Three new species of hagfish (Myxinidae, *Eptatretus*) from the Galápagos Islands

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The Galápagos Islands consist of about ten principal islands and over 100 smaller ones on the equator about 500 miles off the coast of Ecuador. The biodiversity found in these islands by Darwin (1896) is also expressed in the marine fauna (McCosker, 1997); three new species of hagfish were found in only eight specimens from four trap sets (Fig. 1). An exploration off South America and the Galápagos Islands in 1891 by the U.S. Fish Commission Steamer Albatross reported only one species of hagfish, Myxine circifrons Garman (1899), taken off the Gulf of Panama. Several species of Eptatretus with from nine to fourteen gill pouches have since been reported from the eastern Pacific coast (Wisner and McMillan, 1990), and one species with seven gills, E. laurahubbsae McMillan and Wisner (1984), from the Juan Fernandez Islands, Chile. The new species described below are the first hagfish reported from the Galápagos Islands, and the first *Eptatretus* with five, six, and eight gill pouches known from the eastern Pacific. Until the formation of the Panamanian land bridge there was a longstanding connection between the Caribbean and eastern Pacific, which provided a passage for hagfish to move into the eastern Pacific from the Caribbean. These new species may be more closely related to the *Eptatretus* with five to eight gills found in the Caribbean and western Atlantic than to any currently known from the eastern Pacific. Future collecting efforts along the coast of Ecuador may reveal *Eptatretus* similar to those found off the Galápagos Islands; however, until further material is available and genetic studies are made for comparison, we can only speculate on possible origins of these Galápagos hagfishes.

Although the body color of most species of *Eptatretus* is brown to black, Fernholm (1991) described a species of Eptatretus on the basis of one specimen with a highly unusual pink body, stating that its color was probably caused by diet. Of the eight specimens reported here, the seven larger ones are dark purplishbrown to black or dark gray where the slime has not been removed. The smallest specimen (about 142 mm), possibly an albino, is ivory to light tan. Not enough is known about the early development of hagfish to determine if this tiny specimen would have become darker with age. Albinism has been reported by Dean (1903) and Jensen (1959), and I have collected one, an adult E. deani (Evermann and Goldsborough, 1907), which was pinkish-white when alive and light tan color in formalin.

Head grooves are present near the eyespots of the seven large specimens, but not on the smallest hagfish. These grooves, found in many other species of *Eptatretus* (McMillan and Wisner, 1984) and once thought similar to lateral lines,

Manuscript accepted 7 April 1998. Fish. Bull. 97:110–117 (1999). are now believed to have no sensory purpose. Because they are not considered a species-specific character and have no taxonomic value for the present study, they are not included in the species descriptions.

Counts of cusps and total slime pores are somewhat similar for these new Eptatretus species, but there are significant differences in the multicusps, numbers of gill pouches, and prebranchial slime pores which readily distinguish each of them. Some differences are found in the white patterns on the face and barbels of the seven pigmented specimens (Fig. 2), but color cannot be assigned major importance as a species character with such a small sample size.

Material and methods

All new specimens for this study were collected by John E. Mc-Cosker et al using the research submersible Johnson Sea-link

(JSL), on the California Academy of Sciences and Harbor Branch Oceanographic Institute Galápagos Expedition during November 1995. The three new species collected by the California Academy of Sciences and described below were among eight specimens taken in four trap sets; additional specimens (nontypes) of Galápagos Eptatretus were given to the Instituto Nacionál de Pesca, Guayaquil, Ecuador, and were not available for study. Collection data and disposition of specimens are listed in the treatment of each new species. Institutions in which type specimens have been deposited are the California Academy of Sciences, San Francisco (CAS), United States National Museum, Washington, D.C. (USNM), and Scripps Institution of Oceanography, La Jolla, California (SIO).

