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Evidence for enhanced late-stage larval quality, not survival, through maternal carry-over effects in a space monopolizing barnacle

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ABSTRACT

Understanding the effects of maternal and pelagic resource allocation on larval traits is essential to better understand population dynamics of marine benthic invertebrates. We tested how different levels of food supply to adult barnacles and their feeding larvae (nauplii) might alter survival to the settling cyprid larval stage and cyprid quality. Median development time did not vary, except when both parents and larvae were given a low food supply, which delayed the time to metamorphosis by over 40%. Survival to the cyprid stage was only affected by larval feeding, which doubled in better-fed nauplii. In contrast, cyprid size showed a more complex response, prone to additive effects of maternal and larval provisioning. Moreover, the resulting size-range observed for experimental cyprids (spanning over 70% of the minimum cyprid size) mirrored the variation found in the coastal plankton, suggesting that food supply may exert similar effects in nature. Given that barnacles nearly saturate available habitat under favorable conditions, maternal allocation resulting in enhanced late-stage larval quality may be adaptive since competition for available settling space is likely intense. On the other side, severe resource limitation through embryogenesis and larval development may impose delayed metamorphosis and thus enhanced potential for transport and the colonization of marginal habitats, where intraspecific competition may be lower and larval quality less critical.

KEYWORDS: *Chthamalus bisinuatus*; supply side ecology; sessile invertebrates; South Atlantic; tropical coast

INTRODUCTION

Recruitment is an important process controlling population dynamics, but key processes determining input to mature stocks are still not well understood. In marine invertebrates with complex life cycles, the supply of final-stage larvae to benthic habitat has long been considered a critical factor controlling recruitment success (e.g. Jenkins et al., 2008; Pineda et al., 2009). However, larval supply can be largely decoupled from settlement rate (Miron et al., 1995; Moreira et al., 2007), and settlers of the same larval pool can show quite different performance once established in the benthic environment (growth and survival rates combined; Jarrett, 2003; McQuaid & Phillips, 2006), suggesting that larval physiological conditions, i.e. larval quality, may play a very important role. Indeed, an increasing number of studies using different proxies of larval quality, such as size and organic content, have shown that good-quality late-stage larvae of barnacles and crabs develop into juveniles that grow faster and survive longer (e.g. Jarret, 2003; Emlet & Sadro, 2006; Giménez, 2010). Effects of larval quality may endure until adulthood, as shown for barnacles (Torres et al., 2016) and for colonial bryozoans (Marshall et al., 2003). In the case of bryozoans, colonies originating from large larvae reached maturity earlier and produced more embryos compared to colonies originating from smaller larvae (Marshall et al., 2003). Converging evidence from experimental work on different marine invertebrate groups has therefore led to a broad recognition of the importance of larval quality on recruitment strength and population dynamics (Burgess & Marshall, 2011; Van Allen & Rudolf, 2013).

Most marine invertebrates produce feeding or non-feeding larval stages that remain in the plankton for periods of several days to a few weeks (Shanks et al., 2003), during which pelagic transport may take larvae to distant habitat patches where population density and resource availability may be quite different from those at the natal location. How this uncertainty has shaped patterns of maternal provisioning to offspring, and particularly how it affects the number of potential settlers and their quality, is still an open question.

Patterns of maternal provisioning can be variable between the production of lecithotrophic and planktotrophic larvae. Lecithotrophic larvae rely solely on maternal provisioning during their often very short pelagic lifetime. In species producing such non-feeding larvae, environmental variables reflecting benthic habitat quality may shape patterns of maternal resource manipulation during embryogenesis (i.e. maternal effects) that modulate offspring number and quality, as well as their dispersal potential (Marshall & Keough, 2006; Marshall & Uller, 2007). The case of planktotrophic larvae is more complex because maternal provisioning acts in addition to pelagic feeding to improve general larval performance (e.g. Steer et al., 2004; Gagliano & McCormick, 2007). This may be achieved through different mechanisms, such as reducing early offspring mortality through the allocation of essential precursors for basic physiological processes (e.g. Salze et al., 2005; Freuchet et al., 2015), or by inducing plasticity of different functional larval traits (Hart, 1995; George, 1999; McAlister, 2007; Reitzel & Heylan, 2007). Combined effects of maternal and pelagic resource allocation in feeding larvae

