahía Tortugas, Baja California Sur, Mexico, 1 July 1984, 3–4.5 m, dissected (CASIZ 71535).

External morphology (Fig. 2A–D): Length to 25 mm living. Body long, limaciform, widest and highest at level of gill circling. Notal margin a low ridge enlarging anteriorly as flange curving around head (oral veil); four velar processes. Foot long, narrow, slightly expanded anteriorly, transversely grooved. Body surface smooth, with 20 soft conical projections, nearly equal in size. Veil processes and notal margin projections joined by subtle ridge. Tail projections 11. Rhinophores with 11–15 lamellae, without rhinophoral sheath. Oral region rounded, edge of mouth folded, tentacles ear-like. Gill of up to 8 unipinnate branches, smaller in posterior part of gill circling, surrounding

Table 1. Comparison of the Eastern Pacific species of the genus *Polycera*.

	<i>P. atra</i> MacFarland, 1905	<i>P. priva</i> Er. Marcus, 1959	<i>P. alabe</i> Collier & Farmer, 1964	<i>P. hedgpethi</i> Er. Marcus, 1964	<i>P. tricolor</i> Robilliard, 1971	<i>P. kaiseræ</i> Hermosillo & Valdés, 2007	<i>P. anae</i> n. sp.
Colour	Pale grey with narrow or broader lines of black on dorsum and sides. Orange spots within lines and outlining margin of foot	Brownish translucent, with opaque white spots irregularly distributed over body. Slightly prominent mediodorsal opaque white keel running from posterior of gills to tip of tail. Gills tipped with opaque white pigment. Lamellae brownish tipped with opaque white	Blue-black, covered with orange spots arranged more or less in rows posterior to rhinophores. Black gills and rhinophores with orange marks. Velar processes black, white or both. Edge of foot white. Colourless projections on body and black ones on tail	Grey with small black dots. Yellow-orange marks on rhinophores, foot corners and oral and extrabranchial processes. Streaks of same colour on caudal crest, pallial ridge and upper border of foot. Yellow orange spots on tubercles all over body	Translucent grey-white, yellow line around foot margin, on caudal crest and pallial ridge. Row of yellow tubercles on side and centre of notum. Lamellae with white tips and median black line	Pink sprinkled with white spots on dorsum, cephalic region, rhinophoral sheaths and gill plumes. Processes same colour as body; distal half navy blue with white or of-white tip	Black mottled with several rows of interrupted orange lines or dots along body. Rhinophores, tubercles, papillae, extrabranchial processes, oral veil, gill and foot translucent white. Rhinophoral sheath black. Area above eyes translucent
Rhinophores	8–12 lamellae; stout	25 lamellae approx	11 lamellae	12 lamellae; slender	13–20 lamellae	22 lamellae	7–8 lamellae
Gill	8–11; unipinnate	5; bi- to tripinnate	6; unipinnate; 4 anterior higher than posterior ones	9; tripinnate	5–6; bi- and tripinnate	5; unipinnate; 3 anterior larger than posterior ones	5–6; decreasing in size posteriorly
Oral tentacles	Short lobes	Not described	Not described	Very short	Not described	Not described	Short and conical, dorsally grooved
Foot	Linear; prominent anterior corners	Broad, with expanded fronto-lateral corners	Slightly expanded anteriorly	Narrow sole, pointed tail	Relatively narrow	Simple, with pointed pedal corners	Narrow, linear, small anterior corners
Oral veil	4 slender, tapering, pointed processes	6–8 digitiform processes	4 processes	2–3 digitiform processes on either side	8–11 cylindrical pointed processes	6–8 conical processes of unequal lengths	4 digitiform processes, equally spaced
Extrabranchial processes	2 on each side; low compressed and pointed	4–6 on each side; large, inflated, translucent papillae	Absent	3 on each side, increasing in size posteriorly	4–6 on each side, slender and cylindrical	2; digitiform	Absent
Radula	9–11 × 3.2.0.2.3. Two pleural hamate teeth, first smaller than second; second bearing wing-like expansion. Marginals uncinat, triangular, decreasing outwards	18–24 × 3.2.0.2.3. First lateral elongate, with blunt tip; basal denticle poorly developed. Second lateral slightly larger, tip pointed or with low, blunt, subapical denticles; base broad, with well-developed denticle(s). Marginals small, irregular, elongate rectangular plates	9–10 × 3–4.2.0.2.4–3. Laterals with strong denticle at distal end. Second lateral slightly larger than first. Marginals simple, decreasing in size outwards	17 × 3–4.2.0.2.3–4. Innermost of outer teeth with rudimentary cusp	9–16 × 3.2.0.2.3. Innermost laterals hamate with spur near base. Second laterals hamate without prominent spur. Rectangular marginals	11 × 2.1.1.0.1.1.2. Innermost laterals hamate with strong, triangular cusp and smaller cusp at mid-length. Second lateral hamate with wing-like expansion near base. Marginals simple	29–36 × 6–8.2.0.2.6–8. Laterals hamate. Second lateral with triangular distal cusp and second quadrangular cusp at mid-length. Inner marginals with prominent curved spur on anterior end. Outer marginals simple plates

