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Plasticity of thermal tolerance and its relationship with growth rate in juvenile mussels (*Mytilus californianus*)

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Complex life cycles characterized by uncertainty at transitions between larval/juvenile and adult environments could favour irreversible physiological plasticity at such transitions. To assess whether thermal tolerance of intertidal mussels (*Mytilus californianus*) adjusts to post-settlement environmental conditions, we collected juveniles from their thermally buffered microhabitat from high- and low-shore locations at cool (wave-exposed) and warm (wave-protected) sites. Juveniles were transplanted to unsheltered cages at the two low sites or placed in a common garden. Juveniles transplanted to the warm site for one month in summer had higher thermal tolerance, regardless of origin site. By contrast, common-garden juveniles from all sites had lower tolerance indistinguishable from exposed site transplants. After six months in the field plus a common garden period, there was a trend for higher thermal tolerance at the protected site, while reduced thermal tolerance at both sites indicated seasonal acclimatization. Thermal tolerance and growth rate were inversely related after one but not six months; protected-site transplants were more tolerant but grew more slowly. In contrast to juveniles, adults from low-shore exposed and protected sites retained differences in thermal tolerance after common garden treatment in summer. Both irreversible and reversible forms of plasticity must be considered in organismal responses to changing environments.

1. Introduction

Animals' performance in stressful environments could depend on gene–environment 'match–mismatch' [1] or on physiological plasticity expressed either during development or in response to variation in the adult environment [2,3]. Delineating the relative roles and magnitudes of these phenomena is particularly relevant to forecasting the effects of global change [4,5]. Reversible adult plasticity can be an effective strategy in sessile organisms exposed to stressful environments after recruitment to the adult population [6], for example, by allowing long-lived organisms to adjust to predictable environmental changes, such as seasonal temperature cycles [7]. Adult plasticity affects thermal tolerance in diverse terrestrial and marine species [8].

By contrast, developmental plasticity is generally considered irreversible and results from individuals experiencing different environments before adulthood [9]. For example, developmental temperature has been shown to induce irreversible phenotypic changes in *Drosophila*, parasitoid wasps and zebrafish [10–12]. Developmental plasticity may be adaptive in organisms with high gene flow among populations inhabiting disparate environments [13], in cases of high spatial variability within populations [14], and for species that experience considerable temporal environmental variation across generations [14]. These scenarios result in offspring inhabiting environments that vary unpredictably from the parental environment. Complex life cycles, particularly in the numerous

species in which different life-history stages occupy distinct environmental niches, could similarly favour the expression of irreversible plasticity during periods of transition between environments [13]. Aside from studies on insects [10,11,15], relatively little is known about the flexibility of physiological mechanisms during these transitions.

Here, we investigate plasticity of thermal tolerance around such a life-history transition—recruitment of post-larval juveniles to adult aggregations—in the ecologically dominant rocky intertidal mussel *Mytilus californianus*. The dynamic nature and spatial heterogeneity of the rocky intertidal zone [16], along with aspects of this species' life history, may promote both reversible and irreversible forms of plasticity. Adult *M. californianus* are sessile, can live for 50+ years [17], and experience predictable seasonal temperature changes. Accordingly, adult mussels exhibit seasonal plasticity in gill phospholipid membrane composition [18] and in heat shock protein (Hsp) induction temperature [19]. *Mytilus californianus* also experience large temperature fluctuations associated with the semidiurnal tidal cycle, corresponding with short-term plasticity in membrane composition [16] and defenses against oxidative stress [20]. Other aspects of this mussel's life history could promote irreversible, developmental plasticity of thermal tolerance [21]. Adults produce long-lived planktonic larvae (up to 45 days; [22]) that can disperse at least 50 km [23], thereby facilitating gene flow [24] and probably increasing environmental variation across generations. There also is pronounced spatial variability in body temperature within mussel beds [16], further reducing the predictability of a settling larva's future adult micro-environment. However, expressing the regulatory mechanisms that allow juveniles to switch between phenotypes may be costly [7], potentially constraining other processes such as growth [12]. Given *M. californianus*' role as an ecosystem engineer [25], if developmental plasticity does allow juveniles to match their thermal tolerance to their post-recruitment micro-environment—even if such adjustments are limited in magnitude [5,26]—the implications could extend throughout the rocky intertidal community.

We assessed thermal tolerance of juvenile and adult *M. californianus* from multiple intertidal sites and measured growth rates of juveniles to: (i) determine whether fixed genetic differences or phenotypic plasticity determine juveniles' thermal tolerance, (ii) examine potential trade-offs between juveniles' thermal tolerance and growth, and (iii) compare plasticity of thermal tolerance in juveniles and adults from the same sites. Our data suggest that juveniles irreversibly adjust thermal tolerance based on post-recruitment environmental conditions. Such plasticity and its associated costs can affect how taxa that experience unique environments throughout development will cope with environmental change.

