



2004

Variability in broods of the seastar *Leptasterias aequalis*

William B. Jaeckle

Illinois Wesleyan University, wjaeckle@iwu.edu

Brian L. Bingham

Kehualani Giles

Recommended Citation

Jaeckle, William B.; Bingham, Brian L.; and Giles, Kehualani, "Variability in broods of the seastar *Leptasterias aequalis*" (2004). *Scholarship*. Paper 10.
http://digitalcommons.iwu.edu/bio_scholarship/10

This Article is brought to you for free and open access by The Ames Library, the Andrew W. Mellon Center for Curricular and Faculty Development, the Office of the Provost and the Office of the President. It has been accepted for inclusion in Digital Commons @ IWU by the faculty at Illinois Wesleyan University. For more information, please contact digitalcommons@iwu.edu.

©Copyright is owned by the author of this document.

NRC Research Press, doi: 10.1139/Z04-009
Canadian Journal of Zoology (2004), v. 82, 457-463

Variability in Broods of the Seastar *Leptasterias aequalis*

Brian L. Bingham, Kehaulani Giles, and William B. Jaeckle

Abstract: Enormous variation exists in the reproductive output of marine invertebrates (e.g., in the numbers of embryos produced, the volumes of embryos, and the energy that they contain). It is not clear why there is such great variability or what the population-level consequences are. We sampled a population of the brooding seastar *Leptasterias aequalis* (Stimpson, 1862) to collect basic information on brood sizes, embryo volume, and embryo energy content with a goal to better understand the reproductive ecology of this species. We collected brooding females in February and again in April. We measured the size of their broods and sampled the broods to estimate volume and energy content of the embryos. There was great variability in the volume and energy content of embryos produced by individual females and among the embryos in a single female's brood. Larger adults produced larger embryos, which generally had greater energy content and may be of a higher quality. The average energy content of embryos appeared to increase during the brooding period. Larger females produced larger broods but lost a greater proportion of the embryos. The net result is that larger individuals may not produce any more juveniles than smaller individuals, but those that they do produce may be of a higher quality.

Introduction

Seastars in the genus *Leptasterias* are common in the northeast Pacific, ranging from Alaska to California. Occurring in both intertidal and subtidal environments, they are characteristic members of many hard-bottom communities.

These small-bodied seastars are unusual in their reproductive biology. In contrast with most local asteroids, *Leptasterias* females attain reproductive maturity at a body size of only 25 mm (Osterund 1918) and an age of about 2 years (Menge 1975). Reproductive females produce very large eggs (approximately 900 μm in diameter), which they brood externally for several months (November–April; Chia 1966).

Fully developed juveniles eventually crawl away from the brood mass.

Because *Leptasterias* larvae do not have a planktonic stage, their dispersal potential is probably much less than that of free-spawning species. Populations on a single beach, for example, may be largely self sustaining. The dynamics of a population in a particular habitat may, therefore, depend largely on the reproductive success of the resident females.

Menge (1974) studied the influence of habitat on reproductive output of *Leptasterias hexactis* (Stimpson, 1853) in exposed and protected habitats of Washington State (USA). He found that females at a protected inland site allocated significantly more energy to reproduction, increasing the

numbers of eggs that they produced and brooded. George (1994) compared populations of *Leptasterias epichlora* (Brandt) at two inland study sites. She found larger females at one of the sites (presumably because of a better food supply there). Those larger females produced more eggs. In addition, their eggs (*i*) were larger, (*ii*) had a higher protein content, and (*iii*) resulted in larger juveniles. Results of both of these studies suggest significant plasticity in reproductive output based on environment. This has clear implications for the population dynamics of these animals.

It is less clear how much reproductive investment and reproductive success of *Leptasterias* species vary within a site where females are exposed to similar physical conditions and have access to similar food resources. By studying animals from a single limited area, we removed habitat variability and were able to look simply at inherent variation in reproductive characters among a group of brooding females. In particular, we posed the following questions. What is the relative reproductive contribution of large versus small females in a population? Presumably, larger individuals within a site produce more eggs. Are those eggs of a higher quality and do they have a better chance of reaching the juvenile stage? How much variability in egg quality exists within the brood of a single female? Are all embryos within a single brood similar or are there differences that could result in differential survival to metamorphosis?

The goal of this study was to gather initial information on the brood characteristics of female *Leptasterias* collected from a single intertidal beach. The results indicate significant variability both among and within broods and suggest further research directions to clarify the selective forces influencing these basic reproductive features.