may therefore modulate two key determinants of recruitment – the supply and quality of potential settlers. The relevance of such determinants would depend on the strength of density-dependent processes controlling adult benthic populations. For habitat patches where population density is well below carrying-capacity (i.e. recruitment limited sites –Doherty & Fowler, 1994) increased planktonic larval survival rates leading to greater larval supply may be particularly advantageous to mothers, enhancing overall fitness. At sites of high population density where competition for space and food resources is likely intense (i.e. habitat limited – Holbrook et al., 2000), enhanced quality of settling larvae may be critical.

The species used in this study, the intertidal barnacle *Chthamalus bisinuatus* (Pilsbry), is the most common cirripede species along the subtropical Southeastern Atlantic, dominating the upper part of the eulittoral zone of rocky shores, where they frequently cover 80% or more of available substrates (Bueno et al., 2010). These filter feeding barnacles reproduce through cross fertilization - although some cases of self-fertilization have been reported for this genus (Barnes & Crisp, 1956; Anderson, 1994) - and the fertilized eggs are carried in the mantle cavity until the nauplius stage I is fully developed. During embryogenesis, and in the absence of chronic thermal stress, reserve lipids available to mothers are proportionally transferred to their embryos (Freuchet et al., 2015). Once in the plankton, nauplii feed and molt through 6 different stages until they metamorphose into the late non-feeding larval phase, the cyprid. Time-series analyses indicated a pelagic larval duration of 11-12 days in *C. bisinuatus* during summer (Barbosa et al., 2016), but this period may depend on a number of factors, e.g. temperature and food availability, as observed for other chthamalids (Egan & Anderson, 1989; Burrows et al., 1999; Yan & Chan, 2001). The energy accumulated until the cyprid phase will directly influence the exploration capacity to find an adequate settlement surface and the success of metamorphosis into the first juvenile stage (e.g. Jarrett, 2003; Tremblay et al., 2007).

We examined here how trophic resources supplied to parental barnacles and pelagic larvae modulate proxies of three critical recruitment components: *cyprid yield*, as an outcome of larval survival to the competent stage, and a surrogate of the supply of potential settlers; *median development time*, as a metric of pelagic larval duration and dispersal potential; and *cyprid size*, as a proxy of larval quality at the competent stage. This size-to-quality approximation relies on the assumption that larger cyprids metamorphose to larger juveniles, which in turn experience higher growth rates (e.g. Emlet & Sadro, 2006), likely providing a competitive advantage over other recruits and a faster development to a size refuge from predation or bulldozing by limpets (Gosselin & Qian, 1997). Also, this proxy is backed by the allometric scaling of metabolic rate, which predicts a relatively more efficient use of resources as size increases, and explains the generally better performance observed for larger recruits of a wide array of marine invertebrates (Pettersen et al., 2015). The potential for variation in these three larval traits would depend on whether they respond to single or

101 combined manipulations of maternal and pelagic resource allocation. Effects on traits responding to
102 both resources could potentially be more extensive, especially if maternal and pelagic resources act
103 independently and in the same direction (additive effects). We thus anticipated extended effects
104 through maternal resource allocation (latent effects) combined with pelagic resource allocation, as
105 observed for other marine invertebrate larvae (e.g in echinoderms; George, 1996; McAlister & Moran,
106 2013). We tested whether maternal and pelagic allocation contributes to traits affecting larval quality,
107 i.e. cyprid size and traits associated with overall larval supply, i.e. survival rate and development time.
108 Development time was assumed to contribute to larval supply since additional time in the plankton
109 increases the risk of larval mortality through predation or transport away from suitable habitat. Carry-
110 over effects on larval supply would support selection of trait responses under recruitment limitation,
111 while such effects on larval quality would indicate selection of responses under habitat limitation.