2. Material and methods

(a) Plasticity of thermal tolerance and growth rate in juvenile mussels

Juvenile mussels (*M. californianus*, $n = 735$, shell length 5–14.5 mm) were collected in June 2016 from high- and low-shore beds at both wave-exposed (cool) and wave-protected (warm) microsites at Hopkins Marine Station (HMS) in Pacific Grove, CA, USA (36.6217° N, 121.9043° W) [16,20]. Low- and high-shore sites (approx. 1.5 and approx. 1.75 m above mean lower low water, respectively) within each microsite were situated within 10 m of

each other. These exposed and protected microsites differ in wave splash, solar irradiance and mussel body temperatures. Individuals were labelled with coded apiarist tags. Juveniles of this size still inhabit algal tufts and/or interstices of the byssal thread network under and between larger adults [22]; these microhabitats shelter juveniles from the direct solar heating and desiccation risk characteristic of the adult micro-environment [27].

(i) Juvenile one-month and six-month reciprocal transplant studies

Mussels from each of the four origin sites were transplanted to a low-shore location at either their original wave-exposure site or the other site during early summer. Juveniles from each site were placed inside a mesh bag, enclosed within a stainless steel anti-predator cage (3.3 mm mesh), and attached to an acrylic plate (electronic supplementary material, figure S1). The plates were bolted onto open patches of rock, exposing the juveniles to unsheltered conditions, on 21 June 2016. Transplanted individuals were retrieved after one month (July) or six months (December) (electronic supplementary material, figure S2).

Survival was high during the one-month experiments at both transplant sites (99%) (electronic supplementary material, table S1). Owing to tag loss and/or uncertain fate of numerous individuals, survival could not be quantified in the six-month experiment.

(ii) Juvenile one-month common garden study

Shifts in juveniles' thermal physiology could change with recent environmental experience or simply with age [28]. To begin to tease apart these factors, individuals from each origin site ($n = 24$ per site) were exposed to a laboratory common garden treatment that ran in parallel with the one-month transplant study. These mussels were continuously submerged in a flow-through seawater tank (temperature 12.4–15.2°C) at HMS. Phytoplankton food in the flow-through seawater was supplemented once per day by adding approximately 1 ml of concentrated Shellfish Diet 1800 (Reed Mariculture) and stopping water flow for approximately 1.5 h.

(iii) Juvenile six-month reciprocal transplant plus common garden study

Juveniles from the six-month transplant study not assayed immediately for thermal tolerance were kept continuously submerged in common-garden conditions (temperature 14.3°C–15.4°C) in a recirculating aquarium system at Loyola Marymount University (LMU) and fed as described above for one month after retrieval from the field. Thermal tolerance assays were then repeated to test for persistent effects of transplant environment. Owing to tag losses, the origin wave exposure was unknown for these individuals (electronic supplementary material, figure S2).

(iv) Juvenile growth rate

Initial and final measurements of shell length, which is correlated with mass in *Mytilus* [29], were made with digital calipers and used to determine a daily growth coefficient. This coefficient represents proportional change in length per day, assuming a constant growth rate throughout the study. For individuals in the six-month field study, growth coefficients were calculated separately for zero to one month and one to six months. Owing to tag losses, growth could not be quantified in the six-month reciprocal transplant plus common garden study.

(b) Transplant site temperature measurements

To estimate thermal variation between the transplant sites, iButton temperature dataloggers (model DS1921G, Maxim Integrated, San Jose, CA, USA) were embedded in silicon-filled adult

mussel shells (approx. 55 mm shell length; [16]) and deployed within the transplant cages ($n = 5$ per site). Although these adult mimics record different 'body' temperatures than those experienced by juveniles [27], they allow between-site comparisons. The dataloggers recorded temperature (0.5°C resolution) every 30 min for 30 days in summer. They were then reprogrammed to record every 120 min (owing to memory constraints) for the six-month field study. This longer sampling interval probably missed many daily maximum temperatures [30], but it provides an overall assessment of thermal differences between the sites. Data from all functional dataloggers at each site and time were averaged for analyses.

(c) Plasticity of thermal tolerance in adult mussels
In July 2016, we collected approximately 50 adult mussels (40–55 mm shell length) from both the exposed-low and protected-low origin sites. Half of these individuals were assayed for thermal tolerance within 6 days of collection. The remaining individuals were placed in common garden conditions for 19–22 days before repeating the thermal tolerance assay.