Materials and methods

The taxonomy of the genus *Leptasterias* is in a state of flux. Historically, small six-rayed seastars in the northeast Pacific have been referred to as *L. hexactis* (but see George 1994). The most current evidence, however, suggests that the *Leptasterias* complex includes three distinct genetic lineages (Kwast et al. 1990; Hrinkevich et al. 2000; J. Flowers, personal communication). Lineages A and B constitute a clade composed of *Leptasterias aequalis* (Stimpson, 1862). Lineage C is most correctly referred to as *L. epichlora*. The seastars that we studied belong to the *L. aequalis* clade and we refer to them as such in this paper.

We sampled an *L. aequalis* population from the Shannon Point Marine Center beach in Anacortes, Washington, USA. To ensure that we sampled the earliest possible broods, we began, in mid-January, to check seastars on the beach at 2- to 3-day intervals. Broods first appeared on some individuals in mid-February. On 16 February, we collected 12 brooding females, measuring the radius of each as the distance from the middle of the central disk to the end of the longest arm. We used only complete individuals with six undamaged arms.

Within 3 days of collection, we removed the broods from the 12 collected seastars and counted the total number of embryos in each. At this point, the embryos were featureless, yolky balls. Because they were held together in an adhesive mass, the embryos had to be carefully teased apart to make an accurate count. We used an image analysis system

(Bioscan Optimas software, Media Cybernetics, Carlsbad, Calif.) to measure the cross-sectional areas of five arbitrarily chosen embryos from each brood. Embryo volumes were then calculated from these measurements, assuming that the embryos were spherical (Bingham 1997).

We measured organic content of the embryos with a wet ashing method (Parsons et al. 1984; McEdward and Carson 1987). Individual *L. aequalis* embryos were placed on a small piece of Whatman GF/C glass fiber filter paper that had been previously baked in a muffle furnace at 500 °C for 24 h (to remove all organic carbon). Light suction was applied with a Millipore filtration unit and the embryos were lightly washed twice with distilled water to remove salts and residual chloride (Parsons et al. 1984; also see Gosselin and Qian 1999). We placed each filter paper and embryo in a test tube containing 1 mL of 70% phosphoric acid and incubated them at 110 °C for 15 min. We then added 2 mL of 0.484% potassium dichromate in sulfuric acid to each test tube and continued the incubation for an additional 30 min. The samples were diluted to 6 mL total volume with distilled water and their absorbance at 440 nm was measured with a spectrophotometer (1-cm path length). The decreased extinction of the dichromate, relative to a reagent blank, gave an estimate of oxidizable carbon in the embryo. Glucose served as a standard for comparison. Oxidizable carbon was converted to energy assuming that 1 µg of C (from glucose) = 0.039 J (Parsons et al. 1984; McEdward et al. 1988).

While the dichromate oxidation method accurately accounts for the energy content of lipids and carbohydrates in the embryos, it underestimates the energy content of proteinaceous materials (Paine 1971). Despite this analytical uncertainty, we used this technique because it is sufficiently sensitive to explore variation among individuals. Furthermore, the level of uncertainty is equivalent to the mass fraction of organic material unaccounted for by traditional biochemical fractionation techniques (i.e., the remainder fraction; Jaekle and Manahan 1989).

To determine how brood size and embryo energy content change over the brooding period, we collected 12 additional brooding *L. aequalis* from Shannon Point beach on 12 April. This was near the end of the brooding season. The larvae had begun to develop the arms of the preoral lobe and there was some flattening at the site of the developing juvenile. However, the larvae were still very yolky, did not have mouth openings, and were not yet being released. Although the adhesive membrane that held the early embryos in a mass was no longer evident, the larvae continued to adhere to one another and again had to be teased away from the adult and from each other. Brood size and larval energy content were determined as described above. Because larvae were no longer spherical, we did not attempt to estimate their volumes.

Data analysis

To document variability in the broods, we tested for differences in the average volumes and energy contents of embryos from the females collected in February. One embryo was lost during processing, so the total sample size was 59. Comparisons among the broods were made with one-way ANOVA. The data for both analyses (embryo volume and energy content) violated the assumption of equal variance.

Table 1. *Leptasterias aequalis* brood size, embryo volume, and embryo and larval energy content in samples collected in February (at the beginning of the brooding period) and in April (near the end of the brooding period).

Measure	Month	<i>n</i>	Range	Mean	SD
Brood size (no. of embryos)	Feb.	12	142–784	359	243
	Apr.	12	50–198	110	39
Embryo volume (mm ³)	Feb.	60	0.26–0.52	0.37	0.10
Embryo and larval energy (J)	Feb.	59	2.53–8.48	6.23	2.10
	Apr.	60	1.58–15.0	8.75	2.62

Note: Because of the irregular shape of the larvae in the April samples, no volume estimates were made.

