SIZE AND LOCATION RELATIONSHIPS OF STALKED BARNACLES OF THE GENUS *OCTOLASMIS* ON THE MANGROVE CRAB *SCYLLA SERRATA*

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ABSTRACT

Cyprid larvae of the lepadomorph *Octolasmis* colonize the gill chambers of the edible mangrove crab *Scylla serrata* (Forskål, 1755). In a natural population of 856 mangrove crabs from southern Thailand, 260 individuals were infested by 3,670 *Octolasmis cor* and 1,758 *O. angulata*, including 1,614 subadults, 168 cyprids, and 38 peduncles of the two species. This population of gill chamber symbionts was examined to investigate the relationship between barnacle size and barnacle spatial distributions. The habitat of the branchial chamber was partitioned for study into gills one through eight, the inside (hypobranchial) and outside (hyperbranchial) gill surfaces, and the proximal, medial, and distal regions of each gill. The collective data from 260 crabs were pooled for an analysis that showed a nonrandom relationship between the size of octolasmids and their location within the gill chamber. On the inside gill surfaces *O. angulata* attained its largest average size on gills 3, 7, and 8, whereas on the outside surface the barnacles were largest on gills 4 and 5. *Octolasmis cor* attained its greatest average size on the inside surface of gill number 6. Comparisons of barnacles from the three gill regions also revealed some significant differences in average barnacle size. Positive correlations among barnacle size, barnacle number, and barnacle density were present. Moreover, there was a significant correlation between the total numbers of barnacles and the average size of *O. cor* on the inside surfaces of gill numbers 1 to 8, whereas it was not significant for *O. angulata*. There was also a significant correlation between barnacle densities and barnacle size in *O. cor* but not *O. angulata*. Positive associations were also observed among higher numbers of barnacles, larger barnacles, and higher numbers of advanced reproductive stages. Areas with higher densities also were areas of higher average fecundity.

Pedunculate or lepadomorph barnacles are in the Order Pedunculata Newman, 1987, within the Superorder Thoracica Darwin, 1854. Barnacles of the genus *Octolasmis* Gray, 1825 (Crustacea: Cirripedia: Poecilasmatidae Annandale, 1909) are pedunculates but their habitats differ from common gooseneck barnacles such as *Lepas anatifera* Linnaeus, 1767, which live attached to floating objects (Anderson, 1994). The majority of *Octolasmis* species are symbiotic (*sensu lato*, de Bary, 1879) on the integument of decapod Crustacea. As cyprid larvae, the barnacles cement themselves to these ephemeral substrates. They metamorphose on them into juveniles, grow, and live out their adult lives. At host ecdysis the life cycle ends because both the integument (exuviae) and attached barnacles are shed. Thus the life cycle of the symbionts must exist within the life cycle of their hosts, and it is likely that the two are integrated and may be regulated by some of the same hormones.

Colonization of newly molted mangrove crabs *Scylla serrata* (Forskål, 1755) by *Octolasmis* cyprid larvae is significantly pulsed because the cyprids aggregate on premolt crabs (but do not attach) and at the time of host ecdysis the cyprids move from the old exuviae to the integument of the newly molted crab (Jeffries *et al.*, 1989). Thus, cyprid larvae exploring recently molted decapods may interact with each other, but there is no chance of them encountering attached adult *Octolasmis*.

Knowledge of infestation levels, attachment sites, numbers and density of barnacles on the host integument—along with the sex, age, and physiological condition of the hosts—provides a basis for understanding the host symbiont relationship. For example, in a sample of 856 mangrove crabs *S. serrata*, 30 percent harbored *Octolasmis* in their gill chambers, and crabs with carapace widths of less than 34.3 mm bore no *Octolasmis*. The percentage of crabs hosting *Octolasmis* in-

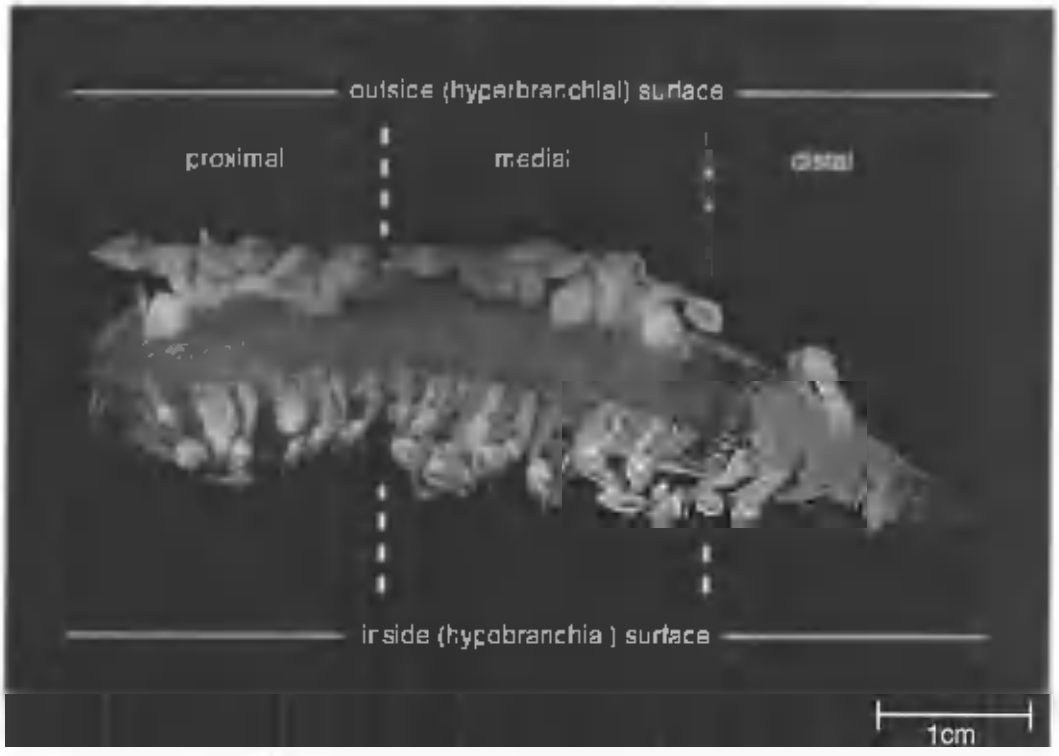

Fig. 1. Posterior view of gill 5 of the mangrove crab *Scylla serrata*. *Octolasmis angulata* can be seen to dominate the outside (hyperbranchial) surface whereas *O. cor* dominates the inside (hypobranchial) surface. The proximal, medial, and distal regions are also shown.