McCosker¹ described the method of capture as follows: "Hagfish were collected with a small, galvanized wire commercial two-fyked minnow trap, about 30 by 50 cm, with 2 cm openings at each end. The



Figure 1

Principal islands of the Galápagos Archipelago, showing collection locations of the three new species of *Eptatretus*: $\blacksquare = E$. grouseri; $\blacklozenge = E$ mccoskeri; and $\blacktriangle = E$. wisneri.

traps were baited with fish or molluscs, or both, weighted and set with a pinger and short float line to be retrieved on the next dive [about 8 h later]; all traps were set among boulders, often on steep slopes." McCosker further reported that every set resulted in hagfish so elusive that many escaped traps during or after retrieval. Hagfish were also seen, but not taken, at 267 m off East Seymour Island, at 274 m off Isla Floreana (Charles Island), and off Isla San Salvador (James Island) at 884 m over sand bottom, at 6.54°C. Although no study material resulted from these sets, the reports furnish information on the little-known habitat of these benthic fish and indicate locations for further collecting effort.

Methods of measuring and counting follow those of McMillan and Wisner (1984) and Wisner and McMillan (1995). Counts of gill pouches (GP), gill apertures (GA), and cusps are noted for both sides, but body proportions and slime pore counts are for the left side only. Low power magnification was required to discern head grooves and fusion of multicusps. Most of the specimens are twisted and previously cut, making some measurements and counts difficult; all specimens were measured several times and averages

¹ McCosker, J. E. 1996. California Academy of Sciences, Golden Gate Park, San Francisco, CA, 94118-4599. Personal commun.



Figure 2

Frontal aspect showing pigmentation on face and barbels of three new species of *Eptatretus* from the Galápagos Islands: (A) holotype of *E. grouseri* (CAS 86428); (B) holotype of *E. wisneri* (CAS 86429); (C) paratype of *E. wisneri* (SIO97-76); (D) holotype of *E. mccoskeri* (CAS 86431); (E) and (F) paratypes of *E. mccoskeri* (SIO 97-75; and (G) paratype of *E. mccoskeri* (USNM 344905).

used to calculate body proportions in thousandths of total length (TL). About 6–9 mm of shrinkage was noted in the total lengths of the seven larger specimens between 24 March 1996 and 20 March 1997. Some shrinkage occurs in all specimens on preservation, mainly in the trunk length, and apparently slightly more when ethanol is the preservative used, as in this case. Selected body proportions and counts of slime pores and cusps are shown in Table 1.

Because variations in color patterns are often noted in large collections, color is not usually considered of much importance. However, the two species with eight gills have distinct differences in the white patterns on their faces and barbels that may provide a useful character in distinguishing between them. The "face" is the anteriormost part of the specimen, the ventral aspect of the head, delineated by three pairs of long, slender protruberances thought to be sensory organs and commonly referred to as barbels. The first two pairs are above and below and adjacent to the nasopharyngeal duct, which is the intake opening for water to the pharynx.

The third pair lies just outside another set of short, fleshy protruberances on each side of the mouth,

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Table 1

Selected measurements and counts of slime pores and cusps of three new species of *Eptatretus* from the Galápagos Islands. Species name followed by number of specimens in parentheses; total length of holotype followed by TL of paratype(s) in parentheses; body proportions in thousandths of TL, average followed by values for all specimens.

