Eggs and late-stage embryos of *Allocareproctus unangas* (family Liparidae) from the Aleutian Islands

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Abstract Four unusual masses of adhesive fish eggs surrounding limbs of the octocoral *Primnoa* sp. were collected in Seguam Pass in the Aleutian Island Archipelago at a depth of 397 m. The eggs contained embryos in the flexion stage of development. When dissected and cleared and stained, counts of dorsal and anal fin rays and vertebrae allowed identification of the eggs as those of *Allocareproctus unangas* Orr and Busby, a newly described snailfish species. This is first report of snailfishes of the genus *Allocareproctus*, and of liparids other than those in the genus *Liparis*, spawning outside of lithodid crabs.

Key words Eggs · Liparidae · *Allocareproctus unangas* · *Primnoa* sp. · Aleutian Islands

There are currently 30 genera with more than 335 species recognized as valid in the family Liparidae (Chernova et al., 2004; Orr, 2004). Very little is known about reproduction and egg development of liparid fishes except for those of the genus *Liparis*, which spawn small demersal eggs in adhesive masses on a wide range of substrates (Ehrenbaum, 1905; Able and Musick, 1976; DeMartini, 1978; Marliave and Peden, 1989). Wenner (1979) reported spawning periods, fecundities, and ovarian egg diameters of *Paraliparis calidus*, *Paraliparis copei*, and *Paraliparis garmani* from the western Atlantic. Stein (1980) provided data on spawning period, fecundity, and ovarian egg diameters for *Paraliparis cephalus*, *Paraliparis latifrons*, *Paraliparis megalopus*, and *Paraliparis mento* from the eastern North Pacific. He also hypothesized that male *Paraliparis* guard nests. Able et al. (1984) summarized available information on the early life history of liparid fishes (mostly from Stein, 1980) and presented several illustrations of larvae. Several authors, including Love and Shirley (1993) and Somerton and Donaldson (1998), have reported liparid fishes of the genus *Careproctus* depositing eggs in the gill cavities of lithodid crabs. We report egg masses and embryos of *Allocareproctus unangas*, newly described by Orr and Busby (2006), as the first example of a liparid other than *Liparis* spawning in nature outside of lithodid crabs.

Materials and Methods

Four egg masses adhered to stalks of the octoral *Primnoa* sp. were collected near Seguam Island in the Aleutian Island Archipelago (Fig. 1). The egg masses were collected on 26 June 1997 with a bottom trawl at a depth of 397 m during a groundfish survey conducted by the National Marine Fisheries Service, Alaska Fisheries Science Center (AFSC), Resource Assessment and Conservation Engineering Division. Complete egg masses were measured along the deposition axis on the coral stalks and perpendicular (90°) to the deposition axis to the nearest 0.1 mm using dial calipers. Individual eggs of one mass (UW 094968 #1) were teased apart with forceps and counted. Minimum and maximum diameters of individual eggs were measured to the nearest 0.01 mm using a stereo dissecting microscope with a calibrated ocular micrometer. Several eggs were dissected, and late-stage embryos were removed from the chorions for illustrations. Seven of these embryos were differentially cleared and stained using the method of Potthoff (1984), and meristic features were counted. The presence of completely formed heamal spines in the embryos allowed us to distinguish abdominal from caudal vertebrae. Differences in meristic characters were assessed by conducting an ANOVA between the embryos and two similar species using data from Orr and Busby (2006).

Results

Identification. Eggs were identified as *Allocareproctus unangas* using meristic features of the embryos, particularly the number of anal fin rays (36–39) (Orr and Busby, 2006; Table 1). Of seven embryos cleared and stained, one had 36, three had 38, and three had 39 anal fin rays. The only other fishes in the family Liparidae from the North Pacific Ocean with similar meristic characters are other members of the genus *Allocareproctus* (Orr and Busby, 2006) and *Careproctus candidus* (Kido, 1988; Mecklenburg et al., 2002). *Careproctus candidus*, by its lower precaudal vertebral count of 9–10, does not overlap with the count of 11 found in all embryos. Among species of *Allocareproctus*, *Allocareproctus ungak* is easily excluded because of its

Table 1. Meristics of cleared and stained embryos and adults of species of *Allocareproctus* and *Careproctus candidus*