Subsequent transformations did not homogenize the variances, so we analyzed the raw data, adjusting α to 0.01 (Underwood 1981).

We hypothesized that embryo energy content would be positively related to embryo volume, with larger embryos containing more energy, thus being better (in the sense of having a greater chance of surviving the brooding period and (or) producing larger juveniles). We tested this with a regression analysis. Because both volume and energy content were subject to measurement error, we used Model II regression. Following the recommendations of Legendre and Legendre (1998), we used the ranged major axis method to approximate the slope and intercept of the regression line. Significance of the slope coefficient was determined by comparing it with slope coefficients generated through 500 random permutations of the raw data.

To determine if embryo volume varied with adult size, we tested for a simple correlation between embryo volume and adult radius. Because late-stage larvae collected in April were no longer spherical, it was not practical to estimate their volumes. As a result, only February embryos were included in analyses involving volume.

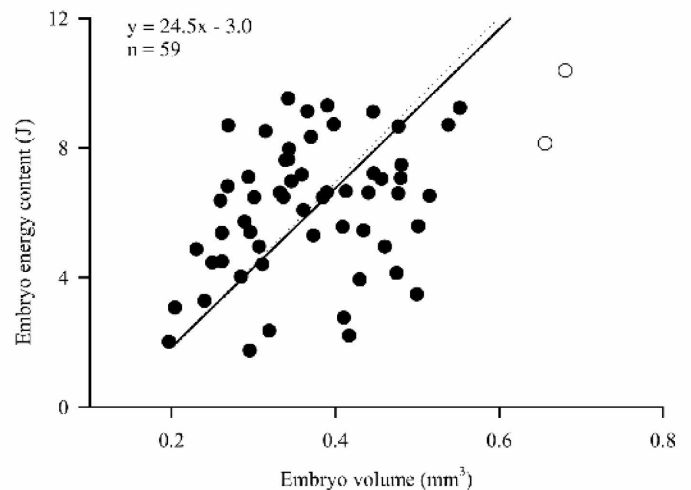
To determine how brood sizes change during the brooding period, we compared broods collected from the seastars in February with those held by the seastars collected in April. We compared the brood sizes with an ANCOVA. This approach was necessary because larger seastars, in general, appeared to produce larger broods. To meet the assumptions of ANCOVA (particularly to stabilize variances and to make the regression lines for early and late broods parallel), we log transformed the brood size data prior to the analysis. We ran the analysis using “date” (February versus April) as the main effect and “adult size” as the covariate.

It also seemed possible that individual embryos might change over the 2-month brooding season. We specifically tested the hypothesis that the energy content of the embryos changes during development by comparing the energy content of embryos taken from the February broods with that of larvae from the April broods. Because multiple embryos were taken from each seastar, our ANOVA included terms for “date” (February versus April), “seastar” (a random factor nested in date), and “larval energy content” (a random factor nested in both individual and date). All statistical assumptions were tested and met prior to analysis.

Results

The *L. aequalis* broods collected in February were extremely variable in size and in the volume and energy con-

Fig. 1. Relationship between *Leptasterias aequalis* embryo volume and energy content in early broods. Results of a Type II regression analysis are shown. Pooled data from 12 brooding adults are included. The dotted line represents the results of a regression analysis with the two extremely large embryos (○) excluded from the analysis.



tent of the embryos that they held (Table 1, Fig. 1). It was common, for example, to find 40%–50% differences in volume between the larger and smaller embryos held by a single female. Despite this high within-brood variability, mean embryo volume still varied significantly among broods ($F_{[11,48]} = 8.67, p < 0.001$) as did mean embryo energy content ($F_{[11,48]} = 5.59, p < 0.001$).

A ranged major axis regression showed a significant positive relationship between embryo volume and energy content despite considerable scatter in the data ($p = 0.002$) (Fig. 1). To ensure that the regression analysis was not unduly influenced by two unusually large embryos (Fig. 1), we re-analyzed the data without those two points. The resulting relationship was still significant ($p = 0.008$), suggesting that larger embryos do have significantly more energy.

Correlation analysis showed a significant positive relationship between embryo volume and adult size, indicating that larger *L. aequalis* produced larger (and more energy rich) embryos (Fig. 2). Again, however, there was a lot of scatter in the data, highlighting the significant within-brood variability.

We expected the number of embryos in a brood to be strongly correlated with size of the adult that produced them. This was true of the February broods ($r = 0.85, p < 0.001, n = 12$) (Fig. 3): larger seastars did produce larger broods.

Fig. 2. Relationship between the size of *L. aequalis* brooding adults and volume of the brooded embryos collected in April. Data are shown for 12 adults with five arbitrarily chosen embryos from each. Results of a correlation analysis are given.

