



Phototactic responses of larvae from the marine sponges *Neopetrosia proxima* and *Xestospongia bocatorensis* (Haplosclerida: Petrosiidae)

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Abstract. Previous studies suggest that phototaxis in sponge larvae is generated by the bending of a tuft of long posterior cilia (LPC). The photoresponsiveness of these cilia is often assayed by examining their reaction to sudden changes in light intensity. Here, we document and describe the larvae of the tropical marine sponges *Neopetrosia proxima* and *Xestospongia bocatorensis* and examine the phototactic behavior of their larvae. Both species brood ovoid, tufted parenchymella larvae, clearly countering an earlier hypothesis that all petrosid sponges are oviparous. Larvae of *N. proxima* were positively phototactic and settled after 2 d, while larvae of *X. bocatorensis* were negatively phototactic and settled in as little as 4 h. In both species, LPC quickly responded to changes in the light intensity. When the light intensity is reduced, the larvae of *N. proxima* fold the cilia inwards immediately without beating, then flare them outwards, beating for a few seconds, and then gradually return to the neutral position while continuing to beat. In contrast, the larvae of *X. bocatorensis* flare the cilia outwards when the light intensity is reduced and fold them inwards when the light intensity is increased. Comparisons with reported ciliary responses to light for other species demonstrate that these responses do not show the hypothesized one-to-one correspondence with phototactic behaviors and are, therefore, of limited use in explaining the mechanisms that coordinate larval swimming.

Additional key words: Porifera, parenchymella larvae, larval ecology

Although relatively little is known about the behavior of sponge larvae compared with the larvae of other marine invertebrates, it is clear that light plays an important role in their ecology. Light can be a key trigger for larval release either via the onset of darkness before release (Amano 1986), via increased light intensity the day before release (Amano 1988), or via intense light exposure (Maldonado & Young 1996). Light can also influence the choice of settlement sites, with most species showing either a preference for shaded sites or no preference at all. In field studies, increased recruitment of *Tedania ignis* (DUCHASSAING & MICHELOTTI 1864) and *Crambe crambe* (SCHMIDT 1862) was observed on shaded areas of settlement tiles, but no difference in the recruitment of

Sigmadocia caerulea (HECHTEL 1965) was observed between shaded and unshaded substrates (Maldonado & Young 1996; Maldonado & Uriz 1998).

Settlement site preferences could be mediated by phototactic larval swimming. There is significant diversity among sponge species in phototactic behavior: of the 88 species with documented phototaxis, 25 display positive phototaxis, 41 show no clear response to light, and 22 are negatively phototactic (Table 1; Wapstra & Van Soest 1987). In addition, phototactic behavior can change during the larval lifespan. For example, larvae of the dictyoceratid *Cacospongia mollior* SCHMIDT 1862 are positively phototactic when they are released from the parent, but become negatively phototactic after 4–6 h (Maldonado et al. 2003).

The mechanisms underlying phototaxis in sponge larvae, which lack neural networks and gap junctions, are not fully understood (Leys & Degnan 2001;

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Table 1. A summary of phototaxis (+, none, or –) and ciliary responses to increased light intensity in sponge larvae. These data are from studies published subsequent to the compilation of similar data by Wapstra & Van Soest (1987). Of these 28 species, seven display positive phototaxis, seven show no clear phototactic response, and 14 are negatively phototactic. Wapstra & Van Soest (1987) provided phototactic response data for an additional 60 taxa; when considering all 88 species, 25 display positive phototaxis, 41 show no clear response, and 22 are negatively phototactic. NA = not available.