(d) Thermal tolerance assays
Thermal tolerance assays were conducted in air to mimic hot, low-tide conditions. To determine the thermal tolerance of field-acclimatized juveniles from each of the four origin sites, we exposed individuals to one of three temperatures ($n = 6$–8 per temperature per site). Each individual was placed in a 0.6 ml microcentrifuge tube, and a 1×1 cm seawater-saturated laboratory tissue was placed at the bottom of the tube to maintain humidity. The tubes were placed in a thermalcycler, which ramped temperature up from 20°C at 0.1°C min^{-1} [16,30]. Upon reaching the target temperature (35.8°C, 37.7°C or 38.6°C), temperature was held constant for 1 h before ramping down to 20°C at 0.3°C min^{-1}. Target temperatures were chosen based on the adult LT$_{50}$ for this species, approximately 38°C [13]. For subsequent assays, we preserved the ramping profiles but used a larger chamber containing a seawater-saturated laboratory tissue: common-garden and one-month reciprocal transplant juveniles in 1.5 ml microcentrifuge tubes; six-month juveniles in 15 ml tubes; adults in 50 ml tubes. The tubes were submerged in a water bath programmed with the ramp profile. Air temperature inside one to two tubes was monitored with a thermocouple to ensure consistency.

Immediately after the ramp, individuals were returned to a flow-through tank at HMS (or the aquarium system at LMU) and fed once per day as described above. Survival was scored after 7 days. Open mussels that did not respond to mechanical stimulation by closing, as well as mussels that opened upon mechanical stimulation, were scored as dead.

(e) Statistics
(i) Thermal tolerance
Survival data were analysed using the *glmulti* package in *R* [31] to perform exhaustive model selection, retaining the terms that provided the most informative binomial general linear model. The full model for juveniles at one month included origin and treatment (field-acclimatized, transplant site (exposed or protected) or common garden) as factors, with assay temperature as a continuous covariate. We included interactions between origin and temperature and between origin and treatment to allow for origin-specific patterns. For the juvenile six months plus common garden analysis, the full model included treatment (exposed or protected), origin shore height (high or low), and temperature, as well as interactions of treatment with the other two predictors. For adults, the full model included origin site and treatment (field-acclimatized or common garden), temperature and interactions of

treatment with the other two predictors. The best model was chosen using a modified Akaike's information criterion (AIC$_c$) adjusted for small sample sizes [32]. Statistical significance ($\alpha = 0.05$) and odds ratios are presented from the best model for each analysis. In each analysis, there was another equally parsimonious model (delta AIC$_c$ < 2.0) [33], but these alternate models did not change the interpretations.

(ii) Growth rate
To analyse growth data, we used the model selection approach described above. For juvenile growth from zero to one month ($n = 658$, electronic supplementary material, table S2), the full model included origin site, treatment and their interaction. For the 48 individuals measured in both the zero to one month and the one to six month intervals, the full model included origin site, treatment and time interval. We also included interactions between origin site and treatment, origin site and time, and treatment and time to allow for more complex patterns. For both analyses, initial shell length was included as a continuous covariate to account for allometric effects.

3. Results
(a) Transplant site temperatures
(i) One-month transplant
The average daily range of temperature was three times greater at the protected site (8.1°C versus 2.7°C) in the one-month study (electronic supplementary material, figure S3a and table S3). The exposed site reached a maximum of 20.3°C; the protected site exceeded this temperature on 15 days, reaching a maximum of 31.3°C.

(ii) Six-month transplant
During the six-month study the protected site had a greater mean daily temperature range, reaching high temperatures more frequently. Daily temperature range decreased at both sites from summer to autumn, and the absolute difference between the sites grew smaller (electronic supplementary material, table S3).

(b) Origin site and recent environment influence juvenile thermal tolerance
(i) One month
The best model for juvenile survival included significant effects of treatment, origin and temperature (table 1). The odds of survival for transplants to the protected site were estimated at more than 10 times those for individuals held in common garden ($p < 0.001$) or transplanted to the exposed site ($p < 0.001$); protected-site transplants had roughly four times the odds of survival as field-acclimatized mussels ($p = 0.019$) (table 1; *post hoc* p-values generated using *multcomp R* package [34]). These treatment differences were most pronounced at 37.7°C (figure 1). It is possible the slightly lower survival of field-acclimatized juveniles (figure 1) was owing to low oxygen content of their small tubes at the end of the heat ramps, but this is unlikely given the propensity of *M. californianus* to seal its valves upon emersion [30] and thus reduce its oxygen consumption rate [35]. Juveniles from the exposed-low origin site had less than one-fifth the odds of survival relative to the exposed-high site ($p = 0.010$) and the protected-high site ($p = 0.005$) (table 1). One other model was supported and