Continued

Table 1. Continued

	<i>P. atra</i> MacFarland, 1905	<i>P. priva</i> Er. Marcus, 1959	<i>P. alabe</i> Collier & Farmer, 1964	<i>P. hedgpethi</i> Er. Marcus, 1964	<i>P. tricolor</i> Robilliard, 1971	<i>P. kaiseræ</i> Hermosillo & Valdés, 2007	<i>P. anae</i> n. sp.
Reproductive system	Prostate gland very large. Penis armed with curved hooks	Ampulla sausage-shaped. Broad, flattened prostate surrounding very large, spherical bursa copulatrix. Penis armed with two kinds of penial spines. Vagina slender. Receptaculum seminis small, pear-shaped	Penis armed with two different kinds of penial spines	Ampulla long, wide. Prostate followed by a fold. Penis with bristle-like spines	Large prostate, folded over itself. Penis armed with elongated spines	Vagina and deferent duct same shape. Bursa copulatrix large, oval. Ampulla long, regularly shaped. Penis armed with hamate spines	Reniform ampulla. Prostate rounded, flattened, folded over itself, covering the large bursa copulatrix. Penis armed with two kinds of penial spines
Geographical range	Oregon to Baja California (Mexico)	S Chile	Baja California, Gulf of California (Mexico)	California, New Zealand, Australia, Japan, South Africa, Italy, Angola, Morocco	British Columbia (Canada) to Baja California (Mexico)	Bahía de Banderas (Mexico)	Costa Rica, Panama, Mexico
References	MacFarland (1905, 1966), Behrens (1991)	Er. Marcus (1959), Schrödl (1996, 2003, 2009), Schrödl et al. (2005)	Collier & Farmer (1964), present study	Marcus (1964), Cenvera et al., (1988, 2010), Miller (2001).	Robilliard (1971), McDonald (1983)	Hermosillo & Valdés (2007)	Camacho-García et al. (2005), Santander & Valdés, (2013), present study

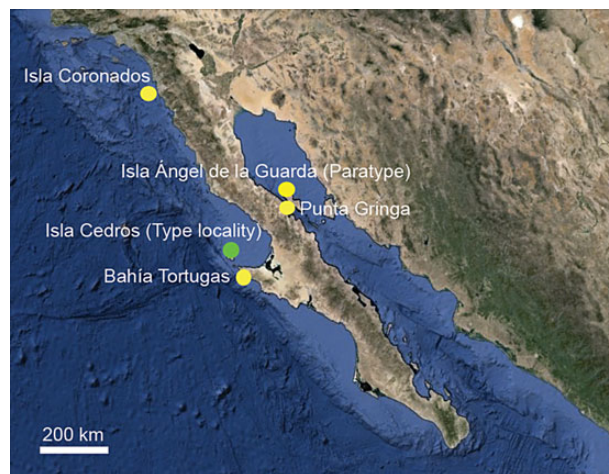


Figure 1. Map of Baja California (Mexico) showing collecting sites of type specimens of *Polycera alabe* and for the present study.

anal papilla. Genital opening on right side, closer to rhinophores than gill. Body colour blue-black with orange spots on dorsum, arranged more or less in rows posterior to rhinophores; spots absent on foot. Oral veil processes black, or translucent white, or they vary (e.g. two laterals translucent white and two centrals black in holotype, Fig. 2A, B). Body projections same colour as veil processes. Small blue-black rounded papillae, sometimes co-occurring with orange spots. Rhinophores and gills blue-black, entirely covered with orange spots (Fig. 2A–D). Area above eyes translucent. Foot edge white.

Internal anatomy: Oral tube soft, elongate; buccal bulb muscular, two times longer than oral tube. Radula (Fig. 3A, B): formulae $9 \times 3.2.0.2.3$ (holotype), $9 \times 4.2.0.2.4$ (CASIZ 71535), $10 \times 4.2.0.2.4$ (CASIZ 72119, 12 mm), $10 \times 3.2.0.2.3$ (CASIZ 72119, 11 mm), $12 \times 3.2.0.2.3$ (CASIZ 57308), $13 \times 3.2.0.2.3$ (CASIZ 66930); no rachidian teeth (Fig. 3A); lateral teeth elongate, bicuspid, square base; inner lateral teeth narrower and smaller than outer laterals; marginal teeth plate-like, much smaller than laterals, decreasing in size towards margin; inner marginals with slightly curved cusps, slightly prominent spur; fourth marginal (if present) very small, not visible in all rows. Labial cuticle with signs of individual jaw elements. Pair of short, digitiform salivary glands attached at sides of oesophageal connection. Blood gland well-developed, beneath intestinal loop. Reproductive system (Figs 3C, D, 4A): triaulic; hermaphroditic duct elongate, narrow; ampulla kidney-shaped; postampullary duct bifurcating into short oviduct leading into female gland and vas deferens with prostatic portion; vas deferens folded before ending penis; prostate broad, large, partially enclosing large bursa copulatrix, prostate slightly bigger than bursa copulatrix; penis armed with many long slender spines distally, shorter hook-shaped spines proximally (Fig. 3C, D); vagina a long, slender tube; receptaculum seminis small, pear-shaped on thin stalk, semi-serially arranged; small uterine duct connecting vagina to female gland.

