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Abstract

Animal personality is often studied within compressed periods of observation that represent narrow windows in comparison to animal lifespans. While much is known about the relations between repeatable personality traits and cross-situational behavioural plasticity, less is known about how such traits might differ across age classes or life-history transitions. We conducted a cross-sectional study of startle response duration in three size classes of *Pagurus bernhardus*, the common European hermit crab. We defined size classes using transitions in the preferred species of gastropod shells that accompany growth, and this change in preference is in turn associated with a transition from intertidal to subtidal habitats. Compared to small and medium sized intertidal individuals the larger subtidal hermit crabs behaved cautiously by showing startle responses of greater duration following disturbance. Startle responses were also repeatable within all three size classes, confirming the presence of animal personality in intertidal hermit crabs and demonstrating that this pattern is retained within the largest size classes, that have undergone the transition from intertidal to subtidal habitat. Interestingly, there was a trend for the pattern of repeatable startle response durations to increase with size class, with the highest value for repeatability and greatest range of startle response durations being present within the large subtidal population. The greater range of startle responses indicates that the longer startle response durations in some larger individuals are more likely due to developmental changes with age and habitat use than reflecting selection against the boldest individuals during earlier stages of life.
Introduction

Animal personality is the presence of behavioural repeatability (Nakagawa & Schielzeth 2010), i.e. between-individual behavioural differences that are consistent across repeated sampling occasions (Dall et al. 2004, Sih et al. 2004, Dingemanse & Réale 2005, Dingemanse et al. 2010). Thus, animal personality defined in this way is dependent on two sources of behavioural variation, between individual variance (which increases repeatability) and within individual variance (IIV, also described as residual variance, which decreases repeatability). Animal personality is present in a wide range of animals. Initial studies on chordates (e.g. see Gosling 2001) were soon followed by studies of personality in animals across a range of phyla, including molluscs (e.g. Sinn et al. 2008), cnidarians (e.g. Briffa & Greenaway 2011) and arthropods (e.g. Pratt et al. 2005; Johnson & Sih, 2007; Reaney & Backwell, 2007; Briffa et al. 2008; see Kralj-Fišer & Schuett 2014 for a general review of personality in invertebrates). Typically, the longitudinal data needed for animal personality studies are collected within short windows relative to animal lifespans. This approach is sufficient to investigate repeatability itself (although the number of samples per individual and sampling interval can influence the results, see Bell et al. 2009). Such data can also allow investigation of a range of ancillary questions relating to animal personality. These include the interrelations between repeatability and behavioural plasticity (where conditions naturally change in the short term, can be experimentally manipulated, or where individuals can be tracked as they encounter heterogeneous habitats). We might also be interested in the presence of among individual differences in that behavioural plasticity (i.e. differences in behavioural reaction norm or the individual x environment interaction, Dingemanse et al. 2010). Finally, longitudinal data can be used to assess the balance between behavioural variation between and within individuals (Stamps et al. 2012) and the circumstances where this may change (e.g. Briffa et al. 2013, Maskrey et al. 2021). In contrast with data collected
in relatively restricted (relative to lifespans) time windows, less attention has been given to investigating animal personality over the longer term, including over developmental timescales and how it might be affected by key life history transitions. Such studies could contribute to our understanding of why animal personality is present (e.g. Stamps & Groothuis 2010, Wilson & Krause 2012). For instance, mean level behavioural differences among cohorts could occur if some behavioural types have enhanced survivorship over others, which are selected against at earlier developmental stages. This scenario underpins the pace of life syndrome (POLS) hypothesis (see Montiglio et al. 2018 for an overview), which suggests that animal personalities derive from among individual differences in lifetime fitness strategies. Proactive individuals should show high boldness and prioritise rapid growth, early maturation and early reproduction at the cost of greater risk exposure and reduced survival. In contrast, shyer reactive individuals resolve this trade-off through slower growth and delayed reproduction but with enhanced longevity. Alternatively, developmental changes in resource requirements and in prioritising current versus future fitness could also lead to differences in behavioural type among cohorts. In this case, individuals might additionally show different behavioural trajectories during development, as in behavioural developmental reaction norms (i.e. individual x age interaction effects, encompassing individual x environment and genotype x environment effects if different environments are experienced during development), which could then produce different behavioural types within cohorts (see Stamps & Groothius 2010).