	Embryos $n = 7$	*A. unangas* $n = 33$	*A. jordani* $n = 58$	*A. tanix* $n = 4$	*A. kallaion* $n = 34$	*A. ungak* $n = 40$	*C. candidus* $n = 15$
Dorsal fin rays	43–45	42–45	41–45	41–42	42–45	39–42	43–48
Anal fin rays	36–39	36–39	34–38	34–36	34–37	33–35	36–41
Pectoral fin rays	23–34[a]	38–42	34–39	38–39	36–40	35–40	33–40
Caudal fin rays	6–10[a]	11–12	11–13	12–13	11–14	12–13	10–13
Precaudal vertebrae	11	10–11	10–12	9–10	12	10–11	9–10
Total vertebrae	48–50	46–50	45–49	45–47	47–49	44–47	47–52

Adult counts from Orr and Busby (2006)

[a] Incompletely developed

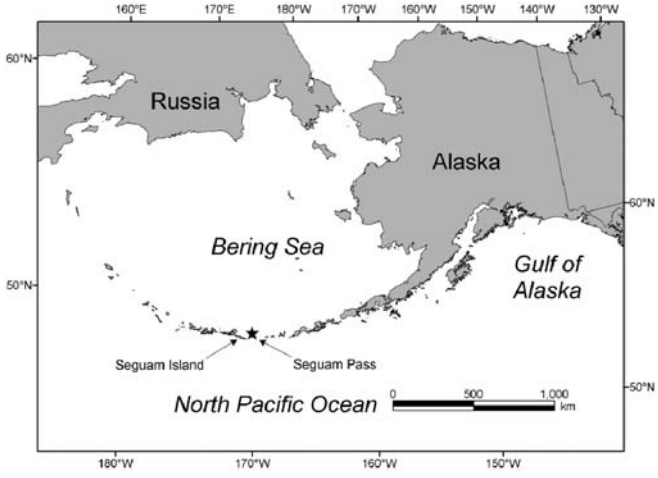

Fig. 1. Collection locality (*star)* of octocoral *Primnoa* sp. with attached egg masses of *Allocareproctus unangas* (UW 094968)

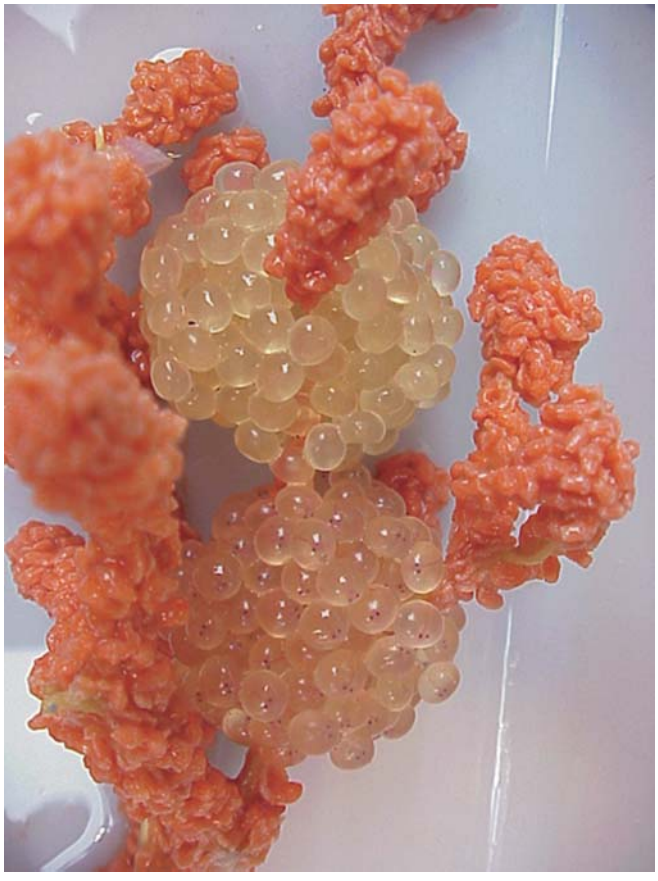

Fig. 2. Live photograph of octocoral *Primnoa* sp. with attached egg masses of *Allocareproctus unangas* (UW 094968)