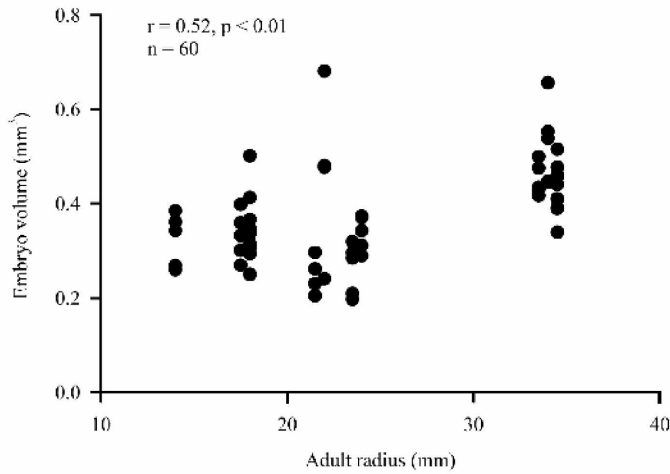
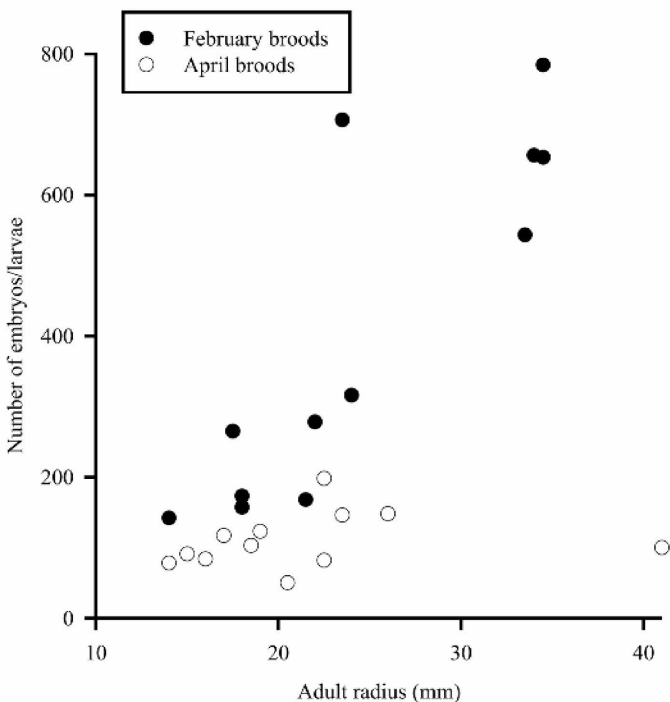


Fig. 3. Relationship between the size of *L. aequalis* brooding adults and the number of embryos and larvae in the brood. Data are shown for broods collected early (16 February) and late (18 April) in the brooding period.



By April, however, overall brood sizes had dropped and there was no longer a correlation between adult size and number of larvae ($r = 0.20$, $p = 0.53$, $n = 12$). ANCOVA showed that brood sizes in April were significantly lower than those in February, independent of adult size (Table 2). This finding reflects the loss of embryos and larvae over the 2-month brooding period.

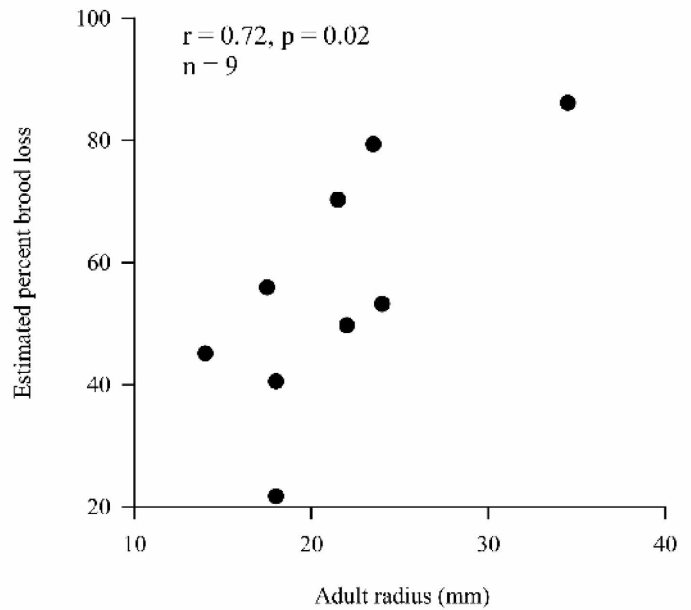
To estimate total embryo and larval loss over the brooding period, we matched adults of similar size from the February and April samples and calculated the difference in their

Table 2. *Leptasterias aequalis* brood size as a function of date (February versus April).