Examples of studies on the development of personality include vertebrates (e.g. see Stamps & Groothuis 2010), molluscs (e.g. Sinn et al. 2008, Dahirel et al. 2017) and insects (Amat et al. 2018). In the case of insects, the metamorphoses that mark major life history transitions provide clear opportunities for investigation of potential causes and underlying mechanisms of personality (Wilson & Krause 2012). Studies of insects show that behavioural
types and behavioural syndromes can persist across these major life history transitions (Brodin 2009, Gyuris et al. 2012, Niemelä et al. 2012, Stanley et al. 2017) at least in heterometabolous species (but see Amat et al. 2018 for a discussion of exceptions). At the same time, average trait values can vary across ontogeny, with adults typically being less bold than larvae (Gyuris et al. 2012, Niemelä et al. 2012, Stanley et al. 2017). Nevertheless, boldness can still change within a life history phase, as in the cricket *Gryllus campestris* where boldness increased during observations over the course of adulthood (ca. 45-50 days) (Fisher et al. 2015). In contrast to the situation in insects, little is known about the ontogeny of personality in other arthropods including crustaceans.

As outlined above, when individuals can be followed through ontogeny (e.g. Sinn et al. 2008, Fisher et al. 2015), this approach can potentially yield rich information, such as the presence of developmental plasticity and among individual differences in developmental reaction norms, as well as signs of niche construction and niche picking (Bergmüller & Taborsky 2010). In many cases, however, there are barriers to collecting longitudinal behavioural data over developmental time-scales, particularly in non-captive animals, such as wild crustaceans. For example, individuals sampled from a wild population may not be amenable to marking and recapture. An alternative approach is to conduct cross-sectional investigations comparing behaviour, already known to be repeatable, across individuals of different life-history stages (Dowling & Godin 2002, Brown & Braithwaite 2004; Magnhagen & Borcherding 2008, Wallis et al. 2020). Although such an approach cannot directly address differences in individual developmental trajectories it can show us (a) whether mean level expression of personality traits differ and (b) whether the personality effect size (i.e. repeatability) differs between life stages / cohorts. In human populations for example, behavioural repeatability increases with age because individuals become more predictable (reduced IIV) in their behaviour (Roberts & DelVeccio 2000), whereas in *G. campestris*
repeatability increases with age due to higher among individual variance (Fisher et al. 2015). In contrast, in ectothermic vertebrates there is an overall pattern for greater behavioural repeatability in samples of juveniles compared with samples of adults (Bell et al. 2009). These examples illustrate the importance of understanding the contribution of population demography in studies of repeatable behaviour. First, if samples of individuals encompass a wide range of size / age classes, then developmental changes in behaviour (if present) could contribute to overall repeatability. Second, one could investigate the potential causes of such differences in personality traits between cohorts. In general, though, studies (longitudinal or cross-sectional) investigating across cohort differences in repeatable behaviour are still relatively rare outside of vertebrates (Stamps & Groothuis 2010, Amat et al. 2018).

In hermit crabs, it is not possible to mark and recapture individuals with much success because they are small animals that do not seem to show much fidelity to a particular territory. Intertidal individuals in particular may undergo some spatial redistribution during each tidal cycle. Additionally, they do not possess body parts that can reliably retain marks in the long term, due to periodic moulting of the exoskeleton associated with growth, and marking their gastropod shells is not feasible as they can change shells (see below). Furthermore, it is not possible to age individuals directly because they do not retain hard body parts that can provide an index of age. Nevertheless, adult body size correlates with age (Lancaster 1988) and growth in hermit crabs is associated with stark changes in their natural history. These changes offer an opportunity to study the links between personality and life history transitions additional to those marked by metamorphoses studied in insects. First, there is a change in resource requirements concerning the empty gastropod shells that they occupy and use as portable shelters. As the size limits of a particular shell species are exceeded, there is a change in preference to a larger species of shell. Second, while smaller individuals inhabit the intertidal zone, the largest size class are only found subtidally, a
change which in part is likely to be due to the presence of larger gastropod species and hence their empty shells. In the European hermit crab, *Pagurus bernhardus*, individuals fall into at least three size classes defined by the species of gastropod shell that they preferentially occupy. Small intertidal crabs typically occupy shells of the smooth periwinkle, *Littorina obtusata*, which they will select in preference to other shells of similar size (various species of top shell such as *Steromphala (= Gibbula) cineraria*, for example). *L. obtusata* shells, however, are not large enough for all intertidal hermit crabs and as growth continues their preferences switch to shells of the common periwinkle, *Littorina littorea*. Subtidal *P. bernhardus* individuals are substantially larger than the two size classes already described. They occupy shells of the common whelk, *Buccinum undatum* a much larger species snail at maturity. Thus, in *P. bernhardus*, changing shell requirements provide a context for investigating differences in repeatable behaviour across an important life-history transition.