Distribution: Although this species has been recorded from Mexico (Collier & Farmer, 1964; Kerstitch, 1989; Behrens, 1991; Hermosillo et al., 2006; Bertsch, 2010; present study), southern California (Behrens, 2004; Behrens & Hermosillo, 2005; Hermosillo et al., 2006), Costa Rica (Instituto Nacional De Biodiversidad, 2011; Hermosillo et al., 2006 as ‘variations B to D’), Panama (Behrens & Hermosillo, 2005; Hermosillo et al., 2006 as ‘variation C’) and northern Chile (Schrödl, 1996, 2003) we only confirm here its presence in Mexico and southern

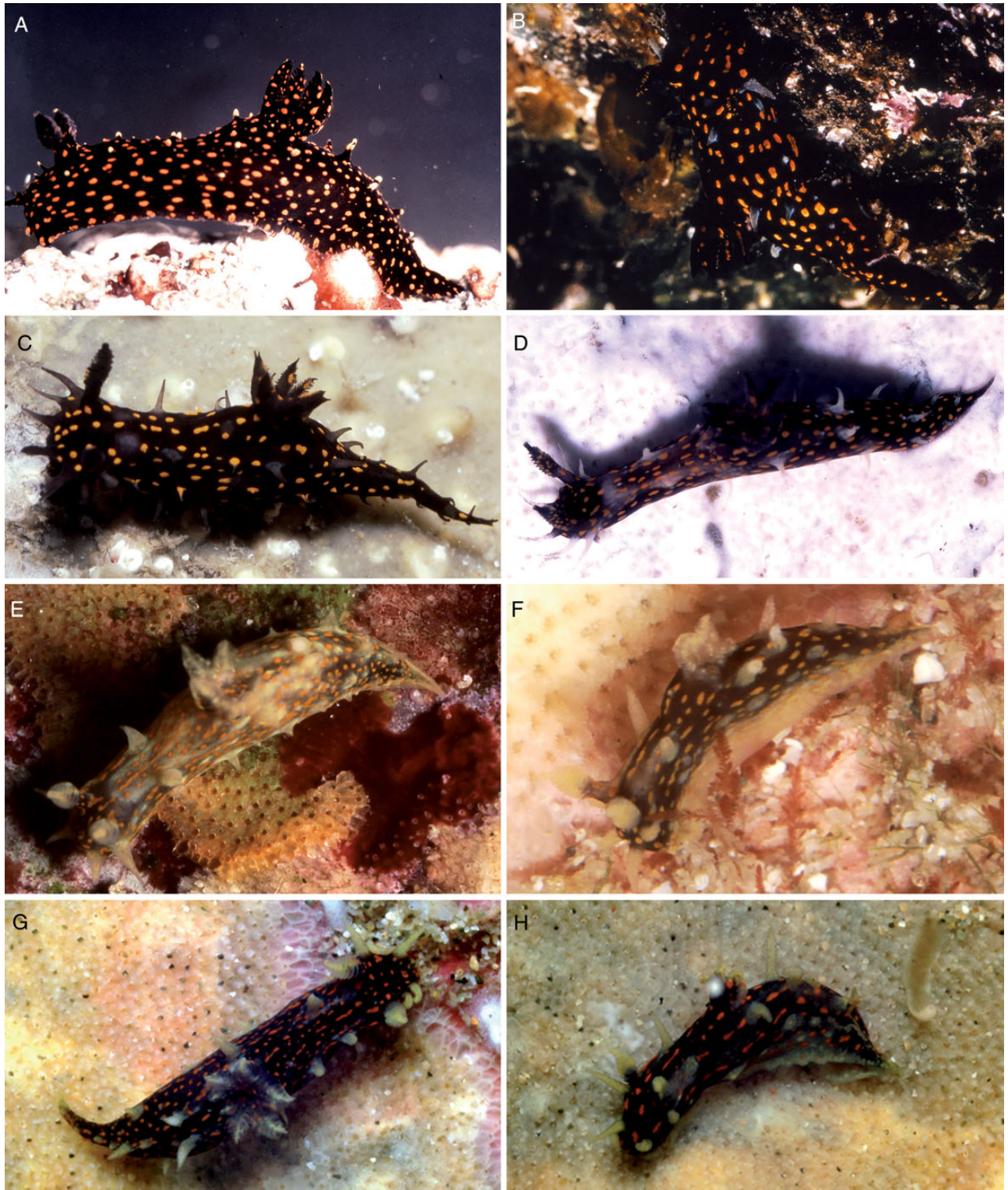


Figure 2. Photographs of living animals. **A–D.** *Polycera alabe* Collier & Farmer, 1964, Baja California, Mexico. **A, B.** CASIZ 18190 (holotype, 25 mm alive, photo by Collier & Farmer). **C.** CASIZ 71535 (20 mm preserved, photo by T.M. Gosliner). **D.** CASIZ 72119 (12 mm preserved, photo by T.M. Gosliner). **E–H.** *P. anae* sp. nov. **E–F.** Panama. Photos by T.M. Gosliner. **E.** CASIZ 088182 (holotype). **F.** CASIZ 088240 (paratype). **G, H.** Guanacaste, Costa Rica. Photos by Y. Camacho-García. **G.** MZUCR8917. **H.** MZUCR8919.

California, where Engle & Richards (2001) stated that the range was extended during the El Niño summer of 1998. The specimens studied here from Costa Rica and Panama belong to *P. anae* n. sp. (described below). Specimens from Chile have not been examined.

Remarks: The studied specimens from Baja California (Fig. 2C, D) are very similar to the holotype (Fig. 2A, B) with the following minor differences: the original description states that the inner veil processes of the holotype are black and the two outer veil processes white, while in our specimens all processes are the

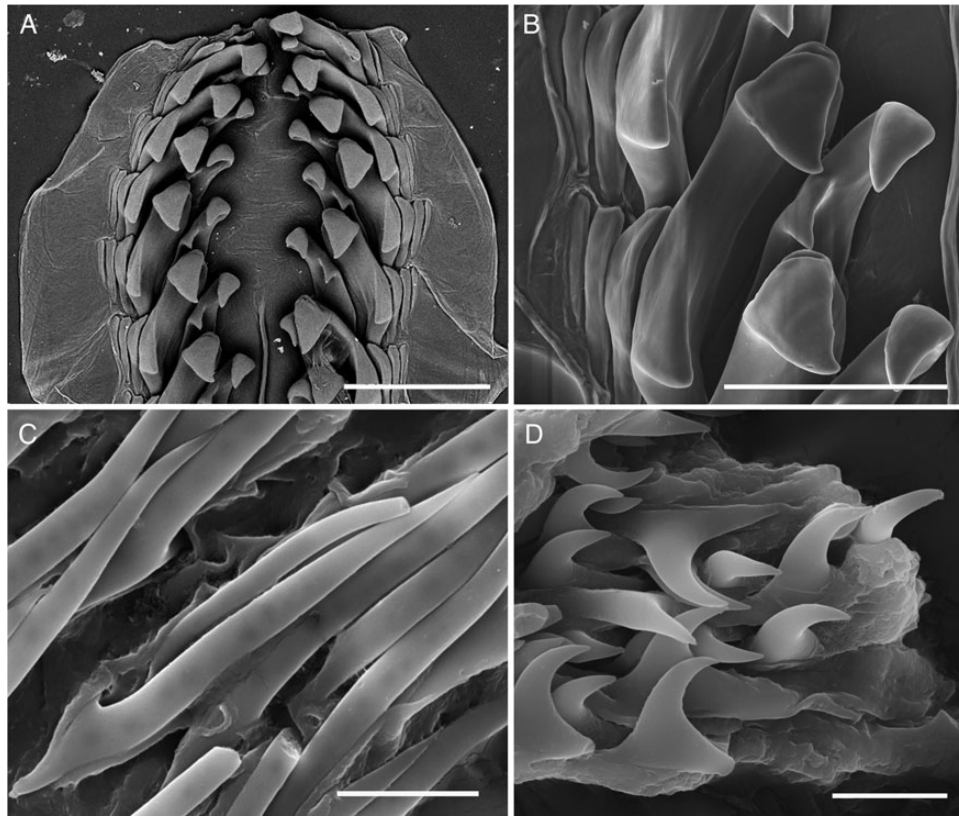


Figure 3. *Polycera alabe* Collier & Farmer, 1964. SEMs of CASIZ 72119. **A.** Radula. **B.** Left half of radula. **C.** Detail of distal penial spines. **D.** Detail of proximal penial spines. Scale bars: **A** = 500 µm; **B** = 200 µm; **C, D** = 10 µm.

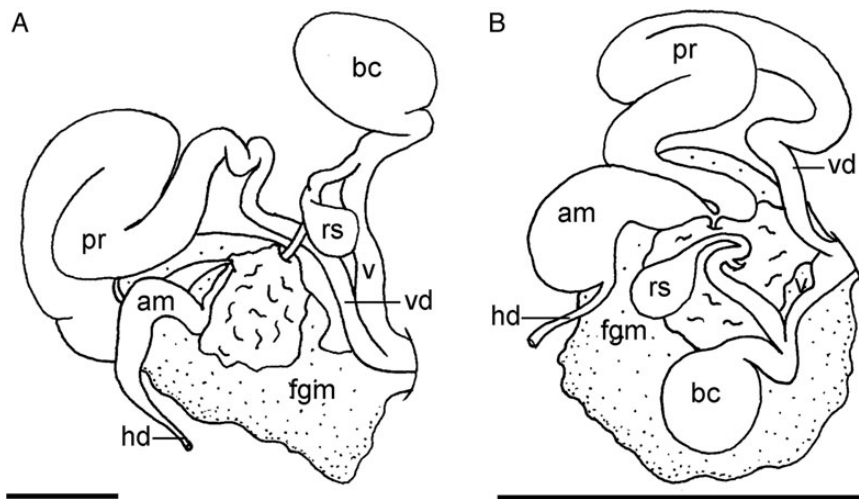


Figure 4. Reproductive system. **A.** *Polycera alabe* Collier & Farmer, 1964 (CASIZ 72119). **B.** *P. anae* n. sp. (CASIZ 088240). Abbreviations: am, ampulla; bc, bursa copulatrix; fgm, female gland mass; hd, hermaphroditic duct; pr, prostate; rs, receptaculum seminis; v, vagina; vd, vas deferens. Scale bars = 1 mm.

same colour, either white or dark grey. The original description gives the number of gill branches as six, while we observed eight gill branches in the holotype as well as in those from Baja California. The two posterior branches are extremely difficult to detect due to their small size, which likely accounts for the discrepancy. The number of rhinophoral lamellae was previously limited to 11, probably because only one specimen was studied,

but we recorded 11–15. The shape of the teeth was poorly described in the original description. Collier & Farmer (1964) recorded only one distal cusp on the outer lateral teeth in the holotype, whereas we observed two cusps on the outer lateral teeth in this specimen, as in the others studied. The radulae of some specimens show a fourth minute marginal tooth in some rows. In this study we describe the reproductive system for the