Source	df	MS	F	p
Date	1	1.126	33.35	<0.001
Error	21	0.033		
Total	23			

Note: Data were analyzed with an ANCOVA with adult seastar radius as the covariate (covariate effect was significant, $p < 0.001$).

Fig. 4. Estimated *L. aequalis* brood loss over the 2-month brooding period. Data were calculated by matching up seastars of similar size collected in February and April and comparing their broods. Correlation coefficient and significance level are shown.



brood sizes (assuming that similarly sized adults produced roughly equivalent broods). Loss estimates ranged from 21% to 86% (Fig. 4). If this is an accurate estimate of brood mortality, it indicates that *L. aequalis* lose a significant portion of the brood. The significant positive correlation suggested that the percent loss was related to adult size; estimated losses were much greater for larger seastars.

A nested ANOVA also showed significant differences in embryo energy content over the brooding period. Larvae from seastars collected near the end of the brooding period (in April) had significantly more energy, on average, than those tested in February (Tables 1 and 3). As expected, there was again significant variability in embryo energy content among seastars (indicated by the significant “seastar” factor in the analysis).

Discussion

Despite low samples sizes, both in the number of females sampled and in the number of embryos measured per brood (between 0.6% and 10%), our sampling revealed intriguing patterns in the brood characteristics of *L. aequalis*. The clearest result was that female reproductive output was extremely variable in this species. This is not unusual; variabil-

Table 3. *Leptasterias aequalis* embryo and larval energy.

Source	df	MS	F	p
Date	1	178.74	10.34	0.004
Seastar	22	17.27	6.18	<0.001
Embryo and larva	92	2.81		
Total	115			

Note: Data were analyzed as a completely nested ANOVA. Date (February versus April) was a fixed factor, while seastar and embryo and larva were nested random factors. Unbalance in the design came from four missing observations.

ity in fecundity and egg size has often been seen both among echinoderm species and among individuals within a species (e.g., Turner and Lawrence 1979; Thompson 1983; Emler et al. 1987; McEdward and Carson 1987; McEdward and Coulter 1987; McEdward and Chia 1991).

Among *Leptasterias* species, differences in fecundity have often been related to female size (Chia 1966; Menge 1970; Niesen 1973; George 1994). This was clearly true in our study. A doubling of the arm radius in brooding females resulted in a three- to fourfold increase in egg production. However, while difference in adult size may explain some of the variation in *L. aequalis* reproductive output, that alone cannot account for all of the variability. This is particularly true for the differences that we saw, not in sizes of broods but in the volume and energy content of embryos within single broods.

Variation among eggs produced by *Leptasterias* females can sometimes result from differences in habitat quality (Menge 1974; George 1994). In our study, however, all of the *L. aequalis* came from a single short stretch of beach. We were unable to detect habitat variability equivalent to that described in these earlier works, and it seems unlikely that habitat variability was sufficient to produce the range of egg volumes and energy contents that we saw. Furthermore, even if habitat variability could explain differences among individuals, it could not explain the variation among eggs in a single brood.

Variation in reproductive output, both among females and within broods of a single female, can have important life history consequences. Egg size has been used as a measure of female investment per propagule and (by summing over total fecundity) as a predictor of female reproductive effort in theoretical models of reproduction and life history evolution (Vance 1973; Smith and Fretwell 1974; Strathmann 1985; Havenhand 1995; McEdward 1997). For echinoderm species like *Leptasterias* spp., with brooded nonfeeding development, most nutrients and structural materials needed to complete development are provided in the egg. If egg volume is related to egg energy content, as it is in this and a number of other species (e.g., Jaekle 1995; McEdward and Morgan 2001), differences in egg volume will be translated through the embryonic and larval stages into juvenile size. Therefore, differences in egg size and volume could ultimately impact juvenile fitness (Lawrence et al. 1984; McClintock and Pearse 1986; Emler et al. 1987).

If we assume that juvenile fitness is directly related to embryo volume, then, based on the variation in embryo volume within the broods of *L. aequalis* examined for this study, some embryos could have twice the fitness potential of their

brood mates. This is probably a conservative estimate, since we sampled only a small proportion of the total broods; it is unlikely that we actually measured the largest and smallest embryos in any brood. However, this estimate fits well with data for *L. epichlora*. George (1994) found 1.5- to 3-fold differences in protein content between the largest and smallest juveniles released from single broods of that species.