Table 1. Statistical summaries of the best binomial models of thermal tolerance. (The first row for each analysis contains *p*-values for each of the included predictors (values in bold are significant, $\alpha < 0.05$), and subsequent rows contain corresponding odds ratios for each term in the model (with 95% confidence intervals). Predictors left blank were excluded from the final model. Odds ratios were produced by exponentiating the model β coefficients and indicate the odds of survival relative to the reference level for each variable. Quasi-complete separation required some model coefficients to be derived by exact conditional logistic regression using the *elrm* package in R (*). Although the interaction between origin and treatment was initially included in the full model for all analyses, it was not included in any of the best models and is thus not shown here.)

analysis	origin[a]	treatment[b]	temperature	origin × temperature[c]
juvenile one month	**$p = 0.002$**	**$p < 0.001$**	**$p < 0.001$**	
	highprot = 1.13 (0.42, 3.03)	exp = 0.73 (0.24, 2.22)	temp = 0.09	
	lowexp = 0.17 (0.05, 0.50)	field = 2.63 (0.90, 8.09)	(0.05, 0.14)	
	lowprot = 0.69 (0.25, 1.88)	prot = 10.82 (3.84, 34.00)		
juvenile six months + common garden			**$p < 0.001$**	
			temp* = 0.75	
			(0.00, 0.85)	
adult one month			**$p < 0.001$**	**$p < 0.001$**
			temp = 0.04	temp × prot = 1.12
			(0.01, 0.12)	(1.05, 1.24)

[a]Reference = high exposed.
[b]Reference = common garden.
[c]Reference = exposed × temperature (adults only).

included effects of treatment, temperature and the interaction between origin site and temperature (electronic supplementary material, table S4).

To ensure that analysing data for juvenile thermal tolerance that were collected one month apart did not introduce a confounding factor of development time, we reran the analysis without the field-acclimatized treatment. The top two, equally parsimonious models were identical to those derived using the full dataset (not shown).

(ii) Six months
Owing to mussel losses, tolerance was only assessed at the two higher temperatures (37.7°C and 38.6°C) that showed the greatest treatment differences at one month. No individuals survived these exposures.

(iii) Six months plus common garden
Although there was a trend of mussels from the protected transplant site showing higher survival at the lowest temperature, the best model included only temperature ($p < 0.001$; table 1). An equally parsimonious model included effects of treatment and temperature ($p = 0.189$ and $p < 0.001$, respectively; electronic supplementary material, table S4). No mussels survived at 37.7°C or 38.6°C (electronic supplementary material, figure S4).

(c) No evidence for plasticity of thermal tolerance in adults
Adults from the protected site were estimated to have 72 times higher odds of survival at 37.7°C than those from the exposed site, both before and after common gardening. Unlike juveniles, adult tolerance did not change with common-garden acclimation (figure 2). The best model included significant effects of temperature and an origin-by-temperature interaction (table 1). An equally parsimonious model included

significant effects of temperature and origin (electronic supplementary material, table S4).

### (d) Juvenile growth rate varies between transplant sites
(i) One month
The best model for growth rate over one month included origin ($p = 0.992$) and three significant terms: treatment, an origin-by-treatment interaction, and a negative effect of shell length (table 2). Overall, transplants to the exposed and protected sites grew 2.45 and 0.75 global standard deviations faster, respectively, than mussels in the common garden (which grew very slowly). For the interaction, juveniles originating from high-shore sites and transplanted to the exposed site grew at more than twice the rate of those at the protected site ($p < 0.001$ for each *post hoc* comparison; figure 3). There were no equally parsimonious models.

(ii) Six months
The best model for juvenile growth over six months included significant effects of transplant site, time interval, their interaction and a negative effect of shell length (table 2). There were no equally parsimonious models. While growth rate at the protected site was consistently slow throughout the study, the interaction term indicated that growth at the exposed site was rapid during the first month (coinciding with reduced thermal tolerance) but slowed to a rate comparable to that of protected-site transplants in the one to six month interval (figure 4).