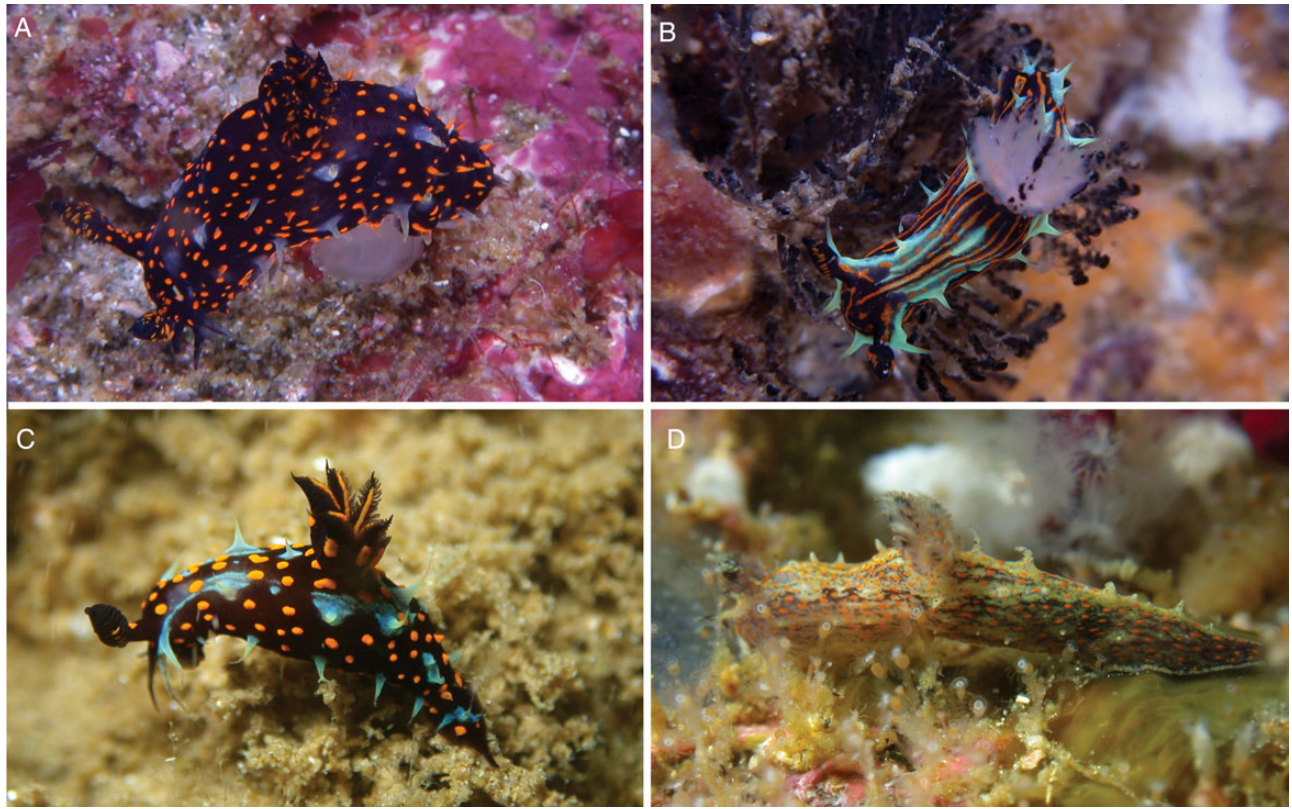


Figure 5. Variations of *Polycera alabe* recognized by Behrens & Hermosillo (2005) and Hermosillo, Behrens & Ríos-Jara (2006). **A.** ‘Variation A’, Isla de Cedros (type locality), Baja California, Mexico. **B–D.** ‘Variations B–D’, Bahía de Banderas, Mexico. Photos by A. Hermosillo.

first time and present SEM and micro-photographs of the penial spines.

Schrödl (2003) gave a relatively short anatomical description of two specimens collected in northern Chile and identified as *P. alabe*. The Chilean specimens show some small differences from typical *P. alabe*, such as 8–10 unipinnate gills and 6 digitiform frontal veil processes, *cf* 8 and 4 respectively. Schrödl (2003) reported for the first time that the first lateral tooth is bicuspid, which matches our observations. The reproductive system was not studied. However, since no specimens from Chile have been studied here we cannot confirm or dismiss the presence of *P. alabe* in this region.

Behrens & Hermosillo (2005) and Hermosillo & Valdés (2007) stated that *P. alabe* displays four distinct colour forms, which appear to be correlated with the geographical range (Fig. 5): variation A found throughout the Gulf of California, the Pacific coast of Baja California and southern California; variation B found on the Pacific coast of Mexico; variation C found from Mexico to Costa Rica and Panama; and variation D found further south. Santander & Valdés (2013) studied the genetic structure of *P. alabe* and concluded that as currently defined it is a paraphyletic assemblage. However, the authors did not sample specimens from the type locality; they included specimens identified as ‘*P. alabe*’ from La Paz (Baja California Sur), Sonora, Jalisco, Colima and Guerrero (all in Mexico), and resolved three well-supported clades: a northern clade (La Paz, Sonora and Jalisco), a southern clade 1 (Sonora and Jalisco) and a southern clade 2 (Colima and Guerrero). Santander & Valdés (2013) concluded that the southern clade 2 was also supported by radular morphological differences and that the two other clades could also possibly constitute different species. The authors noted that it was not possible to conclude which of the two clades found in the Gulf of California (the northern clade

and the southern clade 1) corresponded to *P. alabe* in the strict sense, since the range of both included the type locality (Isla de Cedros, Baja California). A comprehensive morphological comparison between the specimens studied by Santander & Valdés (2013) and our own would hopefully resolve this question. Molecular data are not available for our specimens since all were preserved in formalin.