Measurements	E. grouseri (2)	E. mccoskeri (4)	E. wisneri (2)
Total length in mm	380 (142)	320 (283,298,300)	360 (328)
Prebranchial length	225 (211-239)	249 (237-262)	213 (194-229
Branchial length	71 (63-79)	97 (93-101)	105 (100-111)
Trunk length	544 (535-553)	494 (487-500)	516 (503-28)
Tail length	160 (158–162)	162 (156-177)	168 (167-171
Body depth withVFF	58 (49-68)	100 (94-106)	88 (85,92)
Body depth without VFF	54 (42-66)	100 (94-106)	87 (85-89)
Body depth at cloaca	54 (48-61)	83 (78-88)	72 (72-73)
Tail depth	63 (63-63)	93 (87-102)	77 (76–78)
Slime pore counts (holotypye followed by	paratype(s) in parentheses):		
Prebranchial pores	12 (13)	15 (14,14,14)	9 (9)
Branchial pores	4 (5)	7 (7,7,7)	7(7)
Trunk pores	46 (44)	42 (40,40,41)	46 (47)
Tail pores	15 (14)	10 (10,11,12)	14 (13)
Total pores	77 (76)	74 (72,72,74)	76 (76)
Cusp counts, left, right (holotype followe	d by paratype(s) in parentheses:		
Multicusps, ant./post.	3,3/2,2	3,3/3,3	3,3/2,2
Unicusps(anterior rows)	8,9 (9,9)	$10,10^{\bullet}(9,9-9,10)$	9,9 (9,9)
Unicusps(posterior rows)	8,8 (8,8)	9,10 (9,9–9,10)	8,8 (8,8)
Total cusps	44 (44)	51 (48,50,51)	44 (44)

called "labial barbels," which are immediately adjacent to the oral opening, very similar in size and shape in all hagfish, and are not considered species signifcant. The other three pairs of long, slender barbels are usually measured, and sometimes used as a species character. Because many of the barbels on these specimens were curled and accurate measurements difficult to repeat, this character has not been included in Table 1. Multicusp pattern, and counts of GP and slime pores have proved to be more accurate and less variable intraspecifically than color or barbel size, and thus more important as species characters. The most significant features of taxonomic importance in this study are the counts of gill pouches, prebranchial slime pores, and multicusps. "First" and "last" refer to anteriormost and posteriormost, and tail pore counts are the sum of cloacal and caudal slime pores; other terms used are from Wisner and McMillan (1990, 1995). The water passes from the the nasopharygeal duct through the pharynx, through each afferent branchial duct (ABD) to the gill pouch, and exits through each efferent branchial duct (EBD) to its corresponding external opening (GA). Any excess water passes out of the body through the slightly larger duct known as the pharyngocutaneous duct (PCD), which is ordinarily confluent with, or occasionally just posterior to the last GP on the left side. The numbers of gill apertures are not shown in the table because the number of GA is normally the same as that of GP in the genus Eptatretus; however, GA count is given in each species description and in the key because the apertures are readily counted by external examination and thus provide a useful character for identification in the field. In most *Eptatretus*, including the three species described in the present study, the number of branchial pores is one less than the number of gill apertures, and each pore is slightly below and posterior to each GA anterior to the PCD. The next slime pore, usually found above and behind the PCD, is the first in the trunk pore series.

Key to the species of Eptatretus of the Galápagos Islands

 1b Eight gill pouches and apertures each side 2

- 2b Prebranchial slime pores 9; trunk pores 46–47, tail pores 13–14; total pores 76, total cusps 44, multicusps 10 (3 in anterior, 2 in posterior rows), nasal barbels dark with occasional white tips; face, mouth, and base of labial barbels nearly all white *E. wisneri*, new species

Eptatretus grouseri new species

Holotype CAS 86428, female, 378 mm TL, taken at 00°14.6'S, 91°26.6'W, in a minnow trap at 2370 ft [722 m], 17 November 1995.

Paratype SIO97-77 (formerly CAS 86428), 142 mm TL, (juvenile, sex not determined), taken with the holotype.

Diagnosis Five or six gill pouches and apertures each side, last GA confluent with PCD on left side. Multicusps 3 in anterior and 2 in posterior rows, 9 unicusps in each anterior and 8 in each posterior row, total cusps 44; slime pore counts: prebranchial 12– 13, branchial 4–5, trunk 44–46, tail 14–15, total slime pores 76–77; ventral finfold (VFF) vestigial, caudal finfold (CFF) well developed.