4. Discussion
Our reciprocal transplant and common garden experiments suggest that recent environmental experience contributes to the thermal tolerance of post-settlement juvenile *M. californianus*, but not of adults from the same intertidal sites.
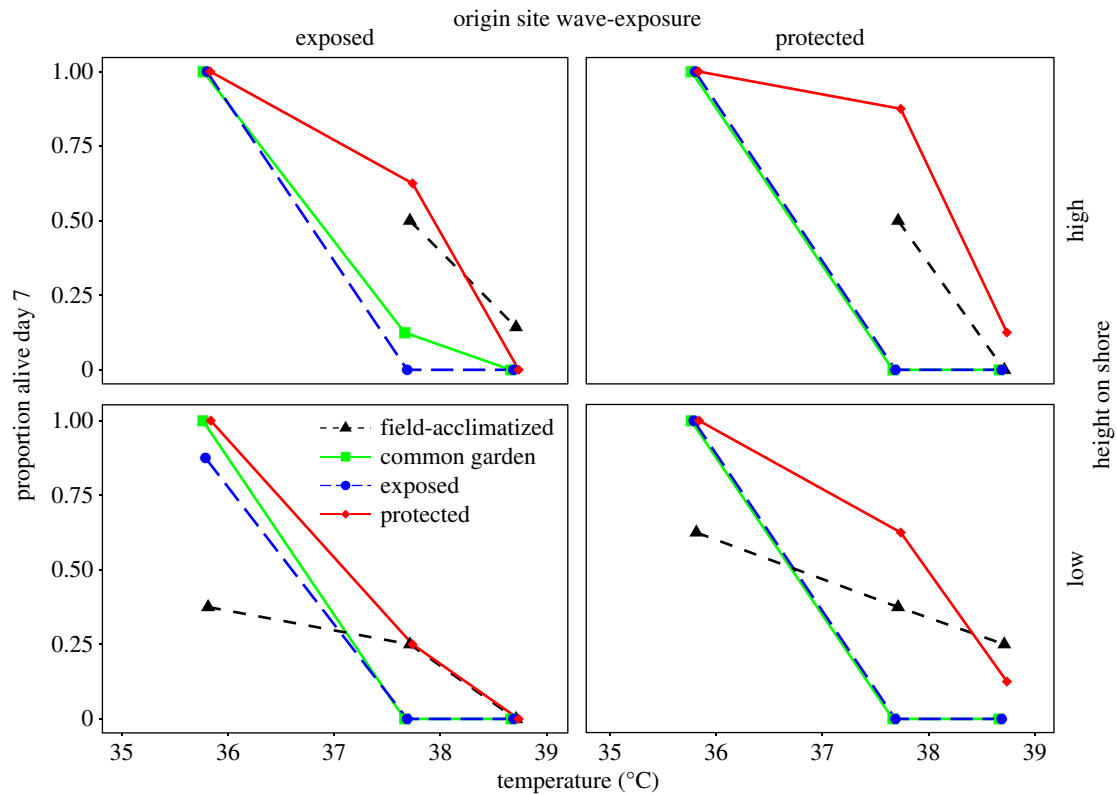
Figure 1. Survival of juvenile mussels after exposure to three heat stress temperatures in the one-month experiment. Mussels from each of four origin sites were assayed for thermal tolerance following four treatments (electronic supplementary material, figure S2). Field-acclimatized mussels from the high sites were not tested at 35.8°C. Points are horizontally staggered to aid in interpretation. The proportion alive on day 7 is the *y*-axis for all panels. 'High' and 'low' height on shore refer to the top and bottom row of panels, respectively.
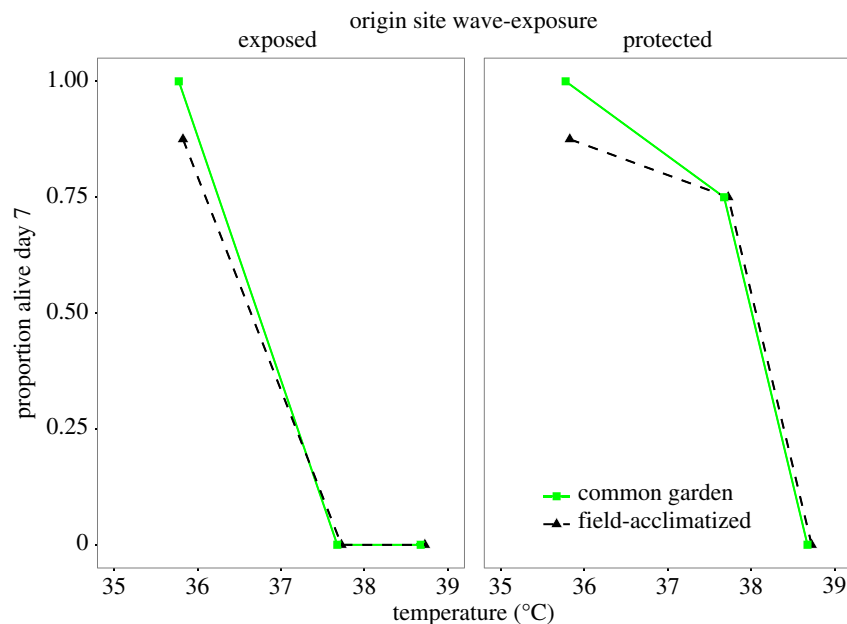
Figure 2. Survival of adult mussels from wave-exposed and wave-protected low sites after exposure to three heat stress temperatures. Adults were assayed directly from the field or after common-garden acclimation.