creased as the crabs approached sexual maturity, and the magnitude of infestation increased with crab size. Among immature crabs all barnacles were on the inside (hypobranchial) gill surfaces, whereas among mature crabs 11 percent were on the outside (hyperbranchial) gill surfaces. The number of barnacles on the hypo- and hyperbranchial gill surfaces of mature crabs was a function of the number of barnacles in the gill chamber (Jeffries *et al*., 1992). *Octolasmis angulata* (Aurivillius, 1894) and *O. cor* (Aurivillius, 1892) were commonly observed living together in the branchial chambers of *S. serrata*. Among the 6,648 barnacles observed, there were about twice as many *O. cor* as *O. angulata*. The spatial distributions of the two species on the gills of *S. serrata* were nonrandom, uneven, and did not reflect available surface area. The two barnacle species were distributed differently on the hypo- and hyperbranchial gill surfaces and were distributed differently on the gills of immature (< 70 mm carapace width) and mature (> 70 mm) crabs (Voris *et al*., 1994).

In this study we investigated the relationship between barnacle size and barnacle spatial distribution for *O. angulata* and *O. cor* residing in the gill chambers of a natural population of the mangrove crab *S. serrata*. The relationships between size and the numbers and densities of barnacles in various locations in the branchial chambers of the crabs were also examined. Abiotic and biotic factors that may influence the settlement patterns of the two barnacle species are presented.

Materials and Methods

Crab and Barnacle Samples.—The same mangrove crab specimens and symbiotic octocorallines used in two previous studies form the foundation for the current report (Jeffries *et al*., 1992; Voris *et al*., 1994). The sample of 856 mangrove crabs, which included 403 males and 453 females ranging in size from 10.9 to 132.2 mm carapace width (males 5–18), were collected from a natural population in southern Thailand. The carapace of each crab was removed and the gills examined for *Octolasmis* cyprids, juveniles, and adults. Adult barnacles were identified to species whereas cyprids, small juveniles, and some intermediate-sized juveniles could not be so identified. The exact site of barnacle attachment (left or right gill chamber, gill number, inside (hypobranchial) or out-

Table 1. The number and mean capitular length (MCL) of *Octolasmis angulata* and *O. cor* taken from the inside and outside surfaces of gills 1 through 8 of 260 infected *Scylla serrata*. In cases where the number is three or less the MCL is not applicable (na).

Gill number	*Octolasmis angulata*				*Octolasmis cor*			
	Inside		Outside		Inside		Outside	
	Number	MCL	Number	MCL	Number	MCL	Number	MCL
1	3	na	15	1.90	60	1.81	6	na
2	2	na	16	1.98	8	1.70	14	1.83
3	72	1.97	23	1.79	442	2.02	1	na
4	84	1.83	92	1.95	1,128	2.08	5	1.75
5	140	1.66	228	1.84	751	2.09	23	1.95
6	265	1.82	88	1.65	769	2.16	3	na
7	514	1.99	21	1.36	368	2.03	1	na
8	187	1.98	1	na	70	1.76	1	na
Totals =	1,267		486		3,616		48	

side (hyperbranchial) gill surface, proximal, medial, or distal gill region (see Fig. 1) and the length (mm) of the capitulum of each barnacle were recorded using methods employed by Jeffries and Voss (1989). A dissecting microscope was used to determine the reproductive status of the barnacles.

Of the 856 crabs examined, 260 hosted the 6,648 individuals of *Octolasmis*. The barnacle population included 1,258 *O. angulata* with a mean capitular length (MCL) of 1.88 mm (range from 0.856 to 4.004 mm), 3,670 *O. cor* with a 2.066 mm MCL (range from 0.572 to 4.719 mm), 1,014 unidentified juveniles with a 0.841 mm MCL (range from 0.572 to 1.287 mm), 98 peduncles only, and 168 cyprids.

Octolasmis angulata and *O. cor* were differentiated on the basis of external features such as overall shape, capitular shape, and capitular plate morphology (Jeffries *et al.*, 1991). In addition, *O. cor* regularly attained a larger size than *O. angulata*, e.g., there were very few *O. angulata* at or above a 3.00 mm capitular length, whereas over 100 *O. cor* equaled or surpassed that capitular length. Juveniles were sub-adult barnacles of both species that could not be assigned to species with certainty.

Treatment of Unidentified Juveniles.—To account for the size of all individuals in each area, an adjusted mean capitular length (AMCL) for both *O. angulata* and *O. cor* was computed. The AMCL for each species in each region was based on the measurements of both the identified adults and unidentified juveniles in the same proportion as the adults in that region. Therefore, the AMCL provides an estimate of the average size of all the individuals of each of the species in a given region. Several conclusions emerged from an analysis of these derived figures.

Gill Area Measurements.—Because the various gill regions have very different surface areas we computed barnacle densities for each region. The average percentage of the total inside surface area of the gills of *S. serrata* are distributed as follows: gill 1, 6.48%; gill 2, 5.07%; gill 3, 10.02%; gill 4, 11.64%; gill 5, 18.82%; gill 6, 18.24%; gill 7, 15.02%; gill 8, 14.66%. Areas for the proximal, medial, and distal regions of each gill were also determined from direct measurements using a digitizer. Densities of barnacles were determined for each region using the measured areas.

Reproductive Stages.—During the data collection phase of this research each *O. angulata* and *O. cor* was assigned to one of four reproductive categories that represent progressively more advanced stages of reproductive condition or readiness. All observations were made using a dissecting microscope. The stages were defined as follows: *Non-reproductive*—there were no pre-ovary cells, no organized islands of cells, and no recognizable strands of pre-ovary cells within the peduncle; *Turgid*—the ovary was clearly recognizable as a branched, lobulated, organ of iridescent cells within the peduncle. This stage was characterized by a distinct ovary; *Gravid*—ovigerous lamellae (Darwin, 1851), i.e., bilateral concave, plate like, aggregates of developmental stages were clearly recognizable within the capitular cavity; *Eyed*—the ovigerous lamellae were punctuated with many minute black spots, each representing the median eye of a larva and collectively indicating an imminent larval release. The black eye spots observed suggested that this ontogenetic stage is comparable to stage N in the development of *Balanus galloprovincialis* (Sowerby, 1823); (see Lewis, 1975).