112 **METHODS**

113 *Experimental design* - The effects of maternal and larval energy provisioning on the larval traits of
114 *Chthamalus bisinuatus* were assessed through manipulation of adult and larval food supply in a
115 factorial experiment [two levels of food supply; high (HF) and low (LF) for each life-stage] under
116 constant room temperature of 21°C, corresponding to air temperature at the time of sampling (see
117 below). Food supply consisted of an even mixture (i.e. equal cell volumes) of three microalgal species
118 cultured in the laboratory, the flagellates *Tetraselmis* sp. and *Isochrysis* sp. and the diatom
119 *Thalassiosira* sp. These species are common in the study region and are commonly used to rear marine
120 invertebrate larvae (Stone, 1989; Egan & Anderson, 1989; Burrows et al., 1999; Helm et al., 2004). This
121 mixture provided all essential fatty acids for larval growth (Ohse et al., 2015) and was delivered to both
122 adults and larvae, at concentrations of either 3 X 10^3 cells.ml^{-1} (HF) or 1 X 10^3 cells.ml^{-1} (LF). Such
123 concentrations were selected based on natural phytoplankton concentrations in the São Sebastião
124 Channel (unpublished data).

125 Adult barnacles (mean opercular length: 2.3 mm ± 0.7 mm SD) were obtained by extracting 40
126 rock chips with dense cover from a 100 m stretch of a rocky coastline at Calhetas Head (23° 49' 54'' S;
127 45° 31' 18'' W), São Paulo State, Brazil, on July 16th 2014. Rock chips hosting a mean barnacle cover of
128 40.13% ± 3.96% (SD) were evenly and randomly assigned to two tanks (one for HF and one for LF
129 treatments), ensuring that similar amount of adults were placed in each tank (54 x 40 x 20 cm). Adult
130 barnacles were subjected to experimental diets for 20 days; a period that exceeds the interval of
131 consecutive larval release events (12-13d; Kasten & Flores, 2013) and therefore should comprise the
132 whole oogenic cycle, as in the tropical congener *Chthamalus malayensis* (i.e. 10 days, Yan et al., 2006).
133 During this period, rock chips were submerged in filtered sea water (3 µm) containing the respective
134 feeding medium twice a day for 1h, so as to simulate the semidiurnal tidal regime that prevails in the

135 area. Owing to logistical limitations, we could not replicate tanks within levels of adult food supply.
136 However, a previous study on the same species, sampled from the same area and manipulated in the
137 same laboratory space, showed that (i) the quantity of food supplied to adult barnacles
138 proportionately translates to the quantity of lipid reserves in early larvae, and that (ii) larval survival
139 did not differ among replicate aquaria for the combination of experimental treatments (adult food
140 supply X thermal stress; Freuchet et al., 2015). As on that occasion, we carefully monitored
141 temperature, light conditions and ventilation within the laboratory and kept them as constant as
142 possible, which should have minimized the chances of any eventual environmental artifacts. Therefore,
143 we are confident that the effects we report for maternal food supply are an outcome of maternal
144 transference of resources, as previously observed.

145 After this manipulation, the nauplii released from experimental adult stocks were sampled by
146 filtering the water through a 100 µm mesh. These larvae were then transferred to 500 ml glass beakers
147 filled with 1 µm filtered seawater, aiming for a density of 0.5 nauplii.ml^{-1} (Moyse, 1960). A total of 12
148 beakers were prepared (n = 3 for each of the four treatment combinations, i.e. HF adult and HF larvae,
149 LF adult and LF larvae, HF adult and LF larvae, LF adult and HF larvae). Beakers were randomly
150 interspersed over the bench space to avoid any influence of environmental gradients within the
151 laboratory. Every other day, nauplii were fed and 90% of the water volume in the beakers was
152 renewed. After 10 days, beakers were carefully checked under a stereomicroscope (6-10X) for the
153 presence of cyprids (either swimming or settled on the bottom or sides of beakers), which were
154 removed from cultures, counted and preserved (70% ethanol) in separate vials. After the first
155 appearance of cyprids, beakers were inspected every other day and the experiment terminated when
156 no new cyprids were observed for a period of 10 days in any given beaker.