Seasonal shifts in tolerance are superimposed on these developmental effects. Our results also indicate a potential trade-off between thermal tolerance and growth for juveniles during summer when food is abundant. It is difficult to prove that the changes we observed are irreversible, because one cannot test the thermal tolerance of an individual multiple times. However, these results are consistent with the existence of a developmental window following settlement during

which juveniles adjust their thermal tolerance to their micro-environment.

(a) Short-term plasticity of thermal tolerance is restricted to juveniles

Evolution of a thermally sensitive window during development in *M. californianus*, as has also been observed in lake

Table 2. Statistical summaries of the best models of juvenile growth rate for the one-month and six-month field studies. (The first row for each analysis contains p-values for each of the included predictors (values in bold are significant, $\alpha < 0.05$). Predictors left blank were excluded from the final model. Partial η^2 quantifies variance explained by each predictor after controlling for all other predictors. Model β coefficients were normalized by the global standard deviation (1.679×10^{-3} or 1.195×10^{-3}, respectively); resulting values indicate the effect (with 95% confidence intervals) of each term as a multiple of standard deviation, relative to the reference level. No equally parsimonious models were found; model weights were 1.00 and 0.76, respectively.)

one-month experiment

origin[a]	treatment[b]	origin × treatment[c]	initial shell length
$p = 0.992$	**$p < 0.001$**	**$p < 0.001$**	**$p < 0.001$**
$\eta^2_{partial} = 0.025$	$\eta^2_{partial} = 0.467$	$\eta^2_{partial} = 0.095$	$\eta^2_{partial} = 0.075$
	exp = 2.45 (2.12, 2.79)	highprot × exp = −0.39 (−0.88, 0.09)	length = −0.10 (−0.13, −0.08)
	prot = 0.75 (0.41, 1.08)	lowexp × exp = −0.86 (−1.31, −0.40)	
		lowprot × exp = −0.88 (−1.34, −0.42)	
		highprot × prot = −0.03 (−0.51, 0.45)	
		lowexp × prot = 0.27 (−0.18, 0.73)	
		lowprot × prot = 0.26 (−0.20, 0.72)	

six-month experiment

| treatment[b]	time interval[d]	initial shell length	time × treatment[e]
$p < 0.001$	**$p < 0.001$**	**$p = 0.006$**	**$p < 0.001$**
$\eta^2_{partial} = 0.149$	$\eta^2_{partial} = 0.011$	$\eta^2_{partial} = 0.075$	$\eta^2_{partial} = 0.154$
prot = −1.53 (−2.06, −1.01)	1−6 = −1.32 (−1.97, −0.67)	length = −0.12 (−0.20, −0.03)	prot × 1−6 = 1.57 (0.82, 2.33)

[a]Reference = high exposed.
[b]Reference = common garden for one-month experiment, exposed site for six-month experiment.
[c]Reference = high exposed, common garden.
[d]Reference = 0−1 month.
[e]Reference = 0−1 month × exposed.
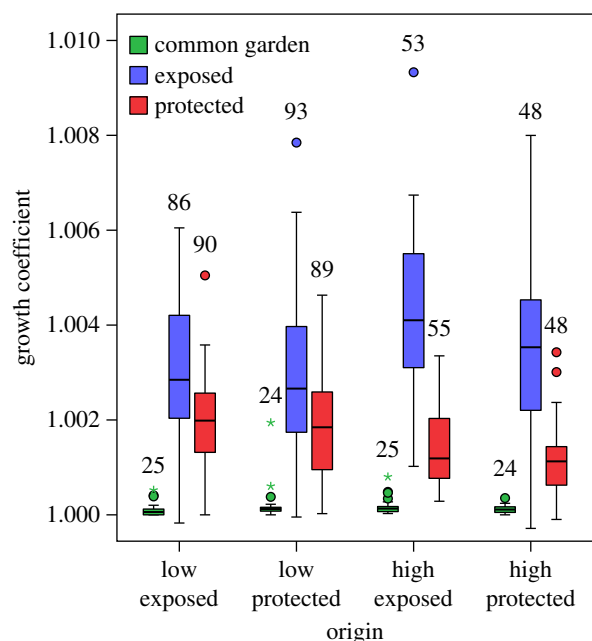
Figure 3. Box plot of daily growth coefficients of juvenile mussels from the four origin sites over one month in summer. Mussels were measured for total shell length immediately after collection from the field and after one month in each treatment. A growth coefficient of 1.0 indicates no growth. Sample sizes are listed above each bar.

whitefish and *Lepidoptera* butterflies [36,37], could be because of unpredictability around the transition between distinct juvenile and adult microhabitats. Juveniles settle in algal tufts or among the byssal threads of adults [22,38]; these microhabitats are moist and sheltered from the sun. The ensuing transition to relatively unsheltered, but spatially variable [16], adult microhabitats as recruits grow could favour physiological plasticity around this period. Subsequently, adult mussels are essentially sessile, with limited capacity to modify their thermal exposure [30]. Consequently, resources devoted to maintaining a plastic juvenile state might best be reallocated to adult growth and reproduction. Analogous trade-offs have been thoroughly examined in plants (e.g. [39]), although rigorous experimental designs are required to quantify the associated costs [40].