Levels of Analysis.—The relationship between the size of barnacles and their location in the gill chambers of the host crabs was considered from several perspectives. In one approach the distribution of each species of barnacle was pooled over all 260 crabs that had one or more barnacles in their gill chambers. Thus, for each species we recorded the number and mean capitular length (MCL) of barnacles located in the left and right gill chambers, on gills 1 to 8, on the inside and outside of each gill surface, and on the proximal, medial, or distal gill regions. Table 1 provides the numbers and MCLs of the two barnacle species over the 8 gills on the inside and outside gill surfaces. For this table the left and right chambers of all crabs were pooled, and the proximal, medial and distal regions along the gills were pooled.

Because the analysis of these data required a large number of comparisons a conservative approach was warranted. For multiple comparisons of MCLs between gill regions we used the Tukey Honest Significant Difference Test for unequal sample sizes (Tukey-HSD Test, see Sprugel and Stohlgren, 1992). In some cases where a limited number of comparisons were involved we also provided the probabilities based on the Least Significant Difference (LSD) test or individual *t* tests, but these tests offer less protection from error resulting from multiple *post hoc* comparisons.

In a second approach we used the patterns of differences observed in the analysis of the pooled samples to

direct specific comparisons within the individual gill chambers of a subset of crabs. In this analysis the replicate was the individual crab chamber with its unique history and assemblage. The within-chamber comparisons were limited by the relatively small number of crabs with large numbers of barnacles. Even among these crabs, sample sizes in specific regions were often so small that they precluded meaningful comparisons. One subset of crabs consisted of 17 individuals (34 chambers) that had more than 100 barnacles (both chambers combined). Within this sample we scored each chamber for a given comparison (e.g., MCL on gill number 5 larger than on gill 6) and then compared the results for all 34 chambers to a binomial distribution using a Chi square test. In addition, in some cases, in a subset of just 5 crabs that had the largest numbers of barnacles (160 to 326 barnacles, two chambers combined) we compared the MCLs of barnacles located in particular regions. For these selected tests we used both the Tukey Honest Significant Difference Test for unequal sample sizes (Tukey ≠N HSD Test) and the Least Significance Difference (LSD) test.

Results

Gill by Gill Comparison on the Inside and Outside Surfaces

Octolasmis angulata.—The mean capitular lengths (MCL) of the *O. angulata* located on the inside surfaces of gills 1 to 8 of all 260 *S. serrata* with one or more octolasmids are presented in Fig. 2A. Sample sizes varied among gills from 2 *O. angulata* on gill number 2 to 514 on gill 7. Gills 1 and 2 were not considered further because of small sample sizes (Table 1). Gills 3, 7, and 8 had *O. angulata* with similar MCLs (Fig. 2A). Gills 4, 5, and 6 had populations with smaller MCLs. The MCLs of *O. angulata* on gills 3, 7, and 8 were each significantly different from the MCL of the group on gill number 5 (Tukey ≠N HSD test, gill number 3 vs. gill 5, $P = 0.013$; gill 7 vs. gill 5, $P < 0.000$; gill 8 vs. gill 5, $P < 0.000$). The MCLs of *O. angulata* on gills 4 and 6 were not significantly different from the MCLs on gill 5 (Tukey ≠N HSD test, $P > 0.05$).

The pattern of size differences on the outside surfaces of the gills was different from that on the inside surfaces (Fig. 2B). Gill number 8 was not included in this analysis because of a sample of only 3 (Table 1), and sample sizes were generally smaller on the outside surface with a maximum of 228 *O. angulata* on gill 5. In general the pattern was of gradually declining MCLs from gill 4 to gill 7. The MCLs of *O. angulata* on gills 1 to 5 showed no significant differences (LSD test, $P > 0.05$; Tukey ≠N HSD test, $P > 0.05$), but the MCL of *O. angulata* on gill 4 differed from both the MCLs on gills 6 and 7 (Tukey ≠N HSD test, $P = 0.003$ and $P = 0.006$ respectively).

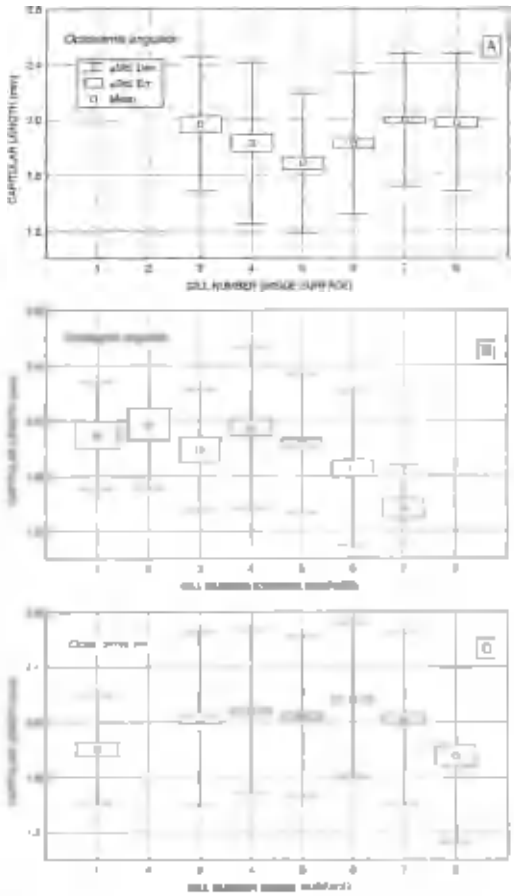

Fig. 2. The mean capitular lengths (MCL), one standard error, and one standard deviation are given for gills 1 to 8 for (A) the *Octolasmis angulata* located on the inside (hypobranchial) surfaces, (B) the *O. angulata* located on the outside (hyperbranchial) surfaces, and (C) the *O. cor* located on the inside surfaces. The barnacles were located in the gill chambers of 260 *Scylla serrata*, and the sample sizes for each gill are given in Table 1.