Species	Phototaxis	Ciliary response	References
Dendroceratida			
<i>Chelonaplysilla noevus</i>	–	NA	Mariani et al. (2005)
Dictyoceratida			
<i>Cacospongia mollior</i> ^a	+/–	Extend then fold	Maldonado et al. (2003)
<i>Dysidea avara</i>	+	NA	Mariani et al. (2005)
<i>Hippospongia lachne</i>	–	NA	Kaye & Reiswig (1991)
<i>Ircinia oros</i>	–	Fold	Maldonado et al. (2003)
<i>Luffariella variabilis</i>	None/–	NA	Ettinger-Epstein et al. (2008)
<i>Pleraplysilla spinifera</i>	+	NA	Mariani et al. (2005)
<i>Rhopaloeides odorabile</i>	+	NA	Whalan et al. (2008)
<i>Spongia (Spongia) barbara</i>	–	NA	Kaye & Reiswig (1991)
<i>Spongia (Spongia) graminea</i>	–	NA	Kaye & Reiswig (1991)
<i>Spongia (Spongia) officinalis</i>	–	NA	Gaino et al. (2007)
Hadromerida			
<i>Cliona viridis</i>	None	NA	Mariani et al. (2005)
Halichondrida			
<i>Halichondria magniconulosa</i>	–	NA	Maldonado & Young (1996)
<i>Halichondria melanodocia</i>	+	NA	Woollacott (1990)
<i>Scopalina lophyropoda</i>	None	NA	Mariani et al. (2005)
Haplosclerida			
<i>Amphimedon queenslandica</i>	–	Extend	Leys & Degnan (2001)
<i>Haliclona</i> sp.	+	NA	Mariani et al. (2005)
<i>Haliclona (Reniera) tubifera</i>	–	NA	Maldonado & Young (1996)
<i>Haliclona (Soestella) caerulea</i>	–	Extend	Maldonado & Young (1996), Maldonado et al. (2003)
<i>Neopetrosia proxima</i>	+	Extend then fold	This study
<i>Niphates digitalis</i>	–	NA	Lindquist et al. (1997)
<i>Xestospongia bocatorensis</i>	–	Fold	This study
Homosclerophorida			
<i>Oscarella</i> sp.	None	NA	Mariani et al. (2005)
Poecilosclerida			
<i>Clathria (Thalysias) jolicoeuri</i>	None	NA	Mariani et al. (2005)
<i>Crambe crambe</i>	None	NA	Mariani et al. (2005)
<i>Phorbas tenacior</i>	–	NA	Mariani et al. (2005)
<i>Tedania (Tedania) ignis</i>	–	NA	Maldonado & Young (1996)
Verongida			
<i>Aplysina aerophoba</i>	None	NA	Maldonado (2009)

^a*C. mollior* is photopositive at release but subsequently becomes photonegative.

Maldonado et al. 2003). Negative phototaxis in the larvae of *Amphimedon queenslandica* HOOPER & VAN SOEST 2006 (identified as *Reniera* sp. in Leys & Degnan (2001)) is thought to be conferred by a shadow response in which the long posterior cilia (LPC) straighten in response to an increase in the light intensity and bend in response to a decrease in the light intensity. The negatively phototactic larvae of *Ircinia oros* (SCHMIDT 1864) and *C. mollior* show the opposite response: the LPC fold inwards clockwise when

exposed to sudden increases in light and flare outwards when the light is reduced below 50 $\mu\text{mol photons m}^{-2}\text{s}^{-1}$ (Maldonado et al. 2003). The prevailing hypothesis, based on these observations, is that when light is directed from one side of the larva, the LPC bend in response to shading, resulting in an angle between the ciliary tuft and the body axis of the larva. Drag from this displacement of the LPC, combined with the clockwise rotation of the larva, causes the larva to turn away from the light (“Maldonado

model" hereafter; Maldonado et al. 2003). However, this is a largely intuitive model that has not been quantitatively examined. Given the low Reynolds numbers experienced by these tiny larvae, the relationship between the orientation of the LPC and the direction of larval movement may well be counterintuitive.

Because ciliary responses to light have been reported for several negatively phototactic but only one positively phototactic species of sponge larvae, observations of additional positively phototactic larvae could shed some light on the mechanisms underlying this behavior. *Neopetrosia proxima* (DUCHASSAING & MICHELOTTI 1864) (Petrosiidae, Haplosclerida; previously *Xestospongia*) is a common sponge on Caribbean reefs. In the Bocas del Toro region of Panama, it inhabits shallow (1–10 m deep) patch reefs and seagrass beds (Diaz 2005). Individuals of *N. proxima* are associated with symbiotic, unicellular cyanobacteria, and their nutrition may be partially derived from these photosymbionts (Erwin & Thacker 2007). *Xestospongia bocatorensis* DIAZ ET AL. (2007) (Petrosiidae, Haplosclerida) is found less commonly, growing in small patches on shallow (0–15 m deep) reefs, coral rubble, and mangrove roots; adults of this species host symbiotic, filamentous cyanobacteria (Thacker et al. 2007). Although larvae of many well-studied aposymbiotic species prefer to settle on shaded substrates and in crevices (Wapstra & Van Soest 1987; Maldonado & Young 1996), well-illuminated sites might be advantageous for the establishment of settlers and recruitment of juveniles of symbiotic species; therefore, we hypothesized that their larvae might be positively phototactic. The objectives of this study were to (1) describe the larvae of *N. proxima* and *X. bocatorensis*, (2) determine whether the larvae of both species are positively phototactic, (3) observe settlement preferences with respect to light, and (4) document changes in the ciliary position in response to changes in the light intensity.