The higher thermal tolerance in juveniles transplanted to the wave-protected site in summer is probably owing to more frequent episodes of high body temperature during low tide. The average daily temperature range at the protected site was triple that at the exposed site, although maximum datalogger temperatures never approached within 4.5°C of the lowest assay temperature. Similar correlations between high temperature variability and thermal tolerance have been observed in a range of taxa [12,41–44]. In the six months plus common garden study, there was a similar, albeit non-significant, trend for greater thermal tolerance among mussels transplanted to the warmer, protected site that was evident only at the lowest assay temperature. Seasonal acclimatization (see below) may have obscured evidence for persistent developmental effects on thermal tolerance in our study design; these may have appeared at lower winter assay temperatures (electronic supplementary material, figure S4).

The pattern of thermal tolerance plasticity in juveniles but not adults that we observed in *M. californianus* is far from universal. Thermal tolerance of adults does respond to recent temperatures in numerous species [15,45–48],

although experimental differences necessitate caution in making generalizations [49]. Notably, adult *M. californianus* seasonally adjust membrane composition and temperature of Hsp induction [18,19]. Similarly, ratios of DNA to RNA in reciprocally transplanted adults quickly adjusted to transplant sites [50]. Adults of other *Mytilus* congeners acclimate both absolute heart rate and acute temperature thresholds for cardiac function [51]. However, definitive links between these parameters and thermal tolerance have not always been demonstrated. Further comparative work is needed to identify the environmental factors and time-scales of variation driving divergence among life-history stages in plasticity of thermal tolerance and to identify the underlying mechanisms.

(b) Seasonal acclimatization of thermal tolerance curves

Juveniles assayed in summer exhibited moderate survival at 37.7°C and 38.6°C, whereas no juveniles survived these temperatures in the groups assayed in winter. Seasonal acclimatization of thermal tolerance curves has been observed in a wide variety of taxa [8]. The observed seasonal differences in the frequency of warm, low-tide episodes probably drive seasonal variation in mussels' thermal tolerance.

The seasonal differences in thermal tolerance of *M. californianus* might also be a result of variation in phytoplankton food supply, which influences both growth rates and biochemical status [52,53], or of interactions between food availability and other stressors [54]. Five years of data from the intertidal zone at HMS (2002, 2008–2011) indicate that chlorophyll *a* levels, a proxy for phytoplankton biomass, are significantly lower during the autumn and winter months (HMS Marine Life Observatory, http://mlo.stanford.edu). Accordingly, daily growth coefficients were significantly lower from 22 July–12 December compared to 21 June–22 July, particularly at the exposed site. Seasonal reductions in autumn and winter growth rate because of reduced food have been observed in other marine invertebrates, including mussels [55–57]. Notably, food restriction decreased thermal tolerance of adult *M. californianus*, presumably owing to energetic constraints [58].