Regarding the variations mentioned by Behrens & Hermosillo (2005), the external description and geographical range (Channel Islands, the type locality Isla Cedros and throughout the Gulf of California) for variation A, match perfectly the type specimens and our studied specimens; therefore we conclude that variation A is the true *P. alabe*. Regarding the other variations, further anatomical and molecular studies are needed to resolve this possible complex of cryptic species. These future studies should include specimens from the Pacific coast of Central and South America.

Polycera anae new species

(Figs 2E–H, 4B, 6)

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Polycera sp.: Camacho-García *et al.*, 2005: 76.

Type material: Holotype: adult (4 mm preserved), Rocas Tiburon off Isla Brincano, Panama, Pacific O. (7°50′03″N, 81°48′00″W), coll. T. M. Gosliner, 20 April 1993, dissected, photo slide (CASIZ 088182). Paratypes: 1 adult (3 mm preserved), Punta Ursula, Isla Jicarita, Panama, Pacific O. (7°14′00″N, 81°48′00″W), coll. T. M. Gosliner, 17 April 1993, 9 m, dissected, photo slide (CASIZ 088240); 1 spec. (2.5 mm preserved), same data, photo slide (CASIZ

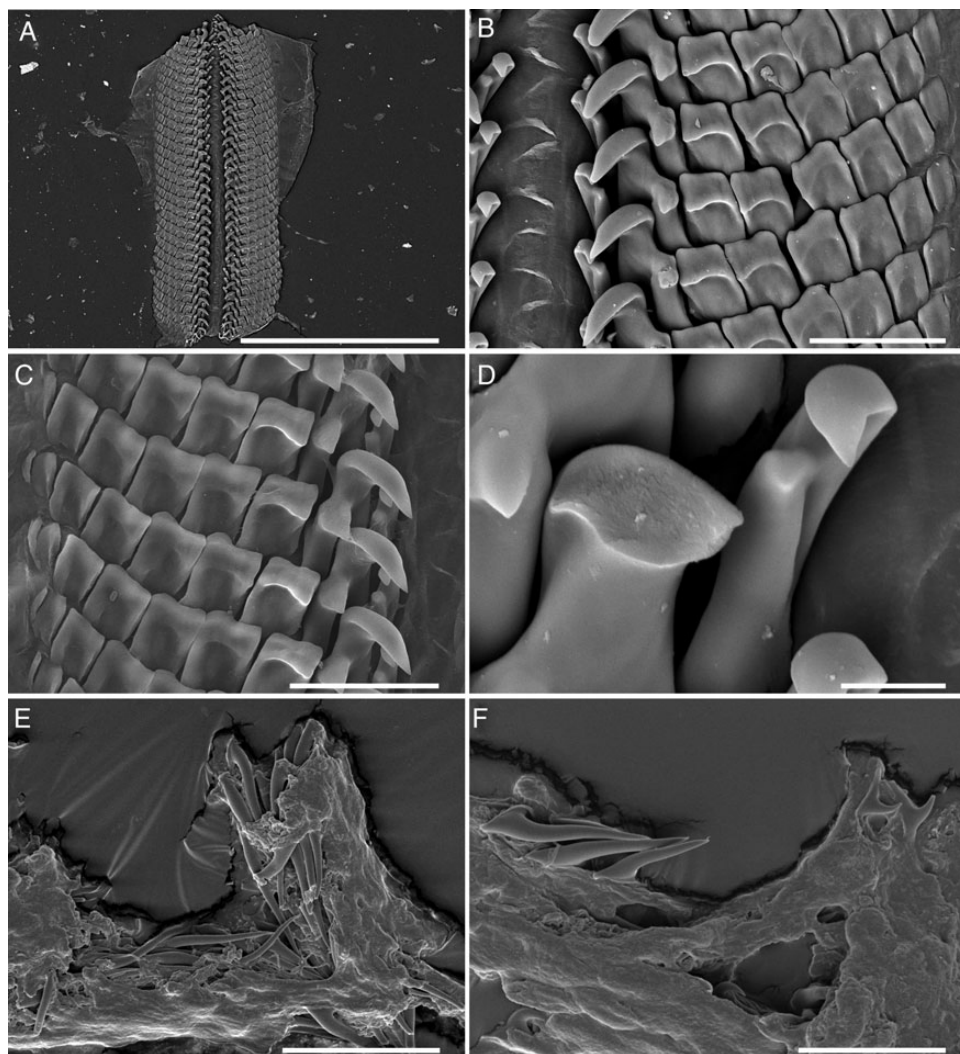


Figure 6. *Polycera anae* n. sp. Scanning electron micrographs. **A, B.** CASIZ 088182 (holotype). **A.** Radula (MZCUR8919). **B.** Right half of radula. **C.** Left half of radula. **D.** Detail of innermost lateral tooth (CASIZ 088182). **E, F.** Penial spines (MZCUR8919). **E.** Distal penial spines. **F.** Proximal penial spines. Scale bars: **A** = 500 μm ; **B, C** = 50 μm ; **D** = 10 μm ; **E** = 30 μm ; **F** = 20 μm .