Methods

Portions of adults of *Neopetrosia proxima* were collected by snorkeling at the base of the mangroves at STRI Point (9°21.10N, 82°15.57W), and adults of *Xestospongia bocatorensis* were collected at Caracol Point (9°22.66N, 82°18.19W) on Isla Colon in Bocas del Toro, Panama in August 2007 and 2009. Fragments of sponges ~1 cm³ in size that contained brood chambers with larvae were placed in a dish of seawater, with light from a fiber optic illuminator directed at the side to stimulate release of the larvae.

Immediately upon release, larvae were collected into a small plastic beaker and their phototactic behavior was observed by shining a fiber optic light on one side of the beaker. In three separate trials, ten (trial 2) or 15 (trials 1 and 3) freshly released *N. proxima* larvae were transferred to each of six replicate Petri dishes containing 30 mL of filtered seawater. Half of each dish was covered with black plastic, and the uncovered side was illuminated with a 150-W fiber optic lamp. To record phototactic behavior, larvae were distributed haphazardly by quickly swirling the dish; the number of larvae in the lighted area of each dish was counted after 5 min and 1, 3, 8, 20, 24, 31, 43, 55, and 67 h for *N. proxima*. For *X. bocatorensis*, 15 larvae were transferred to each of six replicate Petri dishes and 13 larvae to each of two dishes, all in a single trial; the number of larvae in the lighted area of each dish was counted after 5 min, and 1 and 4 h. When these animals had all settled, the dish was uncovered and the number of settlers on each side was counted. A similar, preliminary experiment with more frequent observations was conducted with *N. proxima* in August 2007, using four replicate dishes in a single trial. In this case, the dishes were examined after 0, 1, 3, 6, 9, 12, 18, 24, 36, and 48 h. The percentages of larvae in the lighted area of the dishes were compared among time periods, among trials, and among dishes within trials using repeated-measures ANOVA (factors: time, trials, and dishes within trials) with repeated measures on the time factor. The percentages of larvae that settled in the lighted area of the dishes were compared with a random distribution (50% in light) using a one-sample t-test.

To determine the response of the LPC to abrupt changes in light, freshly collected larvae were placed in a drop of seawater on a microscope slide. Photoresponsiveness of the LPC was observed while the intensity of light was rapidly increased or rapidly decreased using the diaphragm of the condenser, following the protocol of Maldonado et al. (2003). The response of the LPC to rapid changes in the light intensity is simple to observe and has been interpreted in previous studies to reflect the normal response of these cilia to light gradients (Maldonado et al. 2003). A total of ten larvae of each species were observed; *N. proxima* was observed in 2007 and 2009, while *X. bocatorensis* was only observed in 2009.

Results

The larvae of *Neopetrosia proxima* were type IIIa ciliated parenchymella larvae (Fig. 1; Mariani et al. 2005). They were slightly elongated ovoids with bare regions at the anterior and posterior poles, with