(c) Plasticity versus genetic constraint in regulation of growth

Growth rates were similarly affected by an interaction between origin shore height and transplant site. Juveniles originally from the high-shore sites grew 94% (exposed-high) and 70% (protected-high) faster at the exposed transplant site compared to the protected transplant site in summer. Similar, context-dependent origin effects on growth have been observed in *Cerastoderma edule* and *Littorina saxatilis* [59,60]. Filter-feeding invertebrates like *M. californianus* at high-shore sites are submerged for shorter durations than conspecifics at low-shore sites, constraining their feeding opportunities. When food is limited, *Mytilus edulis* adjusts its filtration rate and its ingestion/absorption efficiencies, thereby increasing capacity for growth [61]. Similarly, juvenile *M. californianus* from high-shore sites could be primed to deal with hotter conditions and lower food availability; when these individuals are moved to favourable conditions, they would be able to grow faster than individuals from low-shore sites.
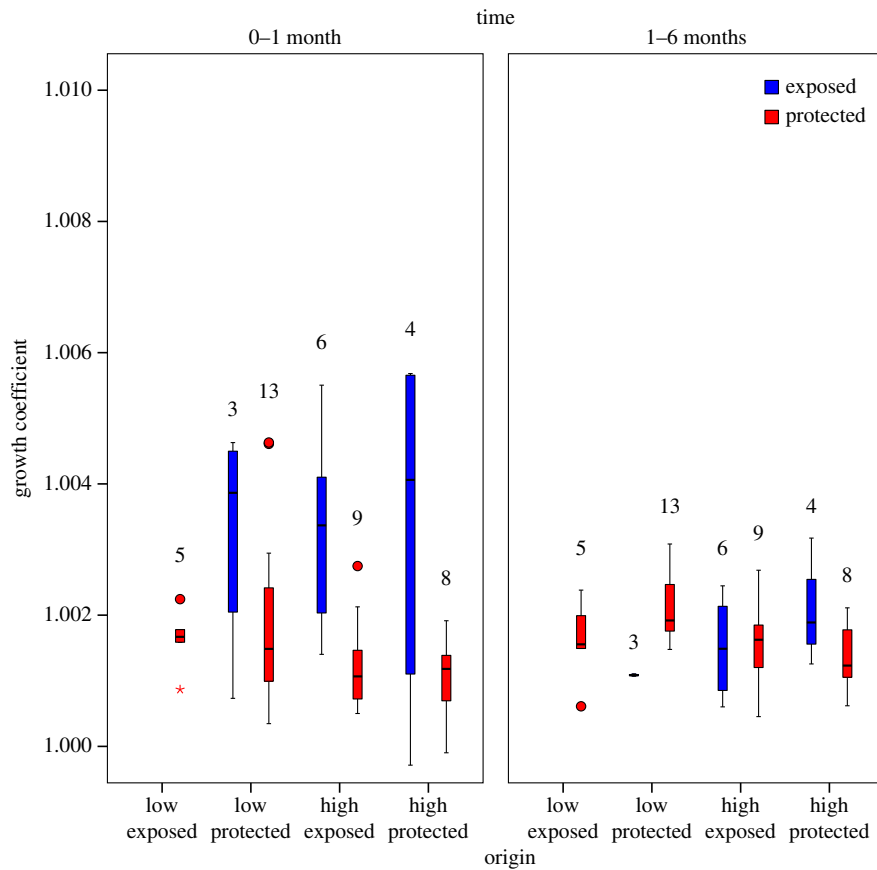
Figure 4. Box plots of daily growth coefficients for the subset of juvenile mussels from the four origin sites that were identifiable after both one and six months in the field. The data were separated between the zero to one month and one to six months intervals. A growth coefficient of 1.0 indicates no growth. Sample sizes are listed above each bar. No individuals transplanted from the exposed-low site to the exposed site were included in this analysis.

(d) Thermal tolerance and growth trade-offs

Juveniles transplanted to the warmer, wave-protected site had the highest thermal tolerance, but they grew more slowly in summer. This finding is consistent with previous results from this species [58], as well as from several fishes [12,62,63]. Broadly, this inverse relationship between thermal tolerance and growth is probably a result of energetic constraints among protected-site transplants. For example, induction of the heat shock response—which protects macromolecules but at an appreciable cost [64]—or of other thermoprotective mechanisms could impinge on growth. It is also possible that differences in food quantity and/or time available for feeding exist between the transplant sites, although there is no reason to expect that food concentrations would vary over the short distance between the exposed and protected sites.

Mussels from all origin sites grew slowly and exhibited low thermal tolerance after being constantly submerged in the common garden, where they did not encounter episodes of high temperature that would prime compensatory tolerance mechanisms. Though this pattern is not universal [12], decreased growth under constant temperatures has also been observed in salmon and other intertidal invertebrates [65,66] and may be attributable to a loss of positive metabolic effects resulting from warm, but not stressful, body temperatures [67]. This pattern may also be attributable to higher cumulative metabolic costs; unlike mussels in the field (electronic supplementary material, figure S3), common-garden juveniles did

not experience low temperatures, which can reduce metabolic rate via Q_{10} effects [68]. Furthermore, common-garden mussels did not benefit from reduced rates of energy expenditure via anaerobiosis during emersion [35].

5. Conclusion

Our results imply environmentally driven developmental plasticity in thermal tolerance that is inversely related to growth rate among recent *M. californianus* recruits. Importantly, the persistent effect of origin site supports the conclusion that both genetics and recent experience contribute to thermal tolerance [4,69,70]. However, there was no evidence for analogous plasticity of thermal tolerance in adults. Without the data for juveniles, the latter result may have been mistaken for fine-scale local adaptation. A more plausible, yet still hypothetical, mechanism for these patterns is irreversible epigenetic programming [71]. The developmental adjustment appears to interact with seasonal acclimatization, evident as horizontal shifts in the thermal tolerance curve. Both irreversible and reversible forms of plasticity should be considered when predicting how species will respond to shifting environments. Under the right circumstances, such plastic responses could mitigate the negative effects of global change.

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