Octolasmis cor.—The number of *O. cor* on the inside surfaces of gills 1 through 8 far exceeded that on the outside surfaces (Table 1); thus, only data from the inside surfaces were analyzed. Because the sample size on gill number 2 is only 8 (Table 1), it was not included in the following comparisons (Fig. 2C). The MCLs of the *O. cor* located on the inside surfaces of gills 3, 4, 5, and 7 did not

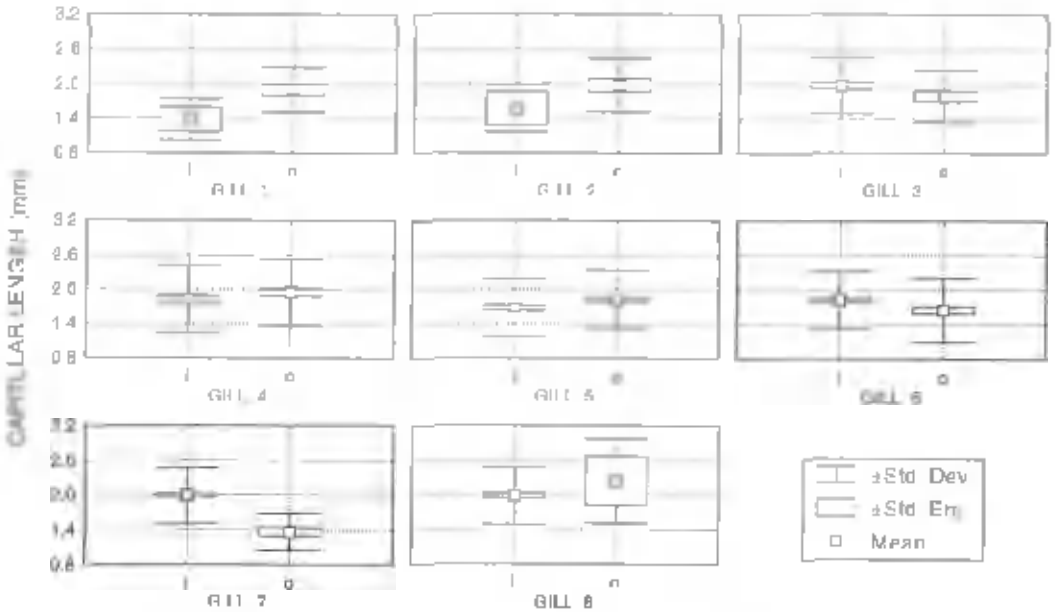

Fig. 3. The mean capitular lengths (MCL), one standard error, and one standard deviation are given for *Octolasmis angulata* located on the inside (hypobranchial) and outside (hyperbranchial) surfaces of gills 1 to 8 of *Scylla serrata*. The sample sizes for each gill are provided in Table 1.

show significant differences (Tukey ≠N HSD test, $P > 0.05$). Both gills 1 and 8 had animals with smaller MCLs, and the *O. cor* on gill 6 had the largest MCL (Table 1, Fig. 2C). In fact, the MCL of *O. cor* on gill 6 was significantly larger than the MCLs on gills 1, 3, 5, 7, and 8 (Tukey ≠N HSD test, $P < 0.05$).

Comparison of the Inside and Outside Surfaces

Octolasmis angulata.—The *O. angulata* on the inside gill surfaces outnumbered those on the outside gill surfaces 2.6 to 1 (Table 1). The MCL of *O. angulata* located on the inside surfaces of all gills (pooled) was larger ($\bar{x}$ = 1.905 mm, s = 0.511, n = 1,272) than the MCL of those located on the outside of the gills ($\bar{x}$ = 1.811 mm, s = 0.532, n = 486; t = 3.422, $d.f.$ = 8, n = 1,756, $P < 0.001$).

A gill by gill comparison of MCLs of *O. angulata* between the inside and outside surfaces using an analysis of variance resulted in a significant F value (F = 7.520, $d.f.$ = 16, $P < 0.001$). Gills 3, 6, and 7 have *O. angulata* with larger MCLs on the inside than the outside (Fig. 3). The differences between inside and outside surfaces on gills 3 and 6 are not significant (gill 3, inside $\bar{x}$ = 1.970 mm, s = 0.486, n = 72; outside $\bar{x}$ = 1.791 mm, s = 0.439, n = 23; Tukey ≠N HSD, $P > 0.05$; gill 6, inside $\bar{x}$ = 1.825 mm, s = 0.506, n = 265; outside $\bar{x}$ = 1.653 mm, s = 0.558, n = 88; Tukey ≠N HSD, $P > 0.05$). The MCL of *O. angulata* on the inside of gill 7 was significantly larger than the MCL of *O. angulata* on the outside (inside $\bar{x}$ = 1.995 mm, s = 0.480, n = 514; outside $\bar{x}$ = 1.362 mm, s = 0.315, n = 21; Tukey ≠N HSD, P = 0.005).

Octolasmis angulata located on the inside surfaces of gills 1, 2, and 8 were smaller in average size than those located on the outside (Fig. 3), but these differences were not credible because the sample sizes were low (Table 1).

Octolasmis angulata located on the inside surface of gills 4 and 5 were smaller in average size than those located on the outside surfaces, but these difference were not significant (gill 4, inside $\bar{x}$ = 1.83 mm, s = 0.583, n = 84; outside $\bar{x}$ = 1.951 mm, s = 0.583; Tukey ≠N HSD, $P > 0.05$; gill 5, inside $\bar{x}$ = 1.68 mm, s = 0.506, n = 140; outside $\bar{x}$ = 1.836 mm, s = 0.502, n = 228; Tukey ≠N HSD, $P > 0.05$).