Description Both specimens have a bluntly rounded rostrum; barbels well developed, first two pairs about equal, third pair slightly longer; greatest body depth in trunk region about the same as tail depth, both only about 6 to 7 percent of TL; prebranchial length about 23 percent of TL, trunk length about 54 percent of TL. Although the two specimens differ greatly in size and color, counts of pores and cusps are very similar (Table 1). The holotype has tiny round eggs less than one mm in diameter, no ellipsoidal developing eggs and no tissue indicating previous large eggs. Body color brownish-black, head region dark brown; eye spots prominent, nearly round; face dark with small white area around mouth and on tips of barbels (Fig. 2A); VFF vestigial, with pale margin on posterior third; cloacal opening white; GA widely spaced with branchial slime pores posterior to and just slightly below each aperture; all GA on the right side and all but the second on the left side have white margins. The second gill pouch on the right side of the holotype is either undeveloped or degenerated; it appears only as thickened muscle tissue connecting the ABD from the pharynx with the EBD leading to the external GA. Only one GP lies along the tip of the dental muscle; the branchial aorta splits after the fifth (last) GP.

The paratype has six normally rounded GP on each side, the first two along the dental muscle with the

aorta branching after the fifth GP. Possibly an albino, this tiny specimen is an unusual ivory to light tan body color, the prebranchial area slightly lighter with eye spots barely discernible. Also, the area around the mouth and cloaca are lighter than the body; the trunk region is just dark enough to show a white margin on the VFF. The slime pore openings are very small, but easily detected because the sacs bulge just under the thin skin.

The specimen is so young that it was not possible to determine gender even with magnification. The total cusp count only differs from that of the holotype by one, but they are very small and the multicusp fusion pattern is too indistinct to discern the number of fused cusps in the posterior rows with any certainty.

Etymology This species is dedicated to my son David "Grouser" McMillan, a Chief Engineer in the U.S. Merchant Marine, for his continued encouragement of my hagfish studies and for his knowledge and love of ships and the sea.

Distribution Known only from Galápagos Islands, Ecuador; both specimens were collected in JSL dive 3958 at 2370 ft [722 m] in Bolivar Channel, off Cabo Douglas, in a minnow trap set on a steep, sedimentladen slope of an invertebrate-rich pinnacle of pillow lava. This was the deepest set in which hagfish were collected on this expedition (Fig. 1).

Comments The body proportions differ by about two percent in prebranchial length and depth of body in trunk region, possibly because one specimen is a juvenile. Pore and cusp counts, which remain the same throughout the individual's life span, are very similar in both specimens. In this tiny paratype the first GP is about 3.5 mm in diameter, with each subsequent GP decreasing in size posteriorly until the sixth is only about 2 mm across. This difference has also been noted in size of unicusps, in which the last one is frequently much smaller at the end of each row, apparently a normal growth pattern. I have not made any study of changes in body proportions of hagfishes from juvenile to adult stage because very small specimens are seldom collected. The capture of this tiny

specimen was possible because of the use of a minnow trap with very small openings. Because of the larger openings of the myxinid traps most often used, hagfishes of less than 200 mm length are seldom caught because they escape before a trap reaches the surface.

The larger specimen with normal pigmentation has been designated as the holotype (in spite of its one abnormal GP), because its adult size allowed determination of gender and multicusp pattern, as well as more accurate measurements and slime pore counts. These two specimens are assigned to the same species because there is such close agreement in cusp and slime pore counts and most body proportion and because they were collected in the same JSL dive. A difference of one gill pouch is common in large collections of Eptatretus having nine or more GP, and this difference may be found occasionally in species having only five or six gill pouches. More specimens will be required to determine the usual number of GP, and whether there are enough other differences to warrant separating into two different species.