157 *Response variables* - Three different larval traits were separately quantified for each replicate beaker
158 in each treatment combination. Cyprid yield (CY) was estimated as the percentage of nauplii that
159 successfully survived to the cyprid stage. Median development time (MDT) was measured as the time
160 (in days) by which 50% of all cyprids were obtained. Cyprid size (CS) was measured as the lateral
161 carapace area (µm^2), from magnified images (63X) using the software ImageJ (Schneider et al., 2012).
162 Linear cyprid metrics were inadequate because of substantial variation in carapace shape. In order to
163 assess how results may actually translate to natural conditions, we compared the range of laboratory-
164 reared cyprid sizes across treatments to the estimated range of near shore cyprids along the São
165 Sebastião Channel. Plankton tows (150 µm) were taken from January 21st to March 28th 2013 at three
166 different sites separated by a few kilometers: Praia do Segredo (23°49'44.06" S, 45°25'21.06" W), Saco
167 Grande (23°49'55.23" S, 45°25'43.57" W) and Itassucê (23°49'56.77" S, 45°26'33.42" W). The general
168 morphology and size were used to identify cyprids in samples. Several different barnacle species are
169 found in the region, including balanids (mostly *Megabalanus* spp and *Balanus* spp), tetraclitids

170 (*Tetraclita stalactifera* [Lamarck]) and cththamalids. The cyprids observed in samples for the first two
171 are much larger and slender (higher length/height ratios) compared to chthamlid cyprids, which is
172 consistent with descriptions of larvae of those genera reared in the laboratory (e.g. Lacombe &
173 Monteiro, 1972; Chan, 2003; Nunes et al., 2017). Within chthamalids, the cypris larvae of
174 *Microeuraphia rhizophorae* (De Oliveira) and *Chthamalus proteus* (Dando & Southward) may be very
175 similar to the ones of *Chthamalus bisinuatus*. However, *M. rhizophorae* and *C. proteus* are estuarine
176 species (Farrapeira, 2008; Farrapeira, 2010; Klôh et al., 2013), and therefore the presence of their
177 larvae along the São Sebastião Channel is very unlikely. We thus assumed that chthamalid cyprids in
178 samples were all larvae of *C. bisinuatus*.

179 *Statistical analyses* - All three response variables were analyzed using separate ANOVAs. Maternal (HF
180 vs LF) and larval (HF vs LF) food supply were considered fixed factors. CY and MDT were analyzed using
181 2-way ANOVAs with a sample size of 3 (i.e. the number of replicate beakers). For CS, the unit of
182 replication was the individual cyprid, and therefore we included a random factor in the analysis,
183 'beaker', nested within the interaction of main factors (maternal and larval food supply). We ensured
184 a balanced design by considering sample size as the minimum number of cyprids found in any beaker
185 (n = 7). Excess cyprids were randomly removed from analyses. In all cases, the variances of data were
186 homogeneous (Cochran's C-test, $p > 0.05$). The Student-Newman-Keuls procedure was used for post-
187 hoc testing when needed (Underwood, 1997).

188 **RESULTS**

189 Manipulations of food supply generally produced positive effects on larval traits, but maternal and
190 larval treatments mediated effects in different ways (Table 1). Cyprid yield (CY) was influenced by only
191 one factor, the amount of food provided to the nauplii. Median development time (MDT) was
192 influenced by both the amount of food offered to the mothers and the larvae, and these effects were
193 interactive. Cyprid size (CS) was also influenced by the two factors, but these effects were additive.

194 Cyprid size was only influenced by the amount of food larvae were offered during their
195 development (p = 0.012; Table 1, Fig 1A), with no significant effects of maternal provisioning. The
196 relative number of nauplii that successfully reached the cyprid stage doubled from the low to the high
197 larval food supply treatment (13.3% ± 6.55% SD to 26.8% ± 6.64% SD), regardless of manipulations of
198 maternal food supply (Fig. 1A).

199 Median development time was influenced by an interactive effect of maternal and larval food
200 supply (p = 0.011; Table 1). MDT was markedly constant, around 18 to 21 days in all treatment
201 combinations, except when both mothers and larvae were supplied low food diets. In this case MDT

202 lasted around 27 days (Figure 1B), an increase of 8-9 days from the mean time observed in all other
203 treatments.