A second pattern that we observed in our study was that brooding *L. aequalis* seem to lose embryos over the brooding period. Brood size decreases in *Leptasterias* species have been previously recognized (Chia 1966; Niesen 1973; Menge 1974; Hamel and Mercier 1995). Menge (1974) estimated a brood mortality of approximately 44% in the *L. hexactis* populations that he studied. Our estimated loss rates were slightly higher (approximately 58%). These levels of mortality are not unusual for brooding echinoderms. Sewell (1996), for example, measured brood mortalities of up to 100% in the intraovarian brooding sea cucumber *Leptosynapta clarki* Heding, 1928.

Our data further suggest that larger females produce more eggs but have a harder time carrying them completely through the brooding season. A positive relationship between adult size and brood size was clear in the February samples but had disappeared by April, primarily because large females were losing a much greater proportion of their broods (Figs. 3 and 4). This inverse relationship between adult size and brooding success was predicted theoretically by Strathmann and Strathmann (1982) and has been demonstrated in the brooding asteroid *Asterina phylactica* Emson and Crump, 1979 (see Strathmann et al. 1984).

There are a number of difficulties inherent in estimating brood mortality. Ideally, measurements would involve direct observation throughout the brooding period (e.g., Sewell 1996). Unfortunately, that is rarely possible. Instead, a series of simplifying assumptions must be made. Our estimates of brood loss rely on two assumptions: (1) that females of similar size produce broods of approximate equal size and (2) that all eggs in the broods are fertilized (for a discussion of this issue, see Rumrill 1990). Our analyses suggest that the first assumption is reasonable; brood size was strongly correlated with adult size in our population. We currently have no data to test the second assumption. If, however, these assumptions are true, a significant portion of the broods of *L. aequalis* never reach the juvenile stage.

In addition to simple changes in numbers, broods appear to change in composition. The decrease in brood size, coupled with a significant increase in average energy content of surviving larvae, indicates differential loss of offspring, with smaller individuals being more at risk. Egg volume in *L. aequalis* may, therefore, be a predictor of survival to the juvenile stage (Lawrence et al. 1984; McClintock and Pearse 1986; Emler et al. 1987).

At present, it is unclear what factors are responsible for the loss of brooded embryos and why those factors act more strongly on smaller embryos. However, two possibilities should be considered: (1) larger individuals consume smaller ones within the brood (adelphophagy) and (2) strong flow strips embryos (primarily the smaller ones) from the brooding female.

Adelphophagy of smaller individuals by larger individuals could easily produce the decreasing brood sizes with a shift

toward larger individuals that we observed in our study. However, brooding seastars for which adelphophagy has been suggested (e.g., *Neosmilaster georgianus* (Studer, 1885); see Bosch and Slattery 1999) possess tissue connections that have been interpreted as conduits for material transport between consumed and consuming individuals. No such structures have been described for *L. aequalis*. While we cannot exclude adelphophagy in our study, the lack of morphological features associated with this process and the absence of prior reports (despite intensive study of *L. hexactis*) lead us to believe that it is not the cause of the brood changes that we saw.

A more likely cause of brood loss is that embryos are simply flushed out of the brood mass by strong water flow. *Leptasterias hexactis* (and presumably *L. aequalis*) embryos hatch at the brachiolaria stage of development and attach to their mother using their brachiolar complex (brachiolar arms and the central adhesive disk; Osterund 1918). The epithelium of the brachiolar complex bears secretory cells that help the larva attach to the substratum (Barker 1978). We hypothesize that the differential loss of smaller individuals from broods in *L. aequalis* is a consequence of a relatively smaller brachiolar surface area for contact with the rest of the brood mass and the maternal body. Smaller individuals, owing to this reduced contact, are more susceptible to dislodgement and removal from the brood by shear stress associated with strong waves and currents.

The study site from which we collected *L. aequalis* is a relatively low-energy environment. Current speeds can be high (maximum currents of approximately 1.8 m/s), but wave action is minimal. However, *L. aequalis* broods during the winter, when occasional storms occur. In the period between our two samples, winds at the study site exceeded 10.2 m/s on five days. On two of these days (15 and 24 March), the winds were blowing directly onshore, producing strong waves. These events conceivably occurred shortly after the larvae had emerged from the fertilization membrane (hatching occurs 21–25 days after fertilization; Strathmann 1987). Although we cannot specifically state that high water flow stripped away the brooded embryos, the observations are consistent with our hypothesis. If wave action is indeed responsible for a differential loss of smaller individuals, it may explain the right shift in the distribution of energy content among individuals collected from older broods and the seeming paradox that older broods possess fewer but more energy-rich individuals.

