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## **Cover Page Footnote**

I thank P.A. Hastings, H.J. Walker, J.S. Stephens, and two anonymous reviewers for reading and commenting on early manuscripts describing this work. B.E. Erisman and O. Aburto-Oropeza organized field expeditions that allowed me to conduct these observations and served as dive buddies, along with J. Lund. C. Sanchez and P. Hastings helped in procuring the necessary permits for collecting fishes in Mexico. Funding for expeditions was provided by Instituto Nacional de Ecología (INE), Fondo Mexicano para la Conservación de la Naturaleza, a Pew Marine Conservation Fellowship awarded to E. Ezcurra, and anonymous donors. Additional funding to support this work was provided by the Center for Marine Biodiversity and Conservation.

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*Acanthemblemaria balanorum* Brock (Clubhead Blenny; Fig. 1) is a chaenopsid tube blenny endemic to the Tropical Eastern Pacific. Adults of this species, as all members of the Chaenopsidae, inhabit vacated invertebrate tubes or tests (Stephens 1963; Lindquist 1985). In the case of *A. balanorum*, the shelter of choice is the vacated test of *Megabalanus* Hoek barnacles, a genus characterized in the Tropical Eastern Pacific by a complex of species (Henry and McClaughlin 1986) that typically live in the upper 10 m on shallow rocky reefs (Brusca and Hendrickx 2008). In the Gulf of California (GOC), Mexico, *A. balanorum* overlaps in distribution with two congeners, *A. crockeri* Beebe and Tee-Van and *A. hastingsi* Lin and Galland, and these species are known to exhibit depth partitioning, with *A. balanorum* inhabiting relatively shallower depths, *A. crockeri* inhabiting relatively deeper depths, and *A. hastingsi* overlapping near the edges of the depth ranges of the other two species at intermediate depths (Lindquist 1985). A detailed study of the relationships among these three congeners in the southern GOC (Lindquist 1985) reported that *A. balanorum* inhabits shelters (=barnacles) down to approximately 7 m depth. Guides to the fishes of the region (e.g., Allen and Robertson 1994; Humann and DeLoach 2004) report a similar depth range.

In November 2010, I observed and collected several individuals of *A. balanorum* at a depth of 21 m at the base of a pinnacle off the south end of Maria Cleofas, the southernmost point in the Islas Marias archipelago, southern GOC. These individuals, like all individuals of this species that I have observed, inhabited vacant barnacles (*Megabalanus*). This observation represents a significant depth range extension for this normally shallow subtidal fish (and may also represent an extension for the barnacle; Brusca and Hendrickx 2008). This ability of a microhabitat specialist to colonize abnormal macrohabitats (in this case much deeper than normal waters) when its microhabitat (=barnacles) is available supports a hypothesis that these specialists are resource (=shelter) limited. Similar shelter limitation has already been experimentally demonstrated in the GOC congener, *A. crockeri*, which increases in average density with shelter addition (Hastings and Galland 2010).

I observed additional evidence that *A. balanorum* is a shelter-limited microhabitat specialist in July 2009 at Las Animas, a small island and a series of small pinnacles in the central GOC. That site proved to be ideal *A. balanorum* habitat, with several large boulders completely covered by broad, very dense *Megabalanus* fields down to 5 m depth. Within these barnacle fields, I observed large numbers of *A. balanorum*, more densely distributed than any other chaenopsid population reported to date (e.g. in Lindquist 1985; Clarke 1996; Thomson and Gilligan 2002; P.A. Hastings, pers. comm.). During an opportunistic survey of the area, I placed a 0.25 m<sup>2</sup> quadrat on five randomly selected areas on the top of a large, flat boulder at 5 m depth in order to survey chaenopsids and



Fig. 1. *Acanthemblemaria balanorum* in shelter at Las Animas in the central Gulf of California, Mexico. Photo taken in July 2009 at 5 m depth during nearest neighbor surveys. Photo by J. Lund.

ascertain densities. Numbers of *A. balanorum* ranged (mean  $\pm$  SD) from 8 – 28 (20.4  $\pm$  8.6) individuals per 0.25 m<sup>2</sup>. These high densities may reflect the very high barnacle densities at this site, though it is difficult to quantify available shelters, as it is not immediately obvious what a chaenopsid considers to be a sufficient shelter.