204 As observed in MDT, both maternal and pelagic allocation played a role in the determination
205 of cyprid size (maternal food supply p = 0.017 and larval food supply p = 0.016, Table 1). In this case,
206 however, effects were additive, with resource allocation from mothers and larvae apparently having
207 independent control, as evidenced by the lack of significance of the interaction term (maternal supply
208 X larval supply, p = 0.623; Table 1). Moreover, enhanced food supply to parents and larvae had
209 remarkably similar effects (c.a. 13% to 14% size increase; Figure 1C). CS was thus prone to extensive
210 variation (~28%), with the smallest size observed after parents and larvae were fed low food
211 concentrations, and the largest size recorded when high food concentrations were offered for both
212 (Figure 1C).

213 Remarkably, laboratory manipulations of a common diet given for both breeding barnacles
214 and their larvae were capable of reproducing the full range of cyprid sizes that can be observed in
215 coastal waters in the São Sebastião Channel (Figure 2).This shows that measures of cyprid size obtained
216 in the laboratory reflect natural variation at these sites, and that treatments of food supply likely
217 spanned the variation of trophic resources in nature.

218 **DISCUSSION**

219 This study shows that resource availability across ontogeny can alter key larval traits (larval
220 survival to the competent stage, development time and larval size) of marine invertebrates. Current
221 understanding of the importance of larval quality in determining benthic performance suggests that
222 such variation is likely to affect population dynamics. In particular, our results demonstrate that
223 maternal inputs can strongly modulate larval traits, indicating surprisingly large carry-over effects,
224 considering that nauplii are broadly considered planktotrophic larvae. Possible additive effects of
225 maternal provisioning were observed for cyprid size, a metric of larval quality, but not for cyprid yield
226 and development time, which rather control larval supply. Interestingly, carry-over effects of maternal
227 allocation seems to have the same effects as pelagic inputs to larvae in the determination of cyprid
228 size (13-14%). This is a surprising outcome considering that the species under investigation, as well as
229 most acorn barnacles (Anderson, 1994; Anil et al., 2010), undergoes a typical planktotrophic larval
230 development. Moreover, the variation in cyprid size obtained in the laboratory through rather simple
231 manipulations of food supply, offering just two experimental diets to adult barnacles and the larvae
232 they produced, was sufficient to replicate the full cyprid size range observed in the field. Although
233 other factors not addressed in this study may play a role, this finding validates extrapolations of
234 laboratory results to natural conditions and suggests that barnacle recruitment can be food-limited in
235 the area.

236 We argue that the larval responses observed may be adaptive for species that frequently face
237 limitation of food resources and potentially aggregate in dense populations which are habitat- rather
238 than recruitment-limited. Extended variation of cyprid size through embryonic allocation suggests that
239 habitat limitation has shaped maternal physiological responses over evolutionary time. In other words,
240 maternal provisioning may be of limited value for larvae settling on rocky shores with only a sparse
241 barnacle cover, but essential at crowded shores where environmental conditions are very favorable
242 for post-larval survival and growth, but available space for potential settlers is scant. Such generally
243 advantageous conditions can be found at wave-swept rocky intertidal shorelines located in productive
244 coastal stretches (Leigh et al., 1987), where the flow of food particles over reef habitat is optimal
245 (Sanford et al., 1994; Leonard et al., 1998). Under these circumstances, the density of chthamalid
246 barnacles may reach saturation, and the short supply of bare rock patches for new recruits will depend
247 on stochastic disturbance delivered mainly by heavy wave action (Barnes & Powell, 1950), particularly
248 where intraspecific competition is intense (Jenkins et al., 2008), or by barnacle mass mortality caused
249 by excessive heat stress (Chan et al., 2006; Harley, 2008). Newly available habitat patches may be
250 disputed by a large number of potential settlers, suggesting a scenario of local selection for high-quality
251 larvae. Positive carry-over effects of maternal resource allocation on cyprid size are likely
252 advantageous in this situation since they translate into better juvenile performance (Pechenik et al.,
253 1998; Moran & Emlet, 2001; Jarrett, 2003; Emlet & Sadro, 2006; Torres et al., 2016), improved
254 competitive ability (Bertness, 1989) and faster growth to a size-refuge from predation (Miller &
255 Carefoot, 1989; Gosselin & Qian, 1997). The benefits of maternal provisioning would therefore be
256 greatest if the bulk of recruits settle close to parental populations (e.g. Kingsford et al., 2002; Levin,
257 2006; Jones et al., 2009), where barnacle density tends to be higher than average.