When threatened, hermit crabs of all sizes show a startle response of rapidly withdrawing into the shell and the duration until re-emergence can be analysed for variation in mean level responses across conditions and for repeatability (Briffa et al. 2008; Briffa & Bibost 2009; Briffa & Twyman 2011; Briffa et al. 2013; Bridger et al. 2015). Within the two intertidal size classes that have been analysed in previous studies, startle response durations are repeatable, but we have yet to compare boldness across size classes. An increasingly common approach to animal personality studies has been to obtain many repeated observations (i.e. n > 2) from each individual such that overall repeatability can be decomposed into the within and between individual variance components mentioned above (e.g. Stamps et al. 2012). In the current study, however, our primary aim is simply to determine whether larger (and hence older) individuals differ in mean boldness compared to smaller individuals. We nevertheless observed each individual twice to provide a basic index of repeatability, in order to establish the extent to which boldness is a personality trait in the
hitherto unanalysed subtidal hermit crabs. We thus assess the possibility that personality in hermit crabs varies across a key life-history transition – the changes in resource requirement that occur with increased size and the transition from life in intertidal to subtidal habitats. Studies of other arthropods have shown that boldness often decreases across life history stages. These changes may reflect either decreased survival of bold individuals in early life, or a developmental reduction in boldness, for example due to a reduced need to perform risky behaviours such as dispersal and resource acquisition. In the case of the former we would expect to see both bold and shy individuals at the early life stages but if shyer individuals only appear at later stages this would more likely represent a developmental shift away from boldness. Additionally, a greater diversity of behavioural types across individuals at later stages would indicate the presence of variation in developmental reaction norms. On the other hand, if boldness increases with age this is more likely to represent a developmental shift away from prioritising future survival over current resource acquisition at early life stages.

Materials and Methods

Small individuals occupying *L. obtusata* shells (size class A: 0.08g to 0.32g, mean = 0.219 ±SE = 0.014g) and intermediate size individuals occupying *L. littorina* shells (size class B: 0.41g to 1.78g, mean = 1.121 ±SE = 0.08g) were collected by hand inter-tidally from Hannafore point, Cornwall, UK between September and November 2009. Large subtidal individuals occupying *B. undatum* shells (size class C: 7.12 to 46.4g, mean = 23.05 ±SE = 2.26g) were obtained from trawls of Plymouth Sound, UK also between September and November 2009. Unsurprisingly, there was clear variation in crab mass across the three size classes as defined by the species of shell occupied (1-way ANOVA: $F_{2,47} = 838.1$, $P < 0.0001$), with *post hoc* Tukey tests indicating significant differences between each pair of
adjacent size classes (A versus B, B versus C; \( P < 0.0001 \) in each case). For each size class, crabs were transported to the lab and housed individually either in 12cm diameter crystalizing dishes (size classes A and B) or in 1L tanks (size class C) containing aerated seawater at 15°C. For all three size classes the individual housing conditions were sufficient for each crab to move around freely without constraint. Each crab was startled using a handling procedure described previously (Briffa et al. 2008), causing the crab to tightly withdraw into its shell. Briefly, crabs were lifted out of the seawater by hand and inverted for 5s, which causes them to withdraw into the shell. They were then replaced on the base of the container, with the aperture facing upwards so that their re-emergence could be clearly observed without the observer needing to overshadow the subject. The duration of withdrawal (henceforth startle response duration) was timed until the crabs re-emerged and made first contact with the container base with the walking legs, to the nearest 0.01s. There was no maximum observation time, i.e. no censoring of the data. Startle response durations were obtained from each crab on two occasions, seven days apart. After behavioural data collection was completed, crabs were removed from their shells by crushing in a vice, the shell fragments were dried and weighed to obtain shell mass (SM) and the crab was weighed and sexed. Data from female crabs were not included in the analyses to avoid sex-specific variation in shell optima associated with reproductive state (Elwood et al. 1979), which can affect startle response durations (Briffa & Bibost 2009). We used the mass of each male crab to estimate its optimal shell mass (OSM) