Abstract. *Cryptasterina hystera* has a highly derived life history with intragonadal development and juveniles that emerge from the parent’s reproductive tract. The gonads are ovotestes with developing eggs separated from sperm by follicle cells. *C. hystera* has typical echinosperm that must enter the gonoduct of conspecifics to achieve fertilization. During oogenesis, an initial period of yolk accumulation is followed by hypertrophic lipid deposition, the major contributor to the increase in egg size. 1-Methyladenine induces egg maturation and ovulation, but the spawning component of the hormonal cascade is suppressed. This is the major alteration in reproduction associated with evolution of viviparity in *C. hystera*. The switch to viviparity was not accompanied by major change in gonad structure, indicating there were few or no anatomical constraints for evolution of a marsupial function for the gonad. Despite their intragonadal habitat, the brachiolaria are equipped for a planktonic life, swimming in gonadal fluid. During the gastrula stage, lipid provisions are released into the blastocoel where they are stored for juvenile development. The eggs of *C. hystera* have light and dark cytoplasmic regions that mark animal-vegetal polarity. The dark pigment provided a marker to follow the fate of vegetal cells. Live birth is rare in the Echinodermata and the incidence of this form of brooding in the phylum is reviewed.

Introduction

A decoupling of selective forces on the larval and adult life stages of marine invertebrates has resulted in a remarkable diversity of life-history patterns. Markedly different larval stages within closely related species often contrast with the comparative similarity of the adults. This is exemplified by cryptic morphospecies, where new species have been discovered through observation of differences in life history, reproductive anatomy, or molecular sequence data (Reid, 1990; Knowlton, 1993; Degnan and Lavin, 1995; Ó Foighil and Smith, 1995; Byrne et al., 1999a, 2003a; Huber et al., 2000). Within major marine groups, some taxa (e.g., syllid polychaetes, littorinid snails, lasaeid bivalves, asterinid sea stars) show greater variation in life-history patterns than closely related taxa (Pocklington and Hutcheson, 1983; Reid, 1990; Ó Foighil and Smith, 1995; Byrne et al., 1999a). Why this is so is a major question for phylogeny and developmental evolution.

Within the Echinodermata, the Asterinidae, a major family of sea stars, is particularly noted for its diverse life histories (Byrne and Cerra, 1996; Hart et al., 1997, 2003, 2004; Byrne et al., 1999a). Species in the asterinid genera *Patiriella* and *Cryptasterina* exhibit a range of developmental modes, including a most derived method of propagation—incubation of progeny in the gonads and birth of juveniles (Byrne, 1996; Byrne and Cerra, 1996; Byrne et al., 2003a). In the Asterinidae, new species have been discovered by observation of developing stages in the gonads or birth of juveniles (Dartnall, 1969; Keough and Dartnall, 1978; Hart et al., 2003; Byrne et al., 2003a; Dartnall et al., 2003). These asterinids constitute a suite of recently diverged cryptic species in which some species exhibit as little as 1% difference in mtDNA sequence compared with congeners that have dispersive larvae (Hart et al., 2003; Byrne et al., 2003a).

This study documents reproduction and development in the most recently discovered viviparous species, *Cryptasterina hystera*, a member of the former pan-tropical *Patiriella pseudoelegia* group (Dartnall et al., 2003). Unexpectedly, *C. hystera* has a typical lecithotrophic brachiolaria
(Byrne et al., 2003a). By contrast, viviparous *Patiriella* species have a vestigial brachiolaria (Byrne and Cerra, 1996). Viviparity in *C. hystera* appears to have arisen from a free-spawning ancestor with a planktonic lecithotrophic brachiolaria larva through retention and intragonadal fertilization of a large egg (Byrne et al., 2003a). The gonad of *C. hystera* is an ovisac and serves as a marsupium for the developing embryos. Reproduction and development of *C. hystera* were examined in this study to determine which life-history traits are conserved and which traits have been modified in association with the evolution of viviparity. Sperm ultrastructure was examined for modifications associated with internal fertilization. The embryos of *C. hystera* are provisioned with a conspicuous store of lipid (Byrne et al., 2003a). Recent studies indicate that maternal lipid reserves are used to support juvenile development (Emlet and Hoegh-Guldberg, 1997; Byrne and Cerra, 2000; Villinski et al., 2002; Byrne et al., 2003b), and particular attention was paid to the fate of these reserves during development of *C. hystera*. The eggs of *C. hystera* have light and dark cytoplasmic regions that mark animal-vegetal polarity. The dark pigment was used as a marker to follow the fate of vegetal cells in development of larval territories.

### Materials and Methods

Mature specimens of *Cryptasterina hystera* were collected from Statue Bay (23°15′ S; 150°45′ E), Queensland, during the reproductive period September–December 1997 and 1999. Ova were obtained by placing gonad lobes that appeared to be entirely female in the ovulatory hormone 1-methyladenine (10⁻⁵ M in filtered seawater [FSW]). Eggs released from the gonad were fertilized by sperm from the same or a different specimen. The larvae were reared in FSW at 21 °C until metamorphosis.

For histology, the gonads were fixed in Bouin’s fluid for 24 h, rinsed in distilled water, dehydrated in graded ethanol, and embedded in paraffin. Serial sections were stained with hematoxylin and eosin. For light and transmission electron microscopy (TEM), gonads, larvae, and juveniles were fixed for 1 h at room temperature (RT) in 2.5% glutaraldehyde in 0.45 μm FSW, and then rinsed in 2.5% NaHCO₃ (pH 7.2). The specimens were post-fixed in 2.0% osmium tetroxide in 1.25% NaHCO₃ for 1 h at RT. Other specimens were fixed in 3% glutaraldehyde in 0.2 M cacodylate buffer (pH 7.4) with NaCl (30 mg/ml) added to the primary fixative for 2 h at 4 °C. This was followed by four rinses in 0.2 M cacodylate buffer with reduced amounts of NaCl added to each rinse, with the final rinse containing no NaCl. The specimens were post-fixed in 1.0% osmium tetroxide in 0.2 M cacodylate buffer for 2 h at 4 °C. Tissues fixed by both methods were rinsed in distilled water, dehydrated in graded ethanol, and embedded in Spurr’s resin. Semithin sections were stained with 1% toluidine blue for light microscopy. Ultrathin sections were stained with 2% uranyl acetate for 30 min and 2.0% lead citrate for 10 min. The sections were viewed with a Phillips EM400 transmission electron microscope.

### Results

#### Hermaphroditic gonad

The gonads of *Cryptasterina hystera* were a mosaic of oogenic and spermatogenic areas (Figs. 1A; 2B, D; 3A). As characteristic of asteroids (Chia and Koss, 1995), the gonad wall was formed by two tissue layers separated from each other by the genital coelom (Fig. 2D). The outer layer consisted of the outer peritoneum, a connective tissue layer, and the epithelial lining of the genital coelom (Fig. 2E). The inner layer also had three layers: the coelomic epithelium, a connective tissue layer, and the inner germinal epithelium (Fig. 2E). Both coelomic epithelia contained myoepithelial cells and occasional neurons. The inner connective tissue was the widest tissue layer in the gonad wall. It contained flocculent hemal fluid and occasional coelomocytes. The germinal layer contained somatic cells and developing gametes.

The eggs developed within a follicle formed by somatic cells (Fig. 2D–F). Follicle cells extended from the germinal epithelium to surround the egg, thereby separating them from adjacent sperm (Fig. 2E, F). Oogenesis in *C. hystera* was characterized by an initial period of yolk granule formation (Fig. 2A) in primary oocytes (x̄ = 25 μm diam; SE = 0.71 μm; n = 15), followed by lipid accumulation. The accumulation of lipid in larger oocytes, starting at a mean diameter of 48 μm (SE = 0.9 μm; n = 21), was responsible for the major increase in egg size (Fig. 2B, C). Yolk granules were interspersed with lipid droplets, and some granules were pushed towards the egg cortex (Fig. 2B). Fully grown eggs (440 μm diameter) were dominated by lipid. A vitelline layer and jelly coat surrounded the eggs, and cortical granules were dispersed along the oolemma (Fig. 2B, E, F). Lipid droplets were not present in the cortical cytoplasm (Fig. 2B). Although looked for, egg maturation and ovulation were not observed and ova were not found in sections.

Large eggs were distinctly marked by pale olive green and dark-colored cytoplasmic regions (Fig. 1A, B). The vegetal hemisphere (confirmed by site of gastrulation) was marked by dark pigment (Fig. 1E). A small dark spot at the animal pole marked the position of the germinal vesicle (Fig. 1A, C). When the gonads were dissected, the eggs floated to the surface, animal pole up (Fig. 1B). Ultrastructural examination of the eggs did not reveal any structures at the vegetal pole that might be connected with the accumulation of dark pigment.

Spermatogenesis was usually allocated to small regions of the germinal epithelium, although entire lobes of some gonads were devoted to sperm production (Fig. 1A). Sperm
development in spermatocyte columns was typical of asteroid spermatogenesis. Spermatozoa were scattered in clumps intermingled with developing oocytes or in small pockets along the gonad wall (Figs. 2A, D; 3A). They remained separated from the oocytes by the follicle layer. The spermatozoa had a spherical head with the acrosome in a depression in the nucleus and surrounded by periacrosomal material, a midpiece, and a flagellum (Fig. 2F, G). These features are typical of asteroid sperm. When placed in seawater, the sperm of *C. hystera* exhibited typical motility.

Figure 1. Light microscopy of *Cryptasterina hystera* gonads and embryos. (A–C) Ovotestes: The eggs have a dark vegetal region (V), and a dark spot marks the location of the germinal vesicle at the animal pole (arrowheads). The eggs float up from the dissected gonad (arrows). The gonad lobes are dominated by oocytes with occasional lobes filled with sperm (T). W, wrinkled blastulae. (D) Wrinkled blastulae. (E) Unhatched gastrula with black pigmented cells (arrow) streaming into the blastopore from one side of the embryo. The blastopore has a dark rim. (F) Hatched gastrula with dark pigment in vegetal hemisphere. Arrowhead, blastopore. (G–I) Brachiolaria larvae: The pigment remains in the posterior region. The lateral brachia (B) and hydroporic canal (H) are marked by darker cytoplasm (J) Gonad filled with near-term juveniles (Ju). (K) Moribund large juveniles dissected from gonad. Scales: A, C = 430 μm; B = 880 μm; D–F = 220 μm; G–I = 300 μm, J = 580 μm, K = 750 μm.
Figure 2. Light and transmission electron microscopy (TEM) of the ovotestis of Cryptasterina hystera (A) The early oocyte (E) contains a few yolk granules (arrow), and sperm (Sp) are present. (B–C) Advanced oocyte filled with lipid droplets (L). Yolk granules (Y) are dispersed among the lipid droplets and along the cortex. (D) Egg (O) and sperm (Sp) separated by the layer of follicle cells (F) around the egg. The arrowheads show the inner and outer tissue layers of the gonad wall. (E, F) TEM of gonad wall and oocyte (O). C, connective tissue layers; CG, cortical granule; E, epithelium of genital coelom; F, follicle cell; G, germinal epithelium; J, jelly coat; P, peritoneum; V, vitelline coat; Y, yolk granule; Arrow, phagocyte. (G) Sperm. A, acrosome. Scales: A, B, D = 50 μm; C = 100 μm; E = 1.5 μm; F = 2.0 μm, G = 0.4 μm.
Development

Embryos in all the gonads of individual *C. hystera* were usually at a similar stage of development, indicating that egg maturation, ovulation, and fertilization were synchronous (Figs. 1J; 3A, B, C). Among individual adults, however, the embryos were often at a different stage of development. Early in the season, cohorts of embryos developed alongside advanced gametes in the same gonad (Fig. 3A). By November and December, only a few unfertilized fully grown eggs remained in the gonad and were likely to be atretic. During these months, most gonads contained a clutch of juveniles (Fig. 1J).

Ovaries placed in 1-methyladenine (1-MA) released eggs, and many of these eggs were fertilized by sperm present in the gonad. Early cleavage was radial and holoblastic. The earliest embryos encountered in the gonad were wrinkled blastulae (Fig. 1C, D). Based on development in laboratory cultures, these embryos were about 12 h post-fertilization. Wrinkled blastulae had a highly contorted epithelium composed of cuboidal epithelial cells filled with large lipid droplets (Fig. 1C, D; 3C). The blastulae were enclosed in the fertilization envelope. The dark cytoplasm remained in the vegetal region throughout early development. In blastulae, it was confined to the lower third of the embryo. On removal from the gonad, blastulae floated at the air-water interface and continued development through the brachiolaria larval stage, metamorphosing into juveniles in 3 weeks.

The blastopore developed at the middle of the dark cytoplasm, about 24 h post-fertilization, and its rim was marked by dark pigment (Fig. 1E). During gastrulation, the dark cells around the blastopore did not move symmetrically into the embryo from around the margin of the opening (Fig. 1E). One side of the embryo appeared to contribute more dark cells to the developing archenteron than the other. As the larva elongated, dark pigmented cells moved anteriorly along the ventral surface and, to a lesser extent, the dorsal surface (Fig. 1F–H). The pigment did not extend beyond the midventral region. Although the distribution of the dark cytoplasm was variable among larvae, cells inheriting the dark pigment always remained in the posterior region of the larva. In many larvae there was a contrast between the light anterior preoral lobe and the dark posterior region at the level of the brachiolar apparatus (Fig. 1I). The central brachium was light green, while the adjacent posterior body region was dark olive. On the dorsal side, dark cytoplasm marked the hydroporic canal (Fig. 1I).

The embryos hatched as late gastrulae at 40 h and the blastopore closed (Figs. 1F; 3A). Gastrulae and larvae swam propelled by their uniform cover of cilia. Their locomotion in gonad fluid depended on the availability of space. During the gastrula stage, lipid droplets were extruded into the blastocoel (Figs. 3D; 4A–C). Prior to extrusion, the epithelial cells changed from cuboidal to columnar, and lipid droplets were shunted to a basal location below the nucleus (Figs. 3D; 4A, B). This basal shunting was a prelude for lipid release. Lipid droplets were released into the blastocoel by an apocrine mechanism, taking part of the cell membrane with them (Figs. 3E; 4B, C). The lipid remained in the blastocoel throughout development (Figs. 3E; 4D).

Formation of the brachiolar attachment complex was first seen on external view in 5-day-old larvae as three protrusions at the anterior end (Fig. 1G). These were the developing brachia. By this stage the larvae swam at the bottom of the dish anterior end up, buoyed by the abundant lipid reserves in the blastocoel of this region of the larva. As the brachiolaria developed, the attachment complex became a prominent feature of the larva. The adhesive disc developed at the base of the brachia (Fig. 4D). Both the adhesive disc and brachiolar arms contained batteries of secretory cells that were presumably the source of adhesive material. Advanced larvae readily adhered to the substratum using their brachia and, prior to metamorphosis, attached to the surface of the culture dishes with the adhesive disc. The juvenile rudiment developed in the posterior region (Fig. 1H, I). Newly metamorphosed juveniles (600 μm in diameter, two pairs of tube feet per radius) from laboratory cultures had an amber hue due to their abundant lipid reserves. These reserves were mobilized during the perimetamorphic period. In sections of juveniles with a newly differentiated gut and well-developed skeleton, the remaining lipid droplets were seen scattered in the body wall (Fig. 3G).

Juveniles brooded by *C. hystera* appear to leave the gonad in a synchronous manner, as indicated by the appearance of hundreds of juveniles (800 μm diameter) in aquaria over a few hours. These juveniles had two pairs of podia in each radius. Some juveniles (1–2 per gonad) remained in the gonads for a longer time (Figs. 1K; 3F). These juveniles were large (1–4 mm diameter) and had four to six pairs of tube feet per radius. How they attained this size in the gonads is not known, but the presence of adjacent moribund juveniles indicated that they cannibalize their siblings. They may also utilize eggs as a source of food. The significance of this phenomenon is not clear because the presence of degenerating juveniles in the gonads indicates that at least some of them were unable to emerge from the gonad (Fig. 1K).

Discussion

Internal fertilization, intragonadal development, and live birth of juveniles is a rare form of propagation in marine invertebrates. Independent dispersive stages are completely deleted from the life history. In echinoderms, intragonadal incubation of embryos is known for one crinoid, four ophiuroids, four holothuroids, and five asteroids (Table 1). In addition, some dendrochirotid, psolid, and apodid sea
Figure 3. Light microscopy of Cryptasterina hystera gonads and embryos. (A, B) Gastrulae (G), brachiorlaria larvae (B), oocytes (O), and spermatocyte columns (SC) in the gonads. (C) Wrinkled blastula with lipid droplets in cuboidal epithelial cells. FE, fertilization envelope. (D) Early gastrulae prior to extrusion of lipid (L). (E) Larva with lipid (L) in blastocoel. Few lipid reserves remaining in the epithelium (Ep). (F) Large juvenile (J), eggs (O), and embryo (E) in gonad. (G) Thin section of fully developed juvenile prior to the onset of feeding. The section extends from the stomach (St) to the edge of the body and shows lipid droplets (L) remaining in the body wall. WV, water vascular system; Am, ampulla of tube foot. Scales: A, B = 125 μm; C = 45 μm; D, E = 13.5 μm; F = 500 μm; G = 40 μm.
cucumbers incubate their offspring in the coelom and juveniles emerge from the adult body (Reviews: Vaney, 1925; McEuen, 1986; O’Loughlin, 1994). Although live birth is rare in echinoderms, it appears that the potential for making the switch to this mode of propagation is more common in some taxa than others. The Holothuroidea contains the highest number of viviparous species (O’Loughlin, 1994). Aside from the aberrant Xyloplax medusiformis, all viviparous asteroids are in the Asterinidae (Table 1).

As is characteristic of viviparous asterinids, the gonads of Cryptasterina hystera were ovotestes (Komatsu et al., 1990; Byrne, 1996). This condition is also reported in viviparous holothuroids (Miller, 1985; Frick et al., 1996). Despite their intragonadal location, the sperm of C. hystera had a well-developed tail, were fully motile, and could fertilize eggs in vitro. Their ultrastructure was the asteroid “echinosperm-type” (sensu Jamieson, 1985) with a round head and embedded acrosome. It is likely that these sperm would be functional if released into the sea. For outcrossing to occur in C. hystera and other viviparous echinoderms, sperm

| \textbf{Table 1.} | \textbf{Taxon} | \textbf{Order} | \textbf{Family} | \textbf{Viviparity} |
|-------------------|--------------|---------------|-----------------|-------------------|
| Asterinidae        | Asterinidae  | Asterinidae   | Viviparous      | Viviparous        |
| Holothuroidea      | Holothuroidea | Holothuroidea | Viviparous      | Viviparous        |
| Xyloplax medusiformis | Xyloplax medusiformis | Xyloplax medusiformis | Viviparous | Viviparous |

Figure 4. Light and transmission electron microscopy of Cryptasterina hystera. (A–C) Lipid reserves (L) are shunted basally below the nucleus (Nu) of the cells in the gastrula epithelium (Ep) before secretion into the blastocoel (Bc). The lipid is released by an exocrine mechanism, often taking a portion of the cell membrane (arrow). (D) Brachiolaria with lipid droplets (L) filling the blastocoel. A, adhesive disc; B, brachia; St, stomach. Scales: A = 3.0 µm; B = 1.5 µm; C = 0.15 µm; D = 85 µm.
would have to gain access to eggs by swimming through the gonopore, an activity that would require sperm chemotaxis. Sperm chemoreception and sperm-activating pheromones have been documented for several echinoderms, including asteroids (Miller, 1989; Miller and Vogt, 1996). Asterinids with benthic development gather to lay communal egg masses, with sperm deposited directly onto the eggs (Tominaga et al., 1994; Byrne, 1995). Sperm exchange in *C. hystera* would be facilitated by similar mating behavior. Hystera (Tominaga masses, with sperm deposited directly onto the eggs with benthic development gather to lay communal egg asteroids (Miller, 1989; Miller and Vogt, 1996). Asterinids have been documented for several echinoderms, including gonopore, an activity that would require sperm chemotaxis. would have to gain access to eggs by swimming through the basal cytoplasm of the epithelium, as seen in *C. hystera* and other viviparous holothuroids (Frick, 1998). From an ancestral state involving planktotrophic development and small eggs dominated by yolk protein (Byrne et al., 1999b, 2003b), the increase in egg size in *C. hystera* was associated with a shift to lipid-dominated eggs. An increase in lipid reserves is characteristic of echinoderms with lecithotrophic development (Emlet et al., 1987; Jaeckle, 1995). The initial phase of oogenesis during which yolk protein is sequestered into yolk granules is typical of the ancestral-type oogenic pattern (Byrne et al., 1999c, 2003b). This is followed by hypertrophic lipid deposition, similar to that seen in the lipid-rich eggs of the sea urchin *Heliocidaris erythrogramma* (Byrne et al., 1999c). Possession of a buoyant egg in *C. hystera* and its congener *C. pacifica* was not expected in species with intragonadal development. Development in *C. hystera* and *C. pacifica* is completely supported by maternal provisioning in the egg. In contrast, *Patiriella vivipara* and *P. parvivipara* have small (120-μm diameter), secondarily reduced negatively buoyant eggs with minimal lipid stores (*P. parvivipara* is completely supported by maternal provisioning in the egg. In contrast, *Patiriella vivipara* and *P. parvivipara* have small (120-μm diameter). In contrast, echinoderms that brood embryos in non-gonadal structures often have specializations for transferring nutrients to developing young (Walker and Lesser, 1989; McClary and Mladenov, 1990). *C. hystera* appears to have no anatomical impediments for acquisition of a marsupial function for the gonads. Considering this lack of morphological constraint, it is surprising that a viviparous life history is not more common in asteroids. Most of the nutritive provisions loaded into the egg of *C. hystera* are reserved for the postlarval perimorphomorphic stage. Early development is burdened by excess nutritive reserves not required for embryogenesis. Shunting of lipid into the basal cytoplasm of the epithelium, as seen in *C. hystera*, is common in lipid-rich echinoderm embryos (Patent, 1968; Henry et al., 1991; Cerra and Byrne, 1995; Byrne and Cerra, 2000; Byrne et al., 2003b). This is suggested to be a mechanism for partitioning excess reserves...
away from the active apical region of the cell (Cerra and Byrne, 1995). In C. hystera, most lipid reserves are extruded into the extracellular blastocoelic space at the gastrula stage by an apocrine mechanism, similar to that documented for the sea urchin *Helioicidaris erythrogramma* (Henry et al., 1991). The blastocoel thus functions as a storage space for nutrients to support juvenile development. Lipid extrusion is now reported for the embryos of two echinoderm species, both of which have highly buoyant, lipid-rich eggs. Such extrusion is the extreme outcome of the basal shunting process and may be associated with the presence of a particular class of lipid.

The presence of differently pigmented cytoplasm, as seen in the eggs of *C. hystera*, has not been reported for asteroids but is known for one echinoid, *Holopneustes purpurascens*. The animal and vegetal hemispheres of the eggs of *C. hystera* could be discerned by light and dark pigment, respectively. This is similar (but opposite) to the situation in the eggs of *H. purpurascens*, where the animal pole has dark yolk and the vegetal pole has light yolk (Morris, 1995). In that urchin, dark pigment is segregated into one blastomere at the 2-cell stage. In *C. hystera*, by contrast, the pigment remained in the vegetal region, with the major cellular segregation occurring at the third equatorial cleavage that separates cells into animal and vegetal fates. The dark cytoplasm remained in the lower third of the embryo, and the blastopore appeared where the dark pigment was most concentrated. During gastrulation, it appeared that one side of the embryo contributed more dark cells to the developing archenteron than the other. The mechanism of gastrulation in *C. hystera* may differ from the radially symmetrical pattern of involution traditionally attributed to echinoderms. Asymmetrical cell movements during gastrulation have been reported for the lecithotrophic embryos of the sea urchin *Helioicidaris erythrogramma* (Wray and Raff, 1994) and also occur in the lecithotrophic embryos of *Patiriella exigua* (Byrne and Cerra, unpubl.). More studies are required to determine whether gastrulation by asymmetrical invagination of cells from the right and left sides of the embryo is a feature of lecithotrophic development in echinoderms.

After closure of the blastopore, dark-pigmented cells that remained evident on the surface view migrated ventrally. This is consistent with migration of the blastopore region to the ventral side of planktotrophic larvae that have a functional anus. In many larvae, the dark cells formed a boundary between the preoral-postoral regions on the ventral surface, at the level of the brachiolar apparatus. The pigment may thus mark the anterior and posterior cellular domains that in feeding larvae would be bordered by the preoral and postoral ciliated bands. This is reminiscent of the oral and aboral territories marked by the ciliary band in sea urchin plutei and for which patterns of gene expression have been determined (Davidson et al., 1998). On the dorsal side, dark pigment was also present in the posterior region, and the hydropore was strongly marked by dark pigment. The animal-vegetal pigment in the eggs of *C. hystera* has potential for use in cell lineage studies.

The presence of a functional brachiolaria in the gonads of *C. hystera* and *C. pacifica* indicates that these sea stars have the potential to brood and broadcast their young—an unusual form of poecilogony that has been reported for the sea star *Pteraster militaris* (McClary and Mladenov, 1988). The highly restricted distribution of *C. hystera* indicates, however, that the brachiolaria are not used for dispersal in nature and that the progeny are likely to leave the parent as juveniles (Byrne et al., 2003a). In aquaria, only juveniles were released. In general, the biogeography of asterinid sea stars correlates with the presence or absence of a dispersive larva; the exception is the benthic developer *Patiriella exigua*, which occurs around the southern hemisphere (Waters and Roy, 2004; Colgan et al., 2005). Viviparous asterinids have the most restricted distribution known for the Asteroida (Byrne et al., 1999a, 2003a). Three of these occur in Australia, and their distributions range from 50 km (three locations, *C. hystera*) to 100 km of coastline (seven locations, *P. vivipara*). It would be interesting to know the distribution of *C. pacifica*, the fourth viviparous asterinid, in Japan. The mangrove habitats where *C. hystera* is found are vulnerable to anthropogenic change, highlighting the need to determine the distribution of this new viviparous species.

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