258 The lack of significant effects of maternal provisioning on planktonic larval survival (and hence
259 on the numbers of potential recruits) could suggest a physiological mechanism which reduces resource
260 wastage, although this was not tested in this study. In this scenario, mothers would not invest energy
261 in facilitating an increase in the number of potential recruits through maternal provisioning as it may
262 make little difference in densely populated habitat patches. Enhanced planktonic larval survival will be
263 beneficial where larvae settle into populations which are recruitment-limited, or when future
264 environmental conditions are uncertain, according to the theory of life-history tactics (e.g. Stearns,
265 1976; Simons, 2007). Therefore, the possible adaptive value of increased larval supply would be
266 proportional to the probability of drifting away from the natal habitat, where the individual chance for
267 successful recruitment will be probably low. Here increased survival through planktonic feeding may
268 allow the colonization of novel habitats through propagule pressure, as framed in studies on invasive
269 biology (e.g. Lockwood et al., 2005). While we speculate here on potential adaptive explanations for
270 the lack of an effect of maternal provisioning on planktonic larval survival, it is important to recognize

that this lack of effect could also be the result of a physiological constraint. We did not discriminate here the effects of food supply on different naupliar stages, but other studies on the same species and in the same area suggest that early stages may be more tolerant to food shortage than late-stage nauplii (Freuchet et al., 2015; Barbosa et al., 2016), in accordance to the results obtained by Hentschel and Emlet (2000) on *Balanus glandula* Darwin. Early nauplii of *Chthamalus bisinuatus* may endure for up to 5 days without any exogenous food sources in the laboratory (Freuchet et al., 2015), suggesting that larvae released in oligotrophic waters may still survive and reach the cyprid stage if oceanographic conditions improve later on.

Our results also indicate that prolonged food shortage may lead to unusual offspring 'spillover' and connectivity among discrete rocky-shore habitats through delayed larval development to the cyprid stage. At 21°C, we estimated a shift from a remarkably steady development time of around 19 days to an extended 28 days period if adults and larvae experience food shortage. Adding to the fact that poorly-fed larvae may probably be less active, and therefore prone to more extensive drifting away from natal populations (Marta-Almeida et al., 2006; Butler et al., 2011), an extended larval period would imply a much higher dispersal potential. Larval loss owing to offshore advection and predation should be frequent for this more dispersive phenotype, but its role in the colonization of distant habitat may be important, showing how variable the recruitment process can be in this species. While these poorly-fed larvae would have a very low chance to successfully recruit to dense barnacle populations, where intraspecific competition is probably intense, they might be able to settle and thrive on marginal habitats, where percent substrate cover is low and bare rock is abundant. These processes are aligned to the original concept of metapopulation dynamics (Pulliam, 1998), in which competition and resource supply determine the chances of individual dispersal from source to sink habitats. Given that the mid-littoral barnacle zone, mostly occupied by *Chthamalus bisinuatus*, is a ubiquitous feature of rocky shores along the study region, it is very likely that sparser barnacle populations at more isolated rocky outcrops rely on episodic recruitment of individuals originating from places with surplus larval production. Extended dispersal of individuals undergoing delayed development to the cyprid stage may be an important means to connect marginal populations.

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COMPLIANCE WITH ETHICAL STANDARDS

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CONFLICT OF INTERESTS: The authors declare that they have no conflict of interest.

ETHICAL APPROVAL: All applicable international, national, and/or institutional guidelines for the care and use of animals were followed.

DATA AVAILABILITY STATEMENT

The datasets generated during and/or analysed during the current study are available from the corresponding author on reasonable request.

483 **Table 1.** Summary results of analyses of variance comparing the effects of maternal and larval food supply on cyprid traits. Statistical significance ($p < 0.05$) is
484 highlighted in bold.