186757); 1 spec. (5 mm preserved) Playa Real, Guanacaste, Costa Rica, Pacific O. ($10^{\circ}23'00''\text{N}$, $85^{\circ}50'00''\text{W}$), coll. Y. Camacho, 13 Jan. 2001, 9 m, dissected (MZCUR-INB0003118315); 1 spec. (4 mm preserved), Playa Grande, Guanacaste, Costa Rica, Pacific O. ($10^{\circ}20'43''\text{N}$, $85^{\circ}51'36''\text{W}$), coll. Y. Camacho, 9 Feb. 2012, intertidal, dissected (MZCUR8917); 1 spec. (4 mm preserved), same data dissected (MZCUR8919); 3 specs (2, 2, 1 mm preserved), same data, not dissected (MZCUR8918); 1 spec. (3 mm preserved), same data, not dissected (MZCUR8920); 1 spec. (3 mm preserved), Playa Herradura, Puntarenas, Costa Rica, Pacific O., coll. Y. Camacho, 19 Sept. 2011, one specimen, intertidal, dissected (MZCUR8090).

Etymology: Dedicated to Ana Ramiro, sister of the last author.

External morphology (Figs 2E–H, 3B, 6): Length to 5 mm living. Body long, limaciform, widest and highest at level of gill cirlet. Notal margin a low ridge enlarging anteriorly as flange curving around head (oral veil); our conical velar processes nearly equally spaced along oral veil; outer pair located next to rhinophores, longer than inner pair. Foot elongate, with short pointed anterior pedal corners. Body surface smooth, with small blunt tubercles scattered on lateral sides. Notal margin with

three main pairs of digitiform papillae: first pair halfway between rhinophores and gills; second pair next to anterior section of gill cirlet; third pair right after gills. After third pair, tail with 4 processes; processes smaller than notal margin processes. Rhinophores with 8–10 wide lamellae decreasing in size towards apex; arising from a smooth, short sheath. Oral tentacles short, tapering. Pericardial swelling anterior to gill cirlet. Gill of up to 6–7 nonretractile unipinnate branches forming semicircle around elevated anal papilla; gill branches decreasing in size posteriorly. Genital opening on right side, below first pair of papillae. Body colour black mottled with several rows of interrupted orange lines or dots along body (Fig. 1E–H). Rhinophoral lamellae, papillae, oral veil processes, gill branches and foot translucent white. Rhinophoral sheath black with orange spots. Area above eyes translucent.

Internal anatomy: Oral tube soft, short; buccal bulb muscular. Labial cuticle with rodlets. Radula (Fig. 6A–D): formulae $29\text{--}31 \times 6\text{--}7.2.0.2.6\text{--}7$ (CASIZ 088182, CASIZ 088240, MZCUR8917 and MZCUR8919) and $36 \times 8.2.0.2.8$ (MZCUR-INB0003118315); no rachidian teeth; inner lateral teeth small, hamate, narrow with strong prominent distal cusp, smaller second cusp approximately at two thirds of length (Fig. 6B–D); outer lateral teeth



Figure 7. Eastern Pacific *Polycera* species. Photographs of living animals. **A.** *P. kaiserae* [Hermosillo & Valdés, 2007](#), Mismaloya, Bahía de Banderas, Pacific coast of Mexico; 14 mm alive. Photo by A. Hermosillo. **B.** *P. tricolor* [Robilliard, 1971](#), British Columbia, Canada; 20 mm alive. Photo by M. Wakeling. **C.** *P. priva* [Er. Marcus, 1959](#), Comau Fjord, Chile; 50 mm alive. Photo by M. Schrödl. **D.** *P. hedgpethi* [Er. Marcus, 1964](#), Morro Bay, California; 20 mm alive. Photo by P. Garner. **E.** *P. atra* [MacFarland, 1905](#), Isla Santa Rosa, Channel Islands, California; 8 mm alive. Photo by A. Hermosillo.

hamate, larger than inner laterals, distal cusp wide triangular, secondary cusp quadrangular, perpendicular to axis of tooth, midway on external margin of tooth (Fig. 6B, C); marginal teeth rectangular plates, decreasing in size towards margin; inner marginals with prominent curved spur at anterior end, outer marginals simple plates (Fig. 6B, C). Pair of elongate, narrow salivary glands attached at the sides of the oesophageal connection. Blood gland well-developed, beneath intestinal loop. Reproductive system (Fig. 3B): triaulic; hermaphroditic duct long, slender; ampulla thicker, kidney-shaped; postampullary duct bifurcating into short oviduct leading into female gland and vas deferens with prostatic portion; vas deferens folded before ending penis; massive prostate gland folded, enclosing bursa copulatrix, then tapering to distal long vas deferens, ending in penis; bursa copulatrix large, oval; penis armed with numerous hooked spines in proximal section of vas deferens, spines elongate, pointed at distal end (Fig. 6E, F); vagina long, connected to bursa copulatrix; receptaculum seminis

pyriform, smaller than bursa copulatrix, on thin but elongate stalk, semiseriably arranged; receptaculum seminis and bursa copulatrix connected by large elongate duct; small uterine duct connects vagina with female gland; female gland mass well developed.

Distribution: Although this species has been reported from Islas Revillagigedos (Mexico), Costa Rica, Panama and the Galapagos Islands ([Camacho-García et al., 2005](#), as '*Polycera* sp.'), we here only confirm its presence in Panama and Costa Rica (present study) and Mexico ([Santander & Valdés, 2013](#), as '*P. alabe* Southern Clade 2') (see Remarks).