Discussion Three species of five- or six-gilled Eptatretus from the Caribbean Sea are E. minor and E. multidens Fernholm and Hubbs (1981), and E. mendozai Hensley (1985). Although these three species may have once shared a common ancestor with E. grouseri, they are readily distinguished by their unusual 3/3 multicusp pattern (three fused cusps on both the anterior and posterior rows of cusps). The 3/2 multicusp pattern of E. grouseri commonly occurs in most species of Eptatretus: two other fivegilled species with this pattern are E. profundis (Barnard, 1923) from South Africa and E. eos Fernholm (1991) from the Tasman Sea, the latter a distinctive pink color and possessing a tube-shaped, elongated "snout." Both of these species have about 26 prebranchial slime pores, an unusually high number for *Eptatretus* and twice as many as found in *E*. grouseri. Four species of Eptatretus with six gill pouches and the 3/2 multicusp pattern are E. burgeri (Girard, 1854) from Japan and Korea, E. hexatrema Barnard (1923) from South Africa, E. longipinnis Strahan (1975) from South Australia, and E. chinensis Kuo and Mok (1994) from the South China Sea. Eptatretus burgeri has a distinctive white middorsal line as well as notably higher prebranchial and total slime pore counts, whereas the other three species have prebranchial pore counts two to three times that of E. grouseri. Although the number of GP is the same, these six species are not considered closely related to E. grouseri because of the significant differences in prebranchial and total slime pore counts, as well as wide geographic separation.

Eptatretus mccoskeri new species

Holotype CAS 86431, male, 320 mm TL, taken at 01°06.3'S, 89°06.9'W, in a minnow trap at 704 ft [215 m], 16 Nov 1995.

Paratypes All males, taken with the holotype; SIO97-75, 2(283,300 mm TL); and USNM 344905, 298 mm TL.

Diagnosis Eight gill pouches and apertures each side, last GA confluent with the PCD on the left side; 12 multicusps (3 fused cusps in each row), unicusps 36– 39 (9,9 to 10,10 unicusps in each anterior and 9,9 to 9,10 in each posterior row), total cusps 48–51; total slime pores 72–74, prebranchial 14–15, branchial 7, trunk 40–42, tail 10–12. Ventral finfold absent or vestigial; caudal finfold with white margin; head slightly lighter than body color, face dark except for small white area around mouth; barbels all white (Fig. 2, D–G).

Description Body color brownish black with head region slightly lighter than body color, face dark except for small white area around the mouth; eye spots visible, but not prominent; face dark, in sharp contrast to completely white barbels; prominent white margin around CFF; rostrum bluntly rounded, nearly straight across; nasal orifice large, face wide with the distance between the pairs of barbels about equal to that of their length. First five (4-6) GP lie along the dental muscle; branchial aorta branches at the sixth or seventh GP; first GA high, series curving downward to the PCD near the ventral midline; series of prebranchial slime pores starts far forward; branchial pores are tiny and close to ventroposterior margins of each GA, trunk pore series starts above and behind the PCD, a space about 5-6 mm between last trunk pore and origin of cloaca. Prebranchial length is about one fourth of TL, trunk length about half of TL; greatest body depth in the trunk region about equal to the tail depth, both about 10 percent of TL (Table 1).

Etymology This species is dedicated to my friend John E. McCosker, Senior Scientist, California Academy of Sciences, San Francisco, California, for providing this new study material, as well as for his important contributions to marine biology.

Distribution Known only from the Galápagos Islands, Ecuador; all four specimens were taken in one dive (JSL 3936) from a minnow trap set on sand bottom at the top of a seamount southeast of Isla San Cristobal (Chatham Island) at about 215 m, the shallowest set where hagfish were found on this expedition (Fig. 1).

Comments Because male hagfish rarely outnumber females in any collection, it is unusual to find that these four specimens are all males; testes are developed, but no study was done to determine presence of sperm. Although both E. mccoskeri and E. wisneri (described below) have eight GP, the pattern of three fused cusps in both anterior and posterior rows, notably higher numbers of prebranchial pores and total cusps, and the sharp contrast of white barbels against the dark face of E. mccoskeri readily distinguish these two species. Also, the prominent white margin around the entire caudal finfold in E. mccoskeri is much less apparent in E. wisneri, showing only as a thin white edge on the tip of the CFF. The numbers of pouches along the dental muscle and position of branching of the branchial aorta also differ slightly in these two 8-gilled species. Three other species of Eptatretus with 8 GP and the 3/2 multicusp pattern are listed below in the discussiom of E. wisneri.