lower dorsal and anal fin ray counts; *Allocareproctus kallion*, because of its higher number of precaudal vertebrae; and *Allocareproctus tanix*, because of lower dorsal fin ray and precaudal vertebrae counts. Since the counts of anal fin rays and precaudal vertebrae in embryos also overlapped with counts recorded for *Allocareproctus jordani* (Orr and Busby, 2006; see Table 1), we conducted an ANOVA using data from Orr and Busby (2006). Although mean numbers of anal fin rays in the embryos ($n = 7$, range 36–39, $\bar{x} = 38.1$, SD = 1.07) were not significantly different from *A. unangas* ($n = 33$, range 36–39, $\bar{x} = 37.1$, SD = 0.74), they were significantly different from *A. jordani* ($n = 58$, range 33–38, $\bar{x} = 35.7$, SD = 0.97, $P < 0.0001$). No statistical difference was found in numbers of precaudal vertebrae. In addition, three of the embryos had 39 anal fin rays, a count not found in *A. jordani*.

Description of egg masses. Egg masses of *A. unangas* are nearly spherical in shape and were found deposited surrounding stalks of the octocoral *Primnoa* sp. (Fig. 2). Diameters of the egg masses measured 34.0–41.0 mm along the deposition axis and 36.3–42.5 mm along the perpendicular axis. Live coloration of the eggs varied from beige to light rose; the stalks of *Primnoa* sp. were a bright reddish-orange. After preservation in 5% formalin, the eggs changed to a pale yellow-green to light amber in color and the coral stalks turned white. The egg mass that was teased apart contained 267 eggs.

Description of individual eggs and embryos. Individual eggs of *Allocareproctus unangas* are slightly aspherical and noticeably flattened or have a slightly cup-shaped depression on the outer chorion at points of contact with adjacent eggs (Figs. 2, 3). Mean diameter in each axis is 5.7 × 6.3 mm (range 5.2–6.0 × 5.8–6.8, SD = 0.2, $n = 241$). The chorion is thick (0.15 mm), leathery, covered with a shallow, very fine honeycomb pattern, light amber in color, and semitranspar-