Remarks: *Polycera anae* may be easily misidentified as *P. alabe* owing to similarities in external appearance. However, there are also differences in distribution, body size, colour pattern, external and internal features. *Polycera anae* is a small species, with a maximum length of 5 mm alive, while living specimens of *P. alabe* reach up

to 25 mm. Although the colour patterns are quite similar, in *P. anae* both the rhinophoral lamellae and the pinnae of the gills are translucent white, whereas in *P. alabe* these are black and with orange spots. The number of lamellae on the rhinophores is also different, *P. anae* has up to 10 and *P. alabe* up to 15. Internally, the radula is an effective diagnostic character: the number of tooth rows is much higher in *P. anae* (29–36) than in *P. alabe* (9–13); *P. anae* has a greater number of marginals than *P. alabe* (6–8 vs. 3–4). In addition, the teeth also vary in size and shape of the cusps, although this may be an artefact related to the angle of the teeth as depicted in the figures. The inner marginals of *P. anae* exhibit a prominent curved spur at the upper end and the outer plates are simple, whereas in *P. alabe* they have a slightly curved cusp and a small spur. Regarding the jaws, we cannot make comparison between the species as unfortunately we were not able to get a good picture of the jaws of *P. alabe*. The salivary glands are also different; in *P. alabe* they are digitiform and rather short, whereas in *P. anae* they are elongate and narrow. Differences in the reproductive system are not clear. Both species have a well-developed prostate, which partially encloses the large bursa copulatrix. The penis in both taxa is armed with at least two different types of penial spines. *Polycera alabe* has a narrower ampulla, but larger bursa copulatrix than *P. anae*, but in the latter the pyriform receptaculum seminis has a longer stalk.

We also studied two specimens from the Galapagos Islands (CASIZ 97507). These are 17 mm in length preserved and have orange spots on the rhinophoral lamellae and gills. The bodies are brownish with orange lines and spots. The projections are translucent but, like the gills and rhinophores, have small orange spots. Each specimen has 16 body projections and 4 foot processes. Internally they only have four marginal teeth (9 × 4.2.0.2.4). Thus, they do not correspond to *P. anae*. They have some similarities to *P. alabe*, but *P. alabe* has 20 body projections and 10 foot processes. The number of lamellae in the rhinophores is also fewer (9–10) than in *P. alabe* (11–15). The colour pattern described above matches the variation D of Behrens & Hermosillo (2005) and also the picture shown by Camacho-García et al. (2005) as typical for the tropical eastern Pacific. More specimens from the Galapagos Islands as well as more detailed morphological and molecular studies are needed in order to resolve their identity.

Santander & Valdés (2013) included two specimens identified as '*P. alabe*' that were recovered in their well-supported 'Southern Clade 2'. These were collected from Colima and Guerrero (central Mexico) and have the same number and morphology of the marginal teeth as our specimens of *P. anae*. This, together with the picture of the living animal given by Santander & Valdés (2003: fig. 1), suggests that *P. anae* is also present in central Mexico. However, further anatomical and molecular studies are needed in order to confirm the geographical range of this species.

DISCUSSION

With the description of the new species *Polycera anae*, the number of species of this genus found in the eastern Pacific rises to seven. Table 1 shows the main external and internal features of these species summarized from the available literature. Almost all are easy to distinguish based on their body colouration (Fig. 7). For example, *P. kaiserae* described from Bahía Banderas (Mexico) is unmistakable because of its pink body sprinkled with white spots (Hermosillo & Valdés, 2007) (Fig. 7A). *Polycera tricolor*, found from British Columbia (Canada) to Baja California (Mexico), has a characteristic translucent grey-white body with a yellow line around the foot margin on the caudal crest and pallial ridge (Robilliard, 1971) (Fig. 7B). *Polycera priva*, so far only found in Chile (Schrödl, 1996, 2003, 2009; Schrödl et al., 2005), can be easily distinguished by its brownish colouration (Fig. 7C). The

potentially invasive species *P. hedgpethi* is also easily recognized by its grey body colour with black spots (Marcus, 1964) (Fig. 7D). All these four species lack the orange spots or marks present in *P. atra*, *P. alabe* and *P. anae*. However, the general body colour of *P. atra* is pale grey with narrow or broader lines of black on the dorsum and sides (MacFarland, 1966) (Fig. 7E), whereas the body colour of *P. alabe* and *P. anae* is blue-black or black covered with orange spots (Fig. 2). However, as discussed under the remarks on *P. anae*, this species has rhinophoral lamellae and gills that are translucent white, whereas in *P. alabe* they are black with orange spots.

Besides the body coloration there are other marked external differences among these species. For instance, *P. priva* and *P. tricolor* have a high number of extrabranchial processes on each side (4–6), whereas the remaining species have only three (*P. hedgpethi*), two (*P. atra*), one (*P. kaiserae*), or they are even absent as in *P. alabe* and *P. anae*. The last two species have some tubercles in the gill area, but they are not clear extrabranchial processes.

Internally the radular formulae are similar for most of the species. In this case, *P. anae* is the easiest to distinguish since the number of marginal teeth is higher than in the other eastern Pacific species (6–8 vs 2–4) (Table 1) and it also has the highest number of rows of teeth (29–31 vs 9–17). Differences between the reproductive systems are difficult to determine. It is clear that all these seven species have a large and differentiated prostate and also a penis armed with spines, but a more detailed and complete comparative anatomical study is required.

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