	Cyprid Yield					Median Development Time					Cyprid Size				
	df	SS	MS	*F*	*p*	df	SS	MS	*F*	*p*	df	SS	MS	*F*	*p*
Maternal supply: M	1	25.9	25.9	0.54	0.549	1	36.8	36.8	11.03	**0.011**	1	11.1	11.1	8.93	**0.017**
Larval supply:L	1	501.4	501.4	10.39	**0.012**	1	90.8	90.8	27.23	**0.001**	1	11.4	11.4	9.16	**0.016**
M x L	1	22.5	22.5	0.33	0.514	1	36.8	36.8	11.03	**0.011**	1	0.3	0.3	0.26	0.623
Beaker (M x L)	-		-	-	-	-	-	-	-	-	8	9.9	1.24	1.51	0.168
Residual	8	386.2	48.3			8	26.7	3.33			72	59.0	0.82		
Total	11	936.1				11	190.9				83	91.7			

485

486 **Figure captions**

487 **Fig 1** Response of larval traits to the combined effects of maternal (= embryonic) and larval (= pelagic)
488 resource supply. CY: cyprid yield; MDT median development time for the cyprid stage; CS: cyprid size.
489 Error bars indicate +1SE. M: maternal; L: larval; 'ns': non-significant differences; *: $p < 0.05$; ** $p < 0.01$

490 **Fig 2** Size-frequency distribution of *Chthamalus bisinuatus* cyprid stages collected in the field and size
491 ranges (mean ± 1 SE) obtained in the laboratory after factorial manipulation of maternal (M) and larval
492 (L) food supply (high: HF vs low: LF). (1) M_{LF} / L_{LF}; (2) M_{LF} / L_{HF}; (3) M_{HF} / L_{LF}; (4) M_{HF} / L_{HF}

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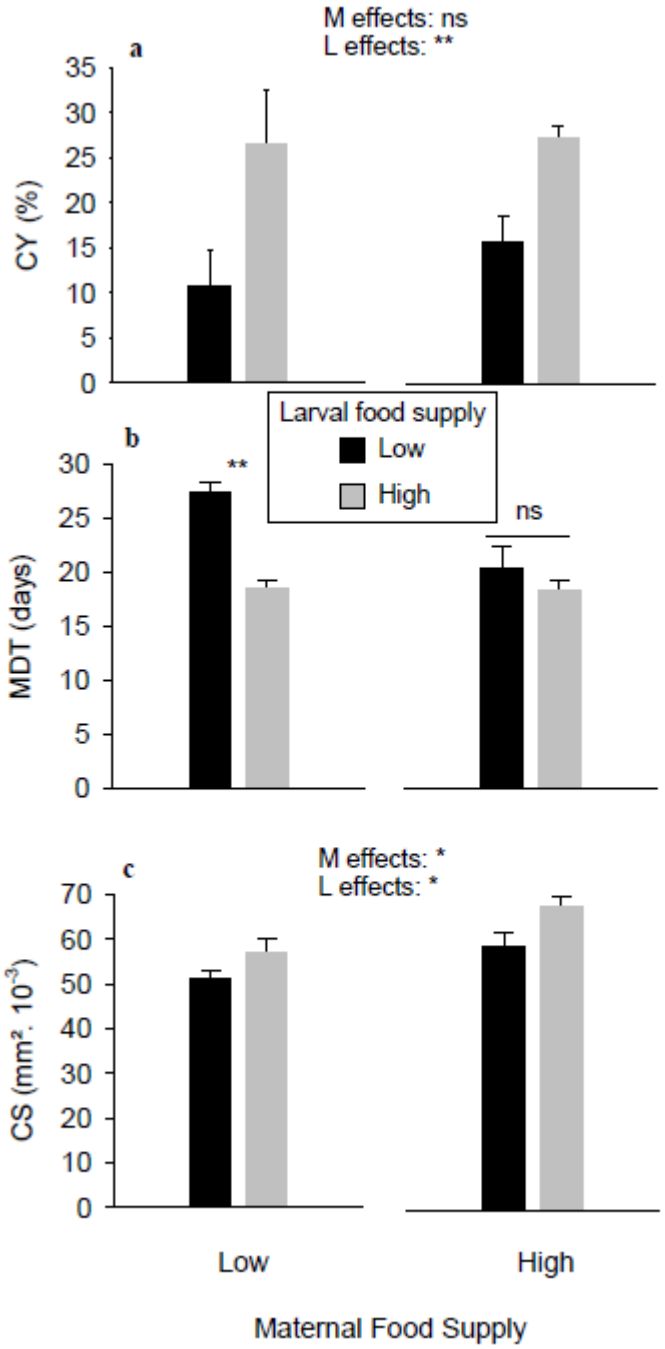