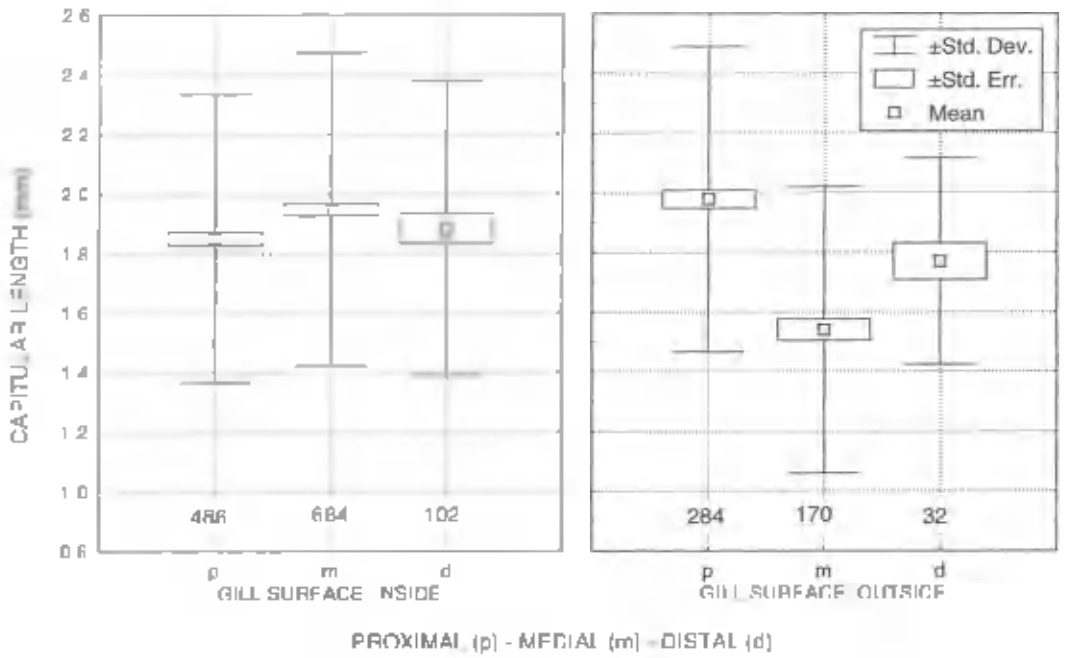

Fig. 4. The mean capitular lengths (MCL), one standard error, and one standard deviation are given for the *Octolasmis angulata* located on the proximal, medial, and distal regions on the inside (hypobranchial) and outside (hyperbranchial) gill surfaces. The number of barnacles in each region is given below each box and whisker plot. Gills 1 to 8 of *Scylla serrata* are combined in this presentation.

Comparison of the Proximal, Medial, and Distal Regions on the Inside and Outside Surfaces of All Gills

Octolasmis angulata.—The MCLs of *O. angulata* showed significant variation between the proximal, medial, and distal regions of the inside and outside surfaces of all gills (ANOVA, $F = 20.715$, $d.f. = 5$, $P < 0.001$).

The MCLs of *O. angulata* located on the proximal, medial, and distal regions of the inside surfaces of all gills combined (Fig. 4) showed one significant difference. *Octolasmis angulata* on the medial region of the inside surface were, on average, significantly larger in MCL than those on the proximal region (medial, $\bar{x} = 1.948$ mm, $s = 0.527$, $n = 684$; proximal, $\bar{x} = 1.850$ mm, $s = 0.486$, $n = 486$; Tukey ≠N HSD, $P = 0.030$). The other two comparisons, proximal with distal and medial with distal, showed no differences (Tukey ≠N HSD test, $P > 0.05$).

Octolasmis angulata located on the proximal, medial, and distal surfaces of the outside of all gills combined (Fig. 4) showed one significant difference in average size. The MCL of *O. angulata* on the medial region was significantly smaller than that on the proximal region (medial, $\bar{x} = 1.540$ mm, $s = 0.479$, $n = 170$; proximal, $\bar{x} = 1.978$ mm, $s = 0.512$; Tukey ≠N HSD, $P < 0.001$).

Octolasmis cor.—An analysis of variance of the MCLs of *O. cor* located on the proximal, medial, and distal regions of the inside (most are inside) and outside surfaces resulted in a significant F value ($F = 3.959$, $d.f. = 5$, $P = 0.001$). The pattern of differences in MCLs for *O. cor* on the inside surfaces is the same as observed in *O. angulata*. Although *O. cor* located on the proximal, medial, and distal regions of the inside of all gills combined showed significant differences in the MCLs using the least significance difference (LSD) test (e.g., proximal, $\bar{x} = 2.047$ mm, $s = 0.577$, $n = 1,558$; medial, $\bar{x} = 2.087$ mm, $s = 0.603$, $n = 2,005$; LSD, $P = 0.044$), none of the differences were significant using the more conservative Tukey ≠N HSD test ($P > 0.05$). The *O. cor* on the distal region were fewer and smaller in MCL ($\bar{x} = 1.925$ mm, $s = 0.639$, $n = 103$) than those on the proximal and dis-

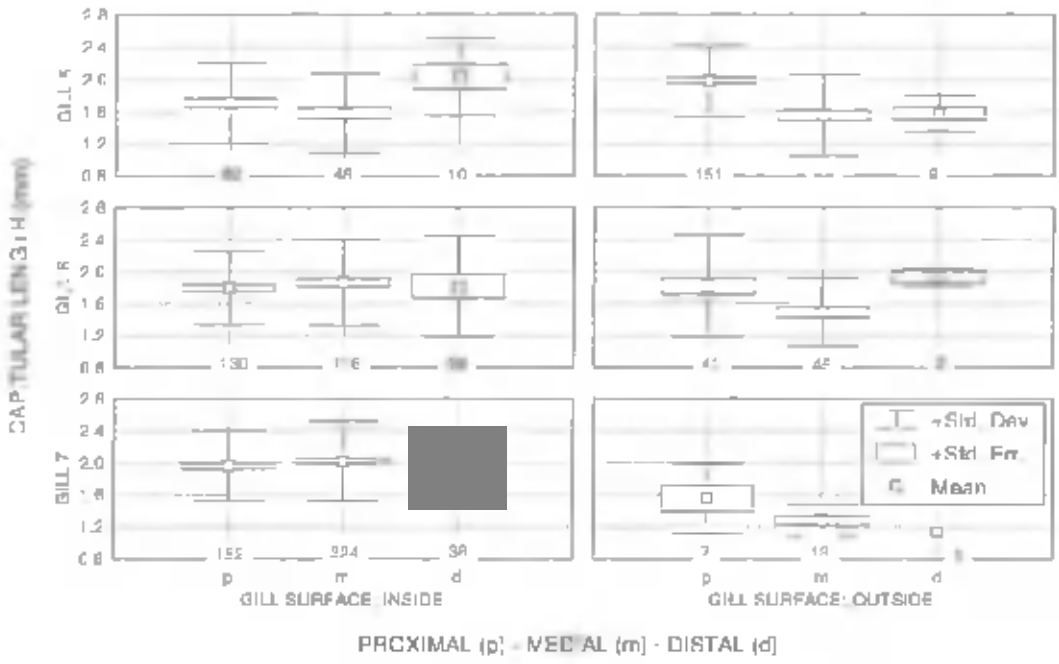

Fig. 5. The mean capitular lengths (MCL), one standard error, and one standard deviation are given for the *Octolasmis angulata* located on the inside (hypobranchial) and outside (hyperbranchial) surfaces of the proximal, medial, and distal regions of *Scylla serrata* gills 5, 6, and 7. The number of barnacles in each region is given below each box and whisker.

tal regions, but the differences were not significant.