To date, no experiments have been designed to determine what a second limiting factor is for these or similar shelter-dwelling microhabitat specialists, but it is quite possibly an issue of territory size. Males of *Acanthemblemaria* and several other chaenopsid genera actively court females, which, upon choosing a suitable male, enter the male's shelter and deposit eggs (Hastings 1986; Hastings 1988; Hastings and Peterson 2011). The males guard the eggs and may simultaneously protect clutches from multiple females (Hastings 1986; Hastings 1988; Hastings and Peterson 2011). Under these conditions, males compete for female choice and for the most desirable shelters (Hastings 1988; Hastings 1992). The high densities of *A. balanorum* observed at Las Animas could lead to agonistic interactions that secondarily limit the population size (or density) of this species.

In order to begin quantifying the distance between occupied barnacle shelters at the densely populated Las Animas site, I randomly chose an individual *A. balanorum* and measured the distance to its nearest neighbor. I then measured the distance to that individual's nearest neighbor and repeated the process until I reached ten total individuals. On average (SD), these ten individuals were only 4.4 cm (1.6) from their nearest neighbor. Upon collecting the specimens, I obtained sex and standard length (SL) for each individual and determined that they were, on average (SD), 28.9 mm SL (3.8) and were all sexually mature adults. In fact, to the best of my knowledge, juvenile habitat preference in this species is unknown. The male to female sex ratio was 7:3, indicating the ability of several adult males to live in close proximity to one another and to mature females, which were somewhat evenly dispersed among the males, within this set of ten individuals (numbers one, three, and eight out of ten; 5, 2.5, and 5 cm from their nearest neighbors, respectively). The presence of some shelters inhabited by less competitive

chaenopsid species, *Coralliozetus angelicus* (Böhlke and Meade) and *Protemblemaria bicirrus* (Hildebrand), increases the overall chaenopsid density, and the availability of some uninhabited shelters perhaps implies that the community at that site is approaching some maximum. It is difficult, however, to determine whether or not empty barnacles represent a choice to avoid high densities, a preference for other more highly desirable shelters, or some additional factor.

While the preliminary evidence presented here supports a hypothesis that these microhabitat specialists are first limited by the presence of their preferred shelter and then by some other factor (possibly territoriality) that prevents higher densities, even with greater shelter availability, it is necessary to design and implement experiments to test this and similar hypotheses empirically. Species like *A. balanorum* that rely on biologically derived microhabitats may be able to utilize a wider range of macrohabitats as their “hosts” move to new areas. In the case of the depth range extension at Maria Cleofas, the ability of *Megabalanus* barnacles to survive down to at least 21 m at this site allows a species that specializes on its empty tests to do the same, and this ability is not unique to *A. balanorum*. A blenniid (*Hypsoblennius brevipinnis* Günther) and another chaenopsid (*A. macrospilus* Brock) were observed and collected at the Maria Cleofas site, inhabiting barnacle tests. *Hypsoblennius brevipinnis* is also a microhabitat specialist, its preferred shelter is also *Megabalanus*, and it is typically confined to the upper 3–4 m of the subtidal zone (Allen and Robertson 1994; Humann and DeLoach 2004; though it may be known from as deep as 10 m; De la Cruz Agüero et al. 1997). Like in *A. balanorum*, this observation represents a significant depth range extension for this specialist and an interesting finding for the Blenniidae, which are characterized by few species reaching depths greater than approximately 25 m (Springer and Smith-Vaniz 1970). *Acanthemblemaria macrospilus*, sister to the more northern *A. hastingsi* discussed above (see Lin and Galland 2010), shares its ability to utilize multiple microhabitat shelters (Lindquist 1985) and typically lives to depths of 15–18 m (Allen and Robertson 1994). Observation of individuals at a depth of 21 m may represent a minor change in depth range, but its flexibility in shelter choice probably allows it to occupy a greater diversity of habitats than *A. balanorum*. Further research into the intra- and inter-specific relationships within this group of species that utilize (and potentially compete for) similar microhabitats will reveal more of the processes at play in determining the makeup of this assemblage and may provide further insight into the relative advantages and disadvantages of microhabitat specialization.

All specimens of all species discussed here were collected using quinaldine and are archived at the Scripps Institution of Oceanography Marine Vertebrate Collection (Las Animas collection number = SIO 09-261; Maria Cleofas collection number = SIO 10-132).

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I thank P.A. Hastings, H.J. Walker, J.S. Stephens, and two anonymous reviewers for reading and commenting on early manuscripts describing this work. B.E. Erisman and O. Aburto-Oropeza organized field expeditions that allowed me to conduct these observations and served as dive buddies, along with J. Lund. C. Sanchez and P. Hastings helped in procuring the necessary permits for collecting fishes in Mexico. Funding for expeditions was provided by Instituto Nacional de Ecología (INE), Fondo Mexicano para la Conservación de la Naturaleza, a Pew Marine Conservation

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