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496 Figure 1

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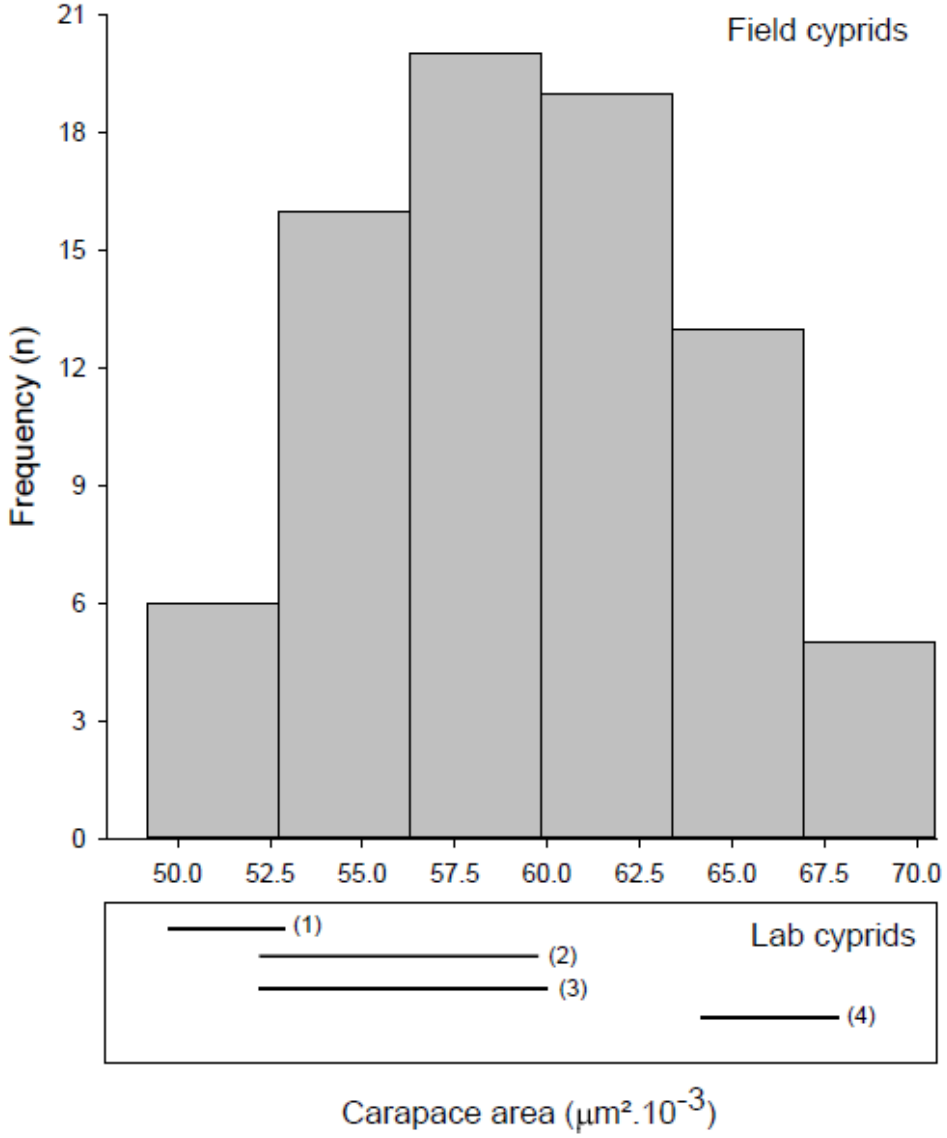

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499 Figure 2