Our study suggests significant variability in the number and energy content of embryos produced by *L. aequalis*, even though the sampled individuals occurred within a single small area. The variability was expressed among females within the population and even within the broods of single females. This variability may (i) simply result from random variation in reproductive output and survival, (ii) indicate different reproductive strategies among brooding females, or (iii) reflect the history of the maternal animal (e.g., Bingham et al. 2000). In any case, the variability may be expressed in differential survival of brooded offspring and fitness of those juveniles that eventually leave the brood. Variation in brooding success and juvenile size could have significant consequences for the population dynamics of *L. aequalis* and for the intertidal communities in which it occurs. Understanding

those consequences will require further careful study with particular attention to the dynamics and energetics of reproduction.

Acknowledgements

We gratefully acknowledge assistance from Steve Heywood and Gene McKeen. Steve Sulkin provided space and facilities at the Shannon Point Marine Center. This work was supported by National Science Foundation grant OCE-9315422.

References

- Barker, M. 1978. Structure of the organs of attachment of brachiolaria larvae of *Stichaster australis* (Verrill) and *Coscinasterias calamaria* (Gray). *J. Exp. Mar. Biol. Ecol.* **33**: 1–36.
- Bingham, B.L. 1997. Light cycles and gametogenesis in three temperate ascidian species. *Invertebr. Biol.* **116**: 61–70.
- Bingham, B.L., Burr, J., and Wounded Head, H. 2000. Causes and consequences of arm damage in the sea star *Leptasterias hexactis*. *Can. J. Zool.* **78**: 596–605.
- Bosch, I., and Slattery, M. 1999. Costs of extended brood protection in the Antarctic sea star, *Neosmilaster georgianus* (Echinodermata: Asteroidea). *Mar. Biol.* **134**: 449–459.
- Chia, F.S. 1966. Brooding behaviour of a six-rayed starfish, *Leptasterias hexactis*. *Biol. Bull. (Woods Hole)*, **130**: 304–315.
- Emlet, R.B., McEdward, L.R., and Strathmann, R.R. 1987. Echinoderm larval ecology viewed from the egg. *In Echinoderm studies. Vol. 2. Edited by M. Jangoux and J.M. Lawrence.* A.A. Balkema Publishers, Rotterdam, the Netherlands. pp. 55–136.
- George, S.B. 1994. Population differences in maternal size and offspring quality for *Leptasterias epichlora* (Brandt) (Echinodermata: Asteroidea). *J. Exp. Mar. Biol. Ecol.* **175**: 121–131.
- Gosselin, L.A., and Qian, P.-Y. 1999. Analysing energy content: a new micro-assay and an assessment of the applicability of acid dichromate assays. *Hydrobiologia*, **390**: 141–151.
- Hamel, J.-F., and Mercier, A. 1995. Prespawning behavior, spawning, and development of the brooding starfish *Leptasterias polaris*. *Biol. Bull. (Woods Hole)*, **188**: 32–45.
- Havenhand, J.N. 1995. Evolutionary ecology of larval types. *In Ecology of marine invertebrate larvae. Edited by L.R. McEdward.* CRC Press, New York. pp. 79–122.
- Hrincevich, A.W., Rocha-Olivares, A., and Foltz, D.W. 2000. Phylogenetic analysis of molecular lineages in a species-rich subgenus of sea stars (*Leptasterias* Subgenus *Hexasterias*). *Am. Zool.* **40**: 365–374.
- Jaekle, W.B. 1995. Variation in the size, energy content, and biochemical composition of invertebrate eggs: correlates to the mode of larval development. *In Ecology of marine invertebrate larvae. Edited by L.R. McEdward.* CRC Press, New York. pp. 49–77.
- Jaekle, W.B., and Manahan, D.T. 1989. Growth and energy imbalance during the development of a lecithotrophic molluscan larva (*Haliotis rufescens*). *Biol. Bull. (Woods Hole)*, **177**: 237–246.
- Kwast, K.E., Foltz, D.W., and Stickle, W.B. 1990. Population genetics and systematics of the *Leptasterias hexactis* (Echinodermata: Asteroidea) species complex. *Mar. Biol.* **105**: 477–489.
- Lawrence, J.M., McClintock, J.B., and Guille, A. 1984. Organic level and caloric content of eggs of brooding asteroids and an echinoid (Echinodermata) from Kerguelen (South Indian Ocean). *Int. J. Invertebr. Reprod. Dev.* **7**: 249–257.

- Legendre, P., and Legendre, L. 1998. Numerical ecology. 2nd English ed. Elsevier Science, Amsterdam, the Netherlands.
- McClintock, J.B., and Pearse, J.S. 1986. Organic and energetic content of eggs and juveniles of antarctic echinoids and asteroids with lecithotrophic development. *Comp. Biochem. Physiol. A Comp. Physiol.* **85**: 341–345.
- McEdward, L.R. 1997. Reproductive strategies of marine benthic invertebrates revisited: facultative feeding by planktotrophic larvae. *Am. Nat.* **150**: 48–72.
- McEdward, L.R., and Carson, S.F. 1987. Variation in egg organic content and its relationship with egg size in the starfish *Solaster stimpsoni*. *Mar. Ecol. Prog. Ser.* **37**: 159–169.
- McEdward, L.R., and Chia, F.-S. 1991. Size and energy content of eggs from echinoderms with pelagic lecithotrophic development. *J. Exp. Mar. Biol. Ecol.* **147**: 95–102.
- McEdward, L.R., and Coulter, L.K. 1987. Egg volume and energetic content are not correlated among sibling offspring of starfish: implications for life history theory. *Evolution*, **41**: 914–917.
- McEdward, L.R., and Morgan, K.H. 2001. Interspecific relationships between egg size and the level of parental investment per offspring in echinoderms. *Biol. Bull. (Woods Hole)*, **200**: 33–50.
- McEdward, L.R., Carson, S.F., and Chia, F.-S. 1988. Energetic content of eggs, larvae, and juveniles of *Florometra serratissima* and the implications for the evolution of crinoid life histories. *Int. J. Invertebr. Reprod. Dev.* **13**: 9–22.
- Menge, B.A. 1970. The population ecology and community role of the predaceous asteroid, *Leptasterias hexactis* (Stimpson). Ph.D. thesis, University of Washington, Seattle, Wash.
- Menge, B.A. 1974. Effect of wave action and competition on brooding and reproductive effort in the seastar, *Leptasterias hexactis*. *Ecology*, **55**: 84–93.
- Menge, B.A. 1975. Brood or broadcast? The adaptive significance of different reproductive strategies in two intertidal seastars *Leptasterias hexactis* and *Pisaster ochraceus*. *Mar. Biol.* **31**: 87–100.
- Niesen, T.M. 1973. Population and reproductive biology of the six-rayed sea star *Leptasterias hexactis* on the protected outer coast. Ph.D. dissertation, University of Oregon, Eugene, Oreg.
- Osterund, H.L. 1918. Preliminary observations on the development of *Leptasterias hexactis*. *Publ. Puget Sound Biol. Stn. Univ. Wash.* **2**: 1–15.
- Paine, R.T. 1971. The measurement and application of the calorie to ecological problems. *Annu. Rev. Ecol. Syst.* **2**: 145–164.
- Parsons, T.R., Maita, Y., and Lalli, C.M. 1984. A manual of chemical and biological methods for seawater analysis. Pergamon Press, New York.
- Rumrill, S.S. 1990. Natural mortality of marine invertebrate larvae. *Ophelia* No. 32: 163–198.
- Sewell, M.A. 1996. Mortality of pentactulae during intraovarian brooding in the apodid sea cucumber *Leptosynapta clarki*. *Biol. Bull. (Woods Hole)*, **190**: 188–194.
- Smith, C.C., and Fretwell, S.D. 1974. The optimal balance between size and number of offspring. *Am. Nat.* **108**: 499–506.
- Strathmann, M.F. 1987. Reproduction and development of marine invertebrates of the northern Pacific Coast. University of Washington Press, Seattle, Wash.
- Strathmann, R.R. 1985. Feeding and nonfeeding larval development and life-history evolution in marine invertebrates. *Annu. Rev. Ecol. Syst.* **16**: 339–361.
- Strathmann, R.R., and Strathmann, M.F. 1982. The relationship between adult size and brooding in marine invertebrates. *Am. Nat.* **119**: 91–101.
- Strathmann, R.R., Strathmann, M.F., and Emson, R.H. 1984. Does limited brood capacity link adult size, brooding, and simultaneous hermaphroditism? A test with the starfish *Asterina phylactica*. *Am. Nat.* **123**: 796–818.
- Thompson, R.J. 1983. The relationship between food ration and reproductive effort in the green sea urchin, *Strongylocentrotus droebachiensis*. *Oecologia*, **56**: 50–57.
- Turner, R.L., and Lawrence, J.M. 1979. Volume and composition of echinoderm eggs: implications for the use of egg size in life-history models. *In* *Reproductive ecology of marine invertebrates*. Edited by S.E. Stancyk. University of South Carolina Press, Columbia, S.C. pp. 25–40.
- Underwood, A.J. 1981. Techniques of analysis of variance in experimental marine biology and ecology. *Oceanogr. Mar. Biol. Annu. Rev.* **19**: 513–605.
- Vance, R.R. 1973. On reproductive strategies in marine benthic invertebrates. *Am. Nat.* **107**: 339–352.