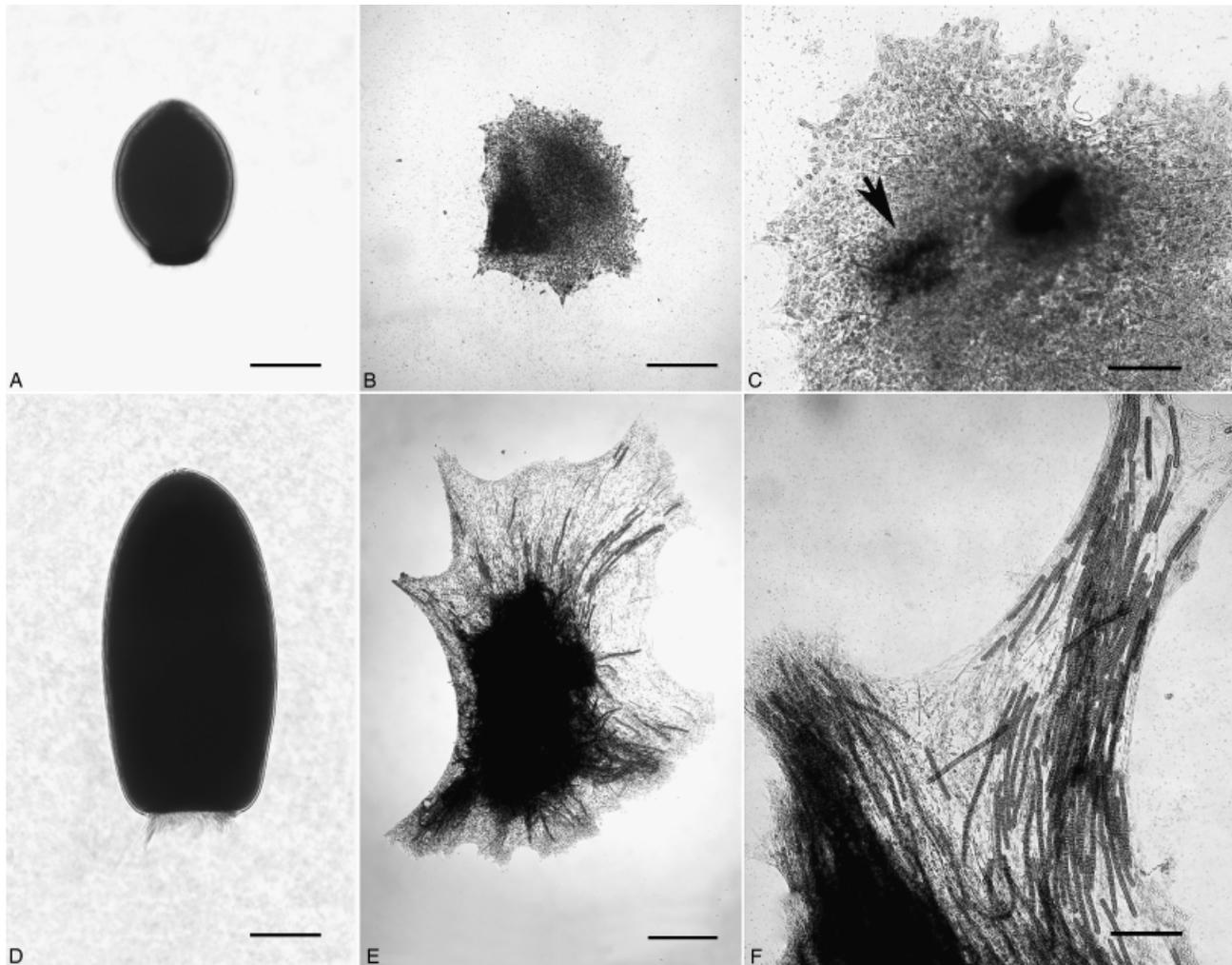


Fig. 1. Larvae and newly settled juveniles of *Neopetrosia proxima* and *Xestospongia bocatorensis*. **A.** Ovoid, type IIIa ciliated parenchymella larva of *N. proxima*, with a pigmented ring of long posterior cilia. Scale bar = 150 μm . **B.** Newly settled juvenile of *N. proxima*. Scale bar = 150 μm . **C.** Higher magnification of a juvenile of *N. proxima*. Individual spicules are visible, but unicellular photosymbionts are too small ($< 1 \mu\text{m}$) to visualize at this magnification. The arrow indicates the remnants of the posterior pigment ring. Scale bar = 125 μm . **D.** Bullet-shaped, type IIIa ciliated parenchymella larva of *X. bocatorensis*, displaying a ring of LPC. Scale bar = 150 μm . **E.** Newly settled juvenile of *X. bocatorensis*. Scale bar = 400 μm . **F.** Higher magnification of a juvenile of *X. bocatorensis*. Many long filaments of a cyanobacterial symbiont, *Oscillatoria spongeliae*, are visible. Scale bar = 100 μm .

an overall cream color and a dark reddish-brown pigment ring at the posterior pole. Newly released larvae were a mean of 367 μm (SD = 22.3 μm ; $n = 10$) in length and 284 μm (SD = 11.7 μm ; $n = 10$) in width. The longer posterior cilia arising from or adjacent to the pigment ring were 67 μm (SD = 9.3 μm ; $n = 19$) in length and very distinct from the 19 μm (SD = 5.3; $n = 5$) long cilia that covered the rest of the body. Larvae swam actively toward the light immediately after release but became noticeably less active after 6 h. After only 12 h, they frequently attached to the substrate at the anterior pole, and they metamor-

phosed after ~ 48 h. The settlers (Fig. 1) were 1.3 mm (SD = 0.33; $n = 6$) across.

Adults of *Xestospongia bocatorensis* also produced type IIIa ciliated parenchymella larvae, but these larvae were longer than those of *N. proxima*, with a rounded anterior end and a planar posterior end (i.e., bullet-shaped; Fig. 1). They were solid dark purple, as is the adult sponge, and had much shorter posterior cilia relative to their body length than did larvae of *N. proxima*. They were a mean of 824 μm (SD = 38.5 μm ; $n = 10$) long and 430 μm (SD = 60.8 μm ; $n = 11$) wide, with posterior cilia

81.7 μm (SD = 13.2 μm ; $n = 22$) in length. Larvae swam actively and strongly away from light upon release, but became less active quickly; all larvae of *X. bocatorensis* used in our experiments settled within 5 h of release. However, some larvae that were not used in the experiment failed to settle in such a short time; these individuals could still swim actively when provoked after 48 h. The settlers had irregular shapes, with some roughly round animals and others with long, string-like morphologies; round individuals were ~ 2 mm across (Fig. 1).

In 2007, counts of larvae of *N. proxima* in Petri dishes revealed a significant change in the percentage of larvae in lighted areas over time (Fig. 2; $F = 3.031$, $df = 9$, $p = 0.034$), with no significant container effect ($F = 2.839$, $df = 3$, $p = 0.080$). In this case, larvae were initially strongly positively phototactic (with 100% of the larvae in the experiment found in lighted areas), but became less positively phototactic over time, resulting in a photoneutral distribution (43% in light after 48 h; one-sample t-test against a random distribution: $t = 3.00$, $df = 1$, $p = 0.205$).

In 2009, counts of larvae of *N. proxima* in the lighted areas of 18 dishes (six dishes with ten larvae per dish, and 12 with 15 larvae per dish) showed that larvae were most often observed in the lighted areas (70% of the total number of larvae in the experiment; Fig. 2). Significantly more larvae settled in the lighted areas than in the shaded areas (79% vs. 21%; one-sample t-test against a random distribution: $t = 5.159$, $df = 17$, $p < 0.001$). The percentage of larvae in the lighted areas increased during the trials ($F = 4.631$, $df = 10$, $p < 0.001$), most significantly in the first hour (Bonferroni-adjusted $p = 0.014$), with no significant differences among subsequent time periods. Significant variability was observed among the 3 d during which trials were conducted ($F = 6.432$, $df = 2$, $p = 0.002$) and among replicate dishes within each day ($F = 6.073$, $df = 15$, $p < 0.001$), likely due to variability in the experimental lighting conditions. Larvae were observed to remain in the same spot for ≤ 24 h without metamorphosing and the number of these stationary larvae increased with time, resulting in the stability of counts in later time periods.

Counts of larvae of *X. bocatorensis* in the lighted areas of eight dishes (six each containing 15 larvae and two each containing 13 larvae) verified our preliminary observations that these larvae are negatively phototactic. The larvae were most frequently observed in the shaded areas (84% of the total number of larvae in the experiment; Fig. 2). During our initial counts (5 min after beginning the assay), the larvae were strongly negatively phototactic, with only 5% of the larvae in light; after 1 h, significantly more

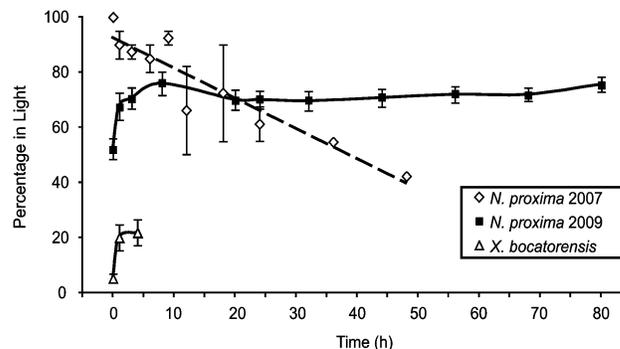


Fig. 2. Percentage (mean \pm SE) of sponge larvae in the illuminated half of the experimental chambers. Open diamonds represent *Neopetrosia proxima* tested in 2007, filled squares represent *N. proxima* tested in 2009, and open triangles represent *Xestospongia bocatorensis*. Note that some error bars are smaller than the symbols used here. For *N. proxima* in 2007, the dotted line represents a linear regression that fits a decline in the positive phototaxis over time ($r^2 = 0.884$, $p < 0.001$). Larvae of both *N. proxima* in 2009 and *X. bocatorensis* also showed significant changes over time ($p < 0.001$ and $p = 0.008$, respectively); for each, the initial values were significantly different from subsequent values, which were then not significantly different from each other.

(20%; Bonferroni-adjusted $p = 0.026$) of the larvae were in the lighted area, yielding a significant effect of time ($F = 7.011$, $df = 2$, $p = 0.008$). The percentage of larvae in lighted areas did not significantly change between 1 and 4 h (Bonferroni-adjusted $p = 1.00$). Those larvae that were in the illuminated side did not cluster near the fiberoptic light, in contrast to the larvae of *N. proxima*. We observed no significant container effect ($F = 1.943$, $df = 7$, $p = 0.137$). Significantly more larvae settled in the shaded than in the lighted areas (74% vs. 26%; one-sample t-test against a random distribution: $t = 3.940$, $df = 7$, $p = 0.006$).

The LPC in both species responded to changes in the light intensity, with the LPC response of larvae of *N. proxima* being more complex than that of the larvae of *X. bocatorensis*. In 2007, preliminary observations showed that larvae of *N. proxima* responded to reduced light intensity by flaring the cilia outwards (Fig. 3A); the larvae folded the cilia inwards when the light intensity was increased (Fig. 3C). More detailed observations in 2009 showed that when the light intensity is reduced, the cilia fold for a fraction of a second before they flare for 2–3 s, beating and gradually returning to the neutral position (Fig. 3B). In response to an increase in the light intensity, the cilia flare very rapidly, and then quickly fold inwards and begin to beat. In both cases, the immediate response

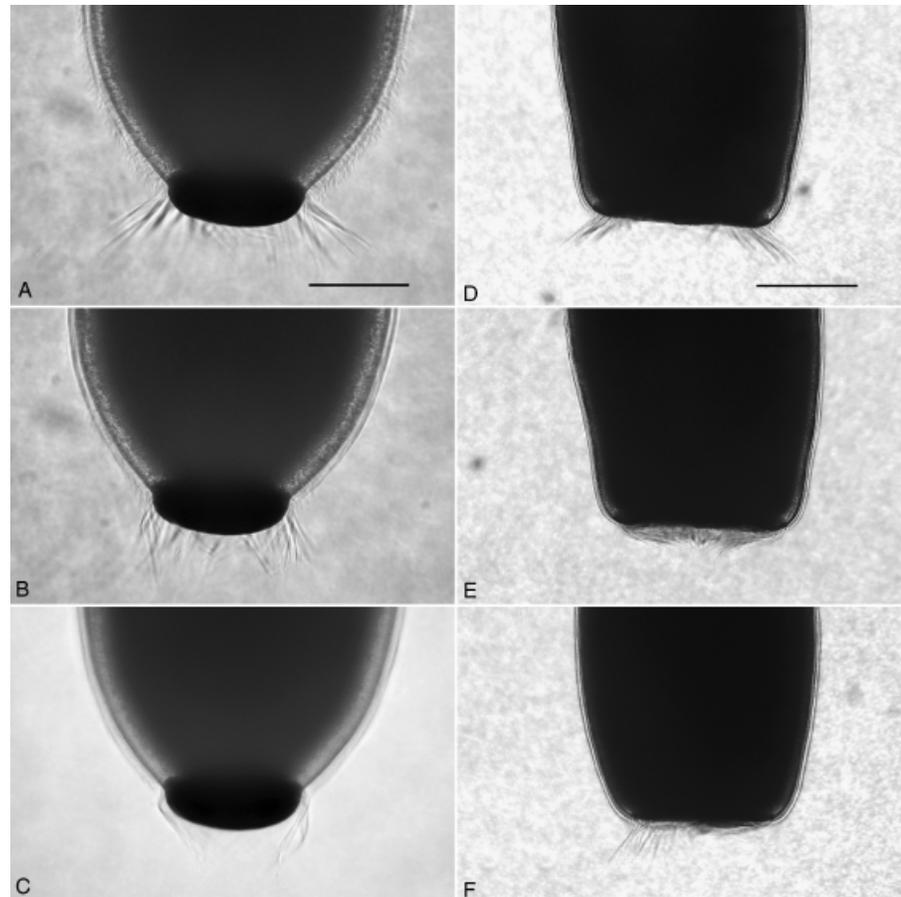


Fig. 3. Responses of long posterior cilia (LPC) to changes in the light intensity. **A.** LPC of *Neopetrosia proxima* flared outwards when the light intensity was reduced. Scale bar = 100 μm . **B.** LPC of *N. proxima* in a neutral position during forward swimming. Scale bar same as in A. **C.** LPC of *N. proxima* folded inwards when the light intensity was increased. Scale bar same as in A. **D.** LPC of *Xestospongia bocatorensis* flared outwards when the light intensity was reduced. Scale bar = 150 μm . **E.** LPC of *X. bocatorensis* folded inwards when the light intensity was increased. Scale bar same as in D. **F.** LPC of *X. bocatorensis* folded asymmetrically when the larvae were turning. Scale bar same as in D.

is very rapid while the secondary response lasts for several seconds and includes beating movements of the cilia. The LPC of larvae of *X. bocatorensis* have a neutral position, with the cilia slightly tucked in toward the center. When the light intensity is reduced, the cilia flare outwards (Fig. 3D) and slowly return to the neutral position after several seconds. When the light intensity increases rapidly, the LPC fold in close to the posterior end of the larvae (Fig. 3E). We also observed asymmetrical folding of the LPC when the larvae were turning (Fig. 3F), although this folding was not associated with lateral differences in illumination. All larvae of both species were consistent in their behavior, except that the response became slower and less pronounced with repeated exposure of any individual to rapid changes in light.

Discussion

Adults of both *Neopetrosia proxima* and *Xestospongia bocatorensis* brood tufted, type IIIa parenchymella larvae that are similar to other reported haplosclerid larvae (Mariani et al. 2005). Larvae have been described from <2% of the known species of

sponges (Maldonado & Bergquist 2002) and no larvae of members of the genera *Neopetrosia* or *Xestospongia* have been described previously in the literature. Although it has been proposed that all members of Petrosiidae may be oviparous, with free spawning of separate sexes (Fromont & Bergquist 1994; Maldonado & Riesgo 2009), our observations counter this hypothesis. These new data demonstrate that modes of reproduction vary among petrosid species, as observed within other families and orders of sponges (van Soest 1991).

The larvae of *N. proxima* are positively phototactic and settled predominantly in the light, while the larvae of *X. bocatorensis* are negatively phototactic and settled predominantly in the shade. Because members of both of these species maintain symbiotic relationships with photosynthetic cyanobacteria (Erwin & Thacker 2007), it is unexpected that *X. bocatorensis* does not show photopositive settlement behavior. Adults of *N. proxima* host unicellular cyanobacteria, classified as *Synechococcus spongiarum* USHER et al. 2004, while adults of *X. bocatorensis* host filamentous cyanobacteria, classified as *Oscillatoria spongeliae* (SCHULZE 1879) (Fig. 1E,F). These two types of

symbionts may interact differently with their hosts, yielding distinct patterns of larval settlement. Alternatively, light intensity may not be the most important factor in settlement site selection; habitat structure, chemotaxis, rheotaxis, biofilm formation, and other variables in the natural environment could clearly influence larval settlement behavior. Of the 100 sponge species (in 29 families) that are known to host symbiotic cyanobacteria (Diaz et al. 2007), phototactic responses of larvae from these species have only been documented in the two species reported here. Thus, it is premature to suggest any general trends in larval phototaxis among species hosting photosymbionts.

Observations and photographs of larvae show that turning is clearly associated with a change in the orientation of the LPC, with or without a light stimulus (Fig. 3; Maldonado et al. 2003; pers. obs.). However, the observed swimming behaviors do not correspond very well to the predictions of the Maldonado model (Table 1). The Maldonado model can account for the swimming behavior of larvae of three of the six species described in the literature (*Ircinia oros*, *X. bocatorensis*, and *Cacospongia mollior* in both its photopositive and photonegative stages). Larvae of two species show the opposite phototaxis of that predicted by the model (*Amphimedon queenslandica* and *Sigmatocia caerulea*). Finally, the response of the LPC of larvae of *N. proxima* is distinct from any of the other species examined previously. For larvae of *N. proxima*, the immediate flaring in response to increased light intensity, which is followed by several seconds of beating, inward-folded cilia, and a gradual return to the original state, is not likely to generate a turn in any straightforward way. It seems likely that the immediate and very short-lived initial response is a “startle” or a stress response that is not indicative of how the cilia would react under normal changes in light intensity experienced in the field. It is possible that the LPC flaring response observed in response to changes in light intensity is independent of any directional swimming or phototactic response.