Comparison of the Proximal, Medial, and Distal Regions on the Inside and Outside Surfaces of Selected Gills

Despite the fact that more than 5,000 barnacles were identified to species and measured (Table 1), the partitioning of barnacles into gill regions often led to sample sizes too small for meaningful comparisons. To overcome this limitation, comparisons in this section were limited to the gills with the largest numbers of barnacles.

Within-side Comparisons. —The *O. angulata* on gills 5, 6, and 7 had the largest sample sizes and merited consideration (Fig. 5). An analysis of variance of the MCLs on these gills resulted in a highly significant F value ($F = 5.544$, $d.f. = 41$, $P = 0.000$, $P < 0.05$).

Comparisons within gill 5 showed some differences. For example, on the inside gill surface, the MCL of *O. angulata* in the distal region ($x = 2.031$ mm, $s = 0.481$, $n = 10$) was larger than the MCLs of those in the proximal region ($x = 1.704$ mm, $s = 0.498$, $n = 82$; LSD test, $P = 0.048$; Tukey ≠N HSD, $P > 0.05$) and in the medial region ($x = 1.576$ mm, $s = 0.497$, $n = 48$; LSD test, $P = 0.008$; Tukey ≠N HSD, $P > 0.05$) regions (Fig. 5). Those on the proximal and medial regions were not significantly different in MCL from each other (LSD test, $P > 0.05$; Tukey ≠N HSD, $P > 0.05$).

On the outside gill surfaces, the MCL of *O. angulata* on the proximal region of gill 5 ($x = 1.981$ mm, $s = 0.449$, $n = 151$) was significantly larger than the MCL of *O. angulata* on the medial region ($x = 1.550$ mm, $s = 0.505$, $n = 68$; LSD test, $P < 0.001$; Tukey ≠N HSD, $P < 0.001$) or distal ($x = 1.573$ mm, $s = 0.226$, $n = 9$; LSD test, $P = 0.016$; Tukey ≠N HSD, $P > 0.05$) regions (Fig. 5). Those in the medial and distal regions were not significantly different from each other (LSD test, $P > 0.05$; Tukey ≠N HSD, $P > 0.05$).

Comparisons within gill number 6 showed only one significant difference. On the outside surface of gill 6 the MCL of the *O. an-*

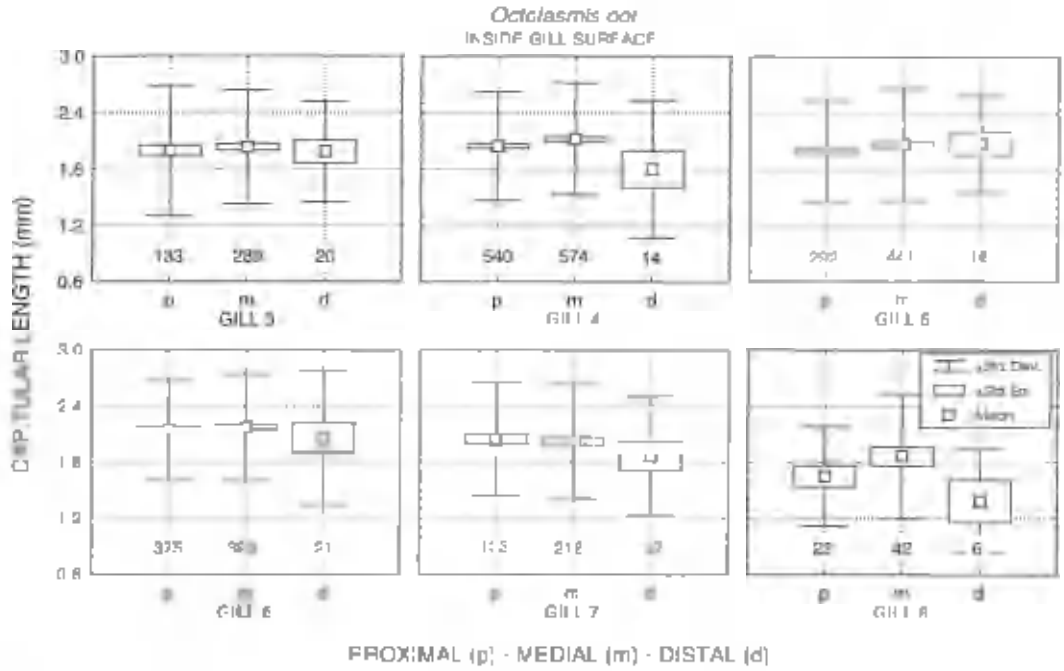

Fig. 6. The mean capitular lengths (MCL), one standard error and one standard deviation are given for *Octolasmis cor* located on the inside (hyperbranchial) surfaces of the proximal, medial, and distal regions of *Scylla serrata* gills 3 to 6. The number of barnacles in each region is given below each plot.

gulata on the proximal region (x = 1.817 mm, s = 0.644, n = 41) was significantly larger than the MCL of those on the medial region (x = 1.490 mm, s = 0.428, n = 45; LSD test, P = 0.002; Tukey ≠N HSD, P > 0.05). All other within-side comparisons of *O. angulata* on gill 6 did not show significant differences (Fig. 5). Comparisons of MCLs on proximal, medial, and distal regions within gill 7 showed no significant differences for all within-side comparisons (Fig. 5). Relatively small sample sizes at this level precluded meaningful within chamber comparisons.