Eptatretus wisneri new species

Holotype CAS 86429, female, 356 mm.TL, taken at 00°28.0'S, 91°37.2'W, from a minnow trap at 1848 ft [563 m], 14 Nov 1995, [JSL dive 3952].

Paratype SIO97-76 [formerly CAS 86430], male, 328 mm TL, taken at 00°17.5'S, 91°38.9'W, from a minnow trap at 1680 ft [512 m], 16 Nov, 1995 [JSL dive 3957].

Diagnosis Eight gills and apertures on each side, last GA on left side confluent with PCD; 10 multicusps, pattern of 3 fused cusps in each anterior row and 2 in each posterior row; unicusps 9 each anterior row and 8 each posterior row; total cusps 44; total slime pores 76, prebranchial pores 9, branchial pores 7, trunk pores 46–47, tail pores 13–14; face mostly white, including mouth and base of labial barbels; other barbels dark, with occasional white spots or tips (Fig. 2, B and C); eye spots distinct, large and irregularly shaped; VFF vestigial or absent, CFF well developed.

Description Body color brownish-black, head slightly lighter, with large, distinct eye spots; gill apertures with white margins; face nearly all white, including area around mouth and base of labial barbels; the VFF appears only as a pale line on the posterior half of the trunk region and the tip of the CFF has a thin pale margin. The first 3 gill pouches lie along the dental muscle; the branchial aorta branches after fifth GP; the prebranchial length is about 21 percent of TL, trunk length about half of TL, the greatest body depth in trunk region about equal to, or slightly more than the tail depth, both about 8 to 9 percent of TL (Table 1).

Etymology This species is named after Robert L. Wisner, my friend and associate at Scripps Institution of Oceanography, for his invaluable assistance with my myxinid research as well as his other contributions to ichthyology.

Distribution Known only from Galápagos Islands, Ecuador, these two specimens were taken by minnow trap on two different JSL dives, off Cabo Douglas, Isla Fernandina (Narborough Island), at 512 and 563 meters (Fig. 1).

Comments The holotye is a young female with round eggs of less than one mm, with no ellipsoidal oocytes and without evidence of tissue indicating previous vitellogenesis. The paratype is a male with well-developed testes, but no examination was done for the presence of sperm. One specimen is more brown than black, and the white face patterns are slightly different; also, the barbels of one specimen are partly white and more robust than those of the other (Fig. 2, B and C). These slight differences could be due to gender, although pigmentation is highly variable and sexual dimorphism has not been reported in hagfish. Barbels are seldom used as a species character because they are often curled, making accurate measurements difficult, and not reliable as a body proportion because their lengths are so small compared with TL. These two specimens have the same pattern of multicusps and unicusp counts, and slime pore counts are the same or only differ by one. Therefore, until further collecting provides more specimens for comparison, the slight differences in pigmentation patterns and size of barbels are considered insignificant, and the two specimens are described as one species.

Discussion Eptatretus wisneri is readily distinguished from *E. mccoskeri* because of its multicusp pattern of 3/2 (three on each anterior and two on each posterior row), lower numbers of total cusps, notably lower number of prebranchial and total slime pores, as well as the darker barbels and the white areas over most of the face. Three other eight-gilled *Eptatretus* with the 3/2 multicusp pattern are *E. octatrema* Barnard (1923) from South Africa, *E. indrambarayai* Wongratana (1983) from the Andaman Sea (N.E. Indian Ocean), and *E. okinoseanus* (Dean, 1904) from Japan). However, they are not considered closely related to *E. wisneri* because of differences in counts, the wide geographical separation, and the fact that this multicusp pattern is common to most species of *Eptatretus*.

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