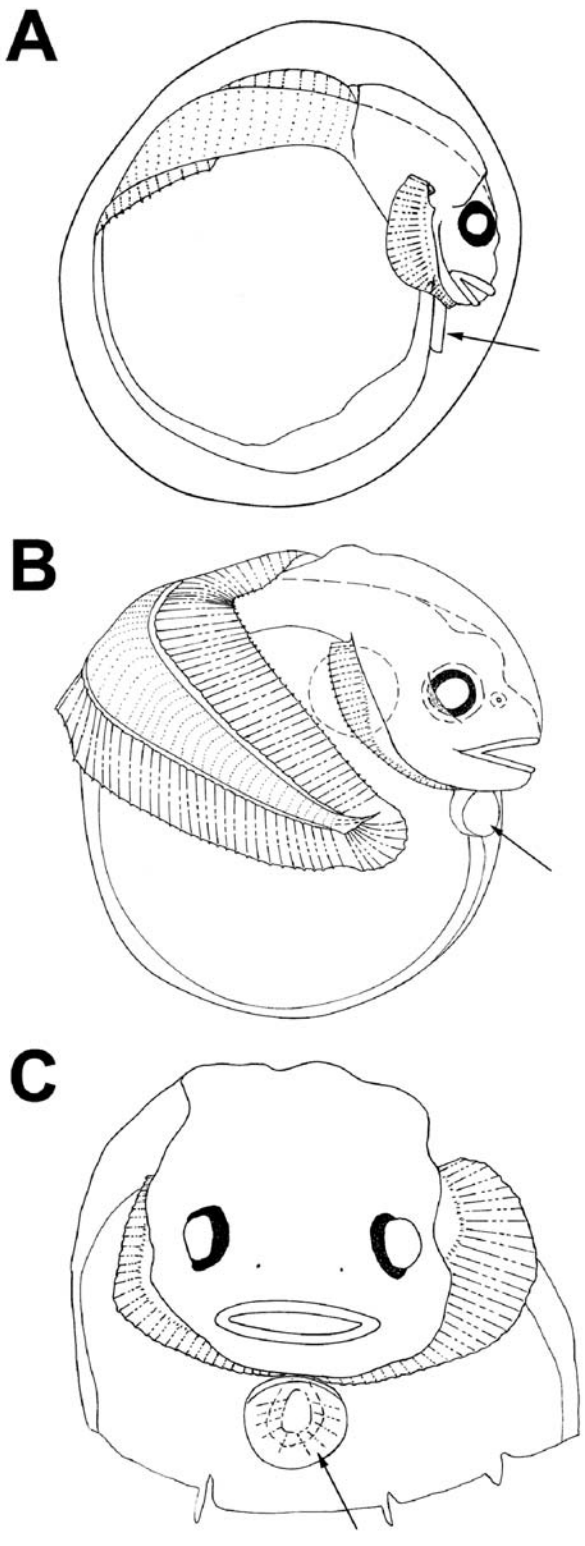

Fig. 3. A Lateral view of *Allocareproctus unangas* egg (from UW 094968 #1). *Arrow* points at ventral sucking disk. **B** Dissected embryo from egg of *Allocareproctus unangas* (from UW 094968 #1). *Arrow* points at ventral sucking disk. **C** Anterior view of embryo dissected from egg of *Allocareproctus unangas* (from UW 094968 #1). *Arrow* points at ventral sucking disk

ent (in preserved state). Eggs contained well-developed, late-stage embryos in middle to late stages of notochord flexion (Fig. 3A,B). The yolk mass is large, homogeneous, amber colored, and covered on its surface with a dense branching network of capillaries. The origin of the capillaries appears to be a large blood vessel originating on the ventral surface of the embryo posterior to the disk and extending ventrally through most of the yolk. Standard lengths (SL) of five embryos measured 7.7–8.7 mm ($\bar{x}$ = 8.3 mm). Embryos possess a large, well-developed disk (Fig. 3B,C) that appears to extend anteriorly from the isthmus over the surface of the yolk mass [disk length, 38.8% of head length (HL), 7.7% SL, $n = 5$]. A faintly outlined spherical structure, likely the gut in early stage of development, is present ventrally behind the head (Fig. 3B). Embryos and yolk masses lack melanistic pigmentation. Counts from seven cleared and stained embryos were compared to adult ranges of similar liparids (Table 1). Elements appeared to be stained only with Alcian blue and were thus considered unossified. Neural and haemal spines were well developed. Elements of the pectoral and caudal fins were not completely formed and contained the larval finfold with no rays present, and thus counts are below adult complements. Hypural bones of the caudal fin were well developed, and 6–10 unossified principal rays were present (adult complement: 1–3, 5 + 6–7, 1–2).

Remarks. This is the first report of eggs of a liparid in nature, other than species of *Liparis*, being deposited in a location other than the gill cavity of a lithodid crab (Love and Shirley, 1993; Somerton and Donaldson, 1998). This unique spawning behavior may likely be an additional character distinguishing *Allocareproctus* and *Careproctus*. *Careproctus reinhardti* was reported to lay its eggs in an aquarium between the wall and a stone or thallus of *Fuscus* (Chernova, 1987). However, this observation was not made under natural conditions, and lithodid crabs were not available as spawning substrate. Shallow-water liparids of the genus *Liparis* have been reported to lay eggs in empty bivalve shells (DeMartini, 1978), on hydroids (Ehrenbaum, 1905; Able and Musick, 1976), and among polychaete tubes and barnacle colonies (Marliave and Peden, 1989).

Another stalk of *Primnoa* sp. collected in the same general area as our specimens had three attached masses of two other unidentified fish eggs (UW 107604). One of these masses was very similar in appearance to that of *A. unangas*, but it contained slightly smaller eggs (4.1–4.6 × 5.0–5.4 mm) and no advanced-stage embryos. The eggs were similar in color and chorion texture and may have been deposited by another species of *Allocareproctus*, of which four others have been collected in the area (Orr and Busby, 2006). The other two masses were smaller in size and contained much smaller (2.5–2.8 mm) spherical eggs. One of these masses contained eggs with eyed embryos. After dissection and subsequent examination of body morphology of the embryos, we believe these to be of a cottoid fish, possibly a liparid, but we could not confirm the presence of a ventral sucking disk. These discoveries, along with recent observations of associations of juvenile rockfishes (*Sebastes* spp.) and other fishes and invertebrates with *Primnoa* sp.

(Heifetz, 2002; Kreiger and Wing, 2002), shed additional light on the importance of subarctic coral beds as critical habitat of marine fishes.

Material examined. *Allocareproctus unangas*, UW 094968, four egg masses deposited on the octocoral *Primnoa* sp., northeast of Seguam Island, Aleutian Islands, Alaska, 52.652° N, 172.251° W, bottom trawl, 26 June 1997, F/V *Dominator*, depth 397 m, collected by J.W. Orr. Other material of *Allocareproctus* cited in Orr and Busby (2006).

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