Octolasmis cor showed many fewer differences and greater uniformity in size over the surface of the gills than did *O. angulata* (Fig. 6). The MCLs on the proximal, medial, and distal regions of the inside surfaces resulted in a significant F value (ANOVA, F = 3.048, $d.f.$ = 35, P < 0.001). There were two significant differences in the MCLs of *O. cor* from within gills, between the proximal, medial and distal regions and both occurred on gill 4 (Fig. 6). *Octolasmis cor* on the proximal surface (x = 2.046 mm, s = 0.577, n = 540) of gill 4 were significantly smaller in MCL than those on the medial surface (x = 2.121 mm, s = 0.594, n = 574; LSD test, P = 0.036; Tukey ≠N HSD, P > 0.05) and *O. cor* on the medial surface were significantly larger in MCL than those on the distal portion (x = 1.801 mm, s = 0.730, n = 14; LSD test, P = 0.044; Tukey ≠N HSD, P > 0.05).

Between side Comparisons.—There were significant differences in MCLs of *O. angulata* found on the inside and outside surfaces of gill number 5 (Fig. 5). The MCLs of *O. angulata* on the inside proximal and inside medial surfaces were significantly smaller than the MCL on the outside proximal surface (inside proximal vs. outside proximal, LSD test, P < 0.001; Tukey ≠N HSD, P > 0.05; inside medial with outside proximal, LSD test, P < 0.001; Tukey ≠N HSD, P = 0.033). However, the MCLs on the inside proximal and inside distal were not significantly different from those on the outside medial and distal surfaces (LSD test and Tukey ≠N HSD, P > 0.05).

The MCL of *O. angulata* on the outside, medial surface of gill 6 was smaller than the MCLs of those on the inside proximal, inside medial, and inside distal surfaces (Fig. 5). All these comparisons were significant us-

ing the LSD planned comparison test ($P < 0.05$) but none were significant using the more conservative Tukey ≠N HSD test ($P > 0.05$). This same pattern of difference was also apparent on gill 7, but the sample size on the medial outside surface was only 13 and the MCLs were not significantly different ($P > 0.05$).

Individual Gill Chamber Analysis

To determine if the above findings based on pooled samples could be demonstrated in individual gill chambers, we investigated a subset of 17 crabs (34 gill chambers) that had more than 100 barnacles in both chambers combined. To do this, we scored each of the 34 chambers with respect to each comparison. For example, the largest between-gill difference in Fig. 2C fell between gills 6 and 8. A scoring of the 34 chambers of the 17 crabs resulted in 6 chambers that had larger *O. cor* on gill number 8 than on gill 6 and 4 chambers that had larger ones on gill number 6 than on gill 8. Twenty-six chambers could not be scored because one of the two gills (usually gill 8) lacked any *O. cor*. Thus, small samples within chambers on gill 8 prevented a conclusive comparison.

However, a chamber tally showed gills 4 and 7 differing significantly, with 15 chambers containing *O. cor* with MCLs greater on gill 4 than on gill 7 and only 5 chambers with MCLs greater on gill 7 (chi-square, $P < 0.025$). A comparison of the MCLs on gills 4 and 5 in the 34 chambers also produced a significant deviation from 1:1, with 24 chambers with MCLs larger on gill 4 than on gill 5 and 5 chambers with MCLs larger on gill 5 (chi-square, $P < 0.01$). Gill 4 was found to have larger MCLs than gill 3 in 26 chambers, whereas gill 3 had larger MCLs in only 7 of the chambers (chi-square, $P < 0.001$). Thus, in several comparisons where sample sizes were sufficiently large, within-chamber comparisons corroborated the results based on pooled samples. However, in the majority of comparisons between particular regions (e.g., gill 4, inside surface, distal region with gill 4, outside surface, distal region), sample sizes proved too small for any meaningful comparisons.

Relationships Between Barnacle Size, Numbers, and Density

The distribution of all barnacles (both species, all stages) over gills 1 to 8 deviated sharply from the expected distribution based on gill area alone (chi-square = 1,614.7; $P < 0.001$). Thus, barnacles were not distributed according to available surface area alone.

It is noteworthy that there were some strong positive correlations between barnacle size, barnacle number, and barnacle density. That is, to some extent, larger numbers, higher densities, and larger barnacles vary together. For example, there was a significant correlation ($r = 0.958$, $P < 0.001$) between the total number of barnacles (all adults, unknowns, and peduncles) on the inside surfaces of gills 1 to 8 and the MCLs of *O. cor* on the inside gill surfaces (Fig. 7A). On the other hand, there was not a significant correlation ($r = 0.610$, $P > 0.05$) between the total number of barnacles on the inside surfaces of gills 1 to 8 and the MCLs of *O. angulata* on the inside surfaces (Fig. 7B). These results suggested that *O. angulata* and *O. cor* respond differently to density-related factors.

There was a strong correlation between barnacle numbers and barnacle densities over gill regions (e.g., numbers of all barnacles vs. density of all barnacles over gills 1 to 8, $r = 0.922$, $P < 0.001$). Thus, it is not surprising that, as with numbers, there was a significant correlation between the density of barnacles on the inside surfaces of gills 1 to 8 and the MCLs of *O. cor* on the inside gill surfaces ($r = 0.908$, $P < 0.01$), and there was no significant correlation between density and the MCLs of *O. angulata* on the inside surfaces ($r = 0.637$, $P > 0.05$).

Consideration of Juveniles

Unidentified juveniles were distributed very much like the adults, and the AMCLs over the various regions were very similar to the MCLs but consistently slightly lower than the MCLs. The same relationships tended to hold when comparing the AMCLs with the numbers or densities of barnacles. For example, the relationships between AMCLs and barnacle density for *O. angulata* and *O. cor* depicted in Fig. 7.

Reproductive Stage, Numbers, Size, and Location

More individual *O. angulata* (44%) and *O. cor* (41%) were assigned to the non-reproductive stage than to any of the other three stages (Table 2). The turgid, gravid, and eyed stages were progressively less common in

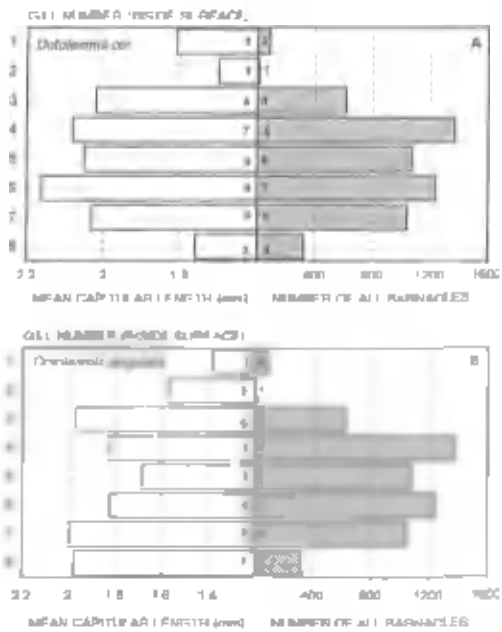

Fig. 3. The total number of all barnacles and the mean capitular lengths (MCL) are compared for the inside (hyprobranchial) surfaces of each of the 8 *Scylla serrata* gills for (A) *Octolasmis cor* and (B) *O. angulata*. Within each bar the rank order from smallest to largest is given. Note that the ranks of numbers and size are very similar for *O. cor* but dissimilar for *O. angulata*.

both species. The turgid and gravid stages occurred as similar percentages, ranging from 24% to 31%, whereas the eyed stage was much less common, ranging from 5.2% to 6.5% (Table 2).

This pattern of relative numbers of reproductive stages was also observed in both *O. angulata* and *O. cor* when the samples were categorized by inside and outside gill surface, proximal, medial, and distal gill region, and by gill number.

However, there were some notable variations. For example, the overall percentages of reproductively active stages (the percentages of turgid, gravid, and eyed stages summed) were 58.2%, 57.5%, and 51.0% for *O. angulata* on the inside proximal, medial, and distal regions, respectively. In contrast, on the outside surfaces, the overall percentages of reproductively active stages were 68.7%, 27.6%, and 46.9% on the proximal, medial, and distal regions, respectively. The relatively low numbers of reproductively active stages on the outside medial region corresponded to a relatively low number of individuals that were also of relatively small size (Fig. 4).

Table 2. The number and percentage of each reproductive stage for scales of the two species of octolasmid barnacle found on 260 mangrove crabs (*Scylla serrata*).

	O. angulata		*O. cor*	
Stage	Number	Percent	Number	Percent
Nonreproductive	773	44	1,490	41
Turgid	454	26	1,129	31
Gravid	417	24	861	24
Eyed	114	6.5	189	5.2

Discussion and Conclusions

The ability of barnacle cyprids to locate highly specific substrates (microhabitats) for colonization is well documented in the literature (Foster, 1987). For example, Moyse (1971) reported that *Megatrema anglicum* (Gray, 1825) is secondarily attracted to conspecifics after the host coral has been located. Most species within the genus *Octolasmis* colonize a limited number of host species (usually decapods) and are typically very selective as to the site of attachment on the body of the host (Jeffries *et al.*, 1982). Moreover, a symbiont may exploit only a subset of a host population, partitioning the population by factors such as age, sex, and environment (Jeffries *et al.*, 1992; Shields, 1992; Hudson and Lester, 1994).

The present work further documents a nonrandom relationship between the size of octolasmids and their location within the gill chambers of their hosts. Thus, it is apparent that *O. angulata* and *O. cor* attain greater average size in some locations over others. Positive correlations among barnacle size, barnacle number, and barnacle density are present. There are also positive correlations among higher numbers of barnacles, larger barnacles, and higher numbers of advanced reproductive stages. Areas with higher densities also are areas of higher average fecundity. Thus, it appears that the selection of the attachment site by the cyprid larvae strongly impacts adult fitness parameters.

In an earlier paper (Voris *et al.*, 1994), it was suggested that the spatial distribution of *O. angulata* and *O. cor* on the gills of *S. serrata* is nonrandom and does not reflect available surface area as measured by gill length or gill area as approximated from measure-

ments on *Callinectes sapidus*. Our current results, although more detailed, are consistent with what has been previously reported (Bullock, 1964; Arudpragasam, 1967; Jeffries *et al.*, 1982, 1992; Venkateswaran and Fernando, 1982; Voris *et al.*, 1994).

There are positive correlations among higher barnacle numbers, larger barnacles, and higher numbers of advanced reproductive stages. It has also been demonstrated for at least two species of octolasmids that larger barnacles have larger brood sizes (Jeffries and Voris, 1983; Matheswari and Fernando, 1989). Thus, regions that have higher numbers of barnacles and larger barnacles also are areas of higher average fecundity.

Dynamic Factors Resulting in Regions of High Numbers and Large Size

The nonrandom distribution of octolasmids observed in this study and previous studies is most likely a result of selection on the part of the cyprid larvae. We have observed very little evidence of differential mortality that could lead to the observed differences. For example, only 38 capitulum-less peduncles were observed in this entire study, and these were not concentrated in low density regions.

The association of relatively large barnacle size with particular locations within the gill chambers could result from one of two mechanisms. Larger size could be attained through earlier colonization and thus a longer period of growth, or larger size could be attained simply through faster growth rates in particular regions. Of course, it is possible that both mechanisms may be operating some or all of the time.

Our results do not allow us to distinguish with certainty between these two processes. However, some of our data support the notion that some regions are better than others for barnacle growth and a preponderance of cyprids select particular regions where the adults attain relatively large size and earlier sexual maturity. This support stems from two observations. First, colonization of newly molted *S. serrata* by *Octolasmis* cyprid larvae has been shown to be pulsed. It is pulsed in the mangrove crab because the cyprids aggregate on premolt crabs and at the time of host ecdysis the cyprids move together from the old exuviae to the integument of the newly molted crab where they soon settle (Jeffries *et al.*, 1989) and have similar amounts of time for growth and development. A second observation comes from the distribution patterns of barnacles in gill chambers bearing either high or low numbers of octolasmids. Both species tend to be found only on the inside gill surfaces when the total number of barnacles within the chamber is less than 20. This pattern holds for *O. cor* until the number within the chamber reaches 160, but *O. angulata* occurs on both the inside and outside surfaces when the numbers within a chamber begin to exceed 20 barnacles (Voris *et al.*, 1994). These data suggest that the inside surface is preferred by cyprids until the density of congenerics reaches certain thresholds.

In conclusion, we assert that site selection within the gill chambers of mangrove crabs by *O. angulata* and *O. cor* cyprid larvae has a direct influence on barnacle growth, fecundity, and, ultimately, fitness.

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