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References

- Amano S 1986. Larval release in response to a light signal by the intertidal sponge *Halichondria panicea*. Biol. Bull. 171: 371–378.
- 1988. Morning release of larvae controlled by the light in an intertidal sponge, *Callyspongia ramosa*. Biol. Bull. 175: 181–184.
- Diaz MC 2005. Common sponges from shallow marine habitats from Bocas del Toro region, Panama. Caribb. J. Sci. 41: 465–475.
- Diaz MC, Thacker RW, Rützler K, & Piantoni C 2007. Two new haplosclerid sponges from Caribbean Panama with symbiotic filamentous cyanobacteria, and an overview of sponge–cyanobacteria associations. In: Porifera Research: Biodiversity, Innovation, and Sustainability, Série Livros 28. Custódio MR, Lôbo-Hajdu G, Hajdu E, & Muricy G, eds., pp. 335–343. Museu Nacional, Rio de Janeiro, Brazil.
- Erwin PM & Thacker RW 2007. Incidence and identity of photosynthetic symbionts in Caribbean coral reef sponge assemblages. J. Mar. Biol. Assoc. UK 87: 1683–1692.
- Ettinger-Epstein P, Whalan S, Battershill CN, & de Nys R 2008. A hierarchy of settlement cues influences larval behaviour in a coral reef sponge. Mar. Ecol. Prog. Ser. 365: 103–113.
- Fromont J & Bergquist PR 1994. Reproductive biology of three sponge species of the genus *Xestospongia* (Porifera: Demospongiae: Petrosida) from the Great Barrier Reef. Coral Reefs 13: 119–126.
- Gaino E, Baldacconi R, & Corriero G 2007. Post-larval development of the commercial sponge *Spongia officinalis* L. (Porifera, Demospongiae). Tissue Cell 39: 325–334.
- Kaye HR & Reiswig HM 1991. Sexual reproduction in four Caribbean commercial sponges. I. Reproductive cycles and spermatogenesis. Invert. Reprod. Dev. 19: 1–11.
- Leys SP & Degnan BM 2001. Cytological basis of photo-responsive behavior in a sponge larva. Biol. Bull. 201: 323–338.
- Lindquist N, Bolser R, & Laing K 1997. Timing of larval release by two Caribbean demosponges. Mar. Ecol. Prog. Ser. 155: 309–313.
- Maldonado M 2009. Embryonic development of *verongid demosponges* supports the independent acquisition of spongin skeletons as an alternative to the siliceous skeleton of sponges. Biol. J. Linn. Soc. 97: 427–447.
- Maldonado M & Bergquist PR 2002. Phylum Porifera. In: Atlas of Marine Invertebrate Larvae. Young CM, ed., pp. 21–50. Academic Press, San Diego, CA, USA.
- Maldonado M, Durfort M, McCarthy DA, & Young CM 2003. The cellular basis of photobehavior in the tufted parenchymella larva of demosponges. Mar. Biol. 143: 427–441.

- Maldonado M & Riesgo A 2009. Gametogenesis, embryogenesis, and larval features of the oviparous sponge *Petrosia ficiformis* (Haplosclerida, Demospongiae). *Mar. Biol.* 156: 2181–2197.
- Maldonado M & Uriz MJ 1998. Microrefuge exploitation by subtidal encrusting sponges: patterns of settlement and post-settlement survival. *Mar. Ecol. Prog. Ser.* 174: 141–150.
- Maldonado M & Young CM 1996. Effects of physical factors on larval behavior, settlement and recruitment of four tropical demosponges. *Mar. Ecol. Prog. Ser.* 138: 169–180.
- Mariani S, Uriz MJ, & Turon X 2005. The dynamics of sponge larvae assemblages from northwestern Mediterranean nearshore bottoms. *J. Plankton Res.* 27: 249–262.
- van Soest RWM 1991. Demosponge higher taxa classification re-examined. In: *Fossil and Recent Sponges*. Reitner J & Keupp H, eds., pp. 54–71. Springer, Berlin, Germany.
- Thacker RW, Diaz MC, Rützler K, Erwin PM, Kimble SJA, Pierce MJ, & Dillard SL 2007. Phylogenetic relationships among the filamentous cyanobacterial symbionts of Caribbean sponges and a comparison of photosynthetic production between sponges hosting filamentous and unicellular cyanobacteria. In: *Porifera Research: Biodiversity, Innovation, and Sustainability*, Série Livros 28. Custódio MR, Lôbo-Hajdu G, Hajdu E, & Muricy G, eds., pp. 621–626. Museu Nacional, Rio de Janeiro, Brazil.
- Wapstra M & van Soest RWM 1987. Sexual reproduction, larval morphology and behaviour in demosponges from the southwest of the Netherlands. In: *Taxonomy of Porifera*. Vacelet J & Boury-Esnault N, eds., pp. 281–307. Springer-Verlag, Berlin, Germany.
- Whalan S, Ettinger-Epstein P, Battershill C, & de Nys R 2008. Larval vertical migration and hierarchical selectivity of settlement in a brooding marine sponge. *Mar. Ecol. Prog. Ser.* 368: 145–154.
- Woollacott RM 1990. Structure and swimming behavior of the larvae of *Halichondria melanodocia* (Porifera: Demospongiae). *J. Morphol.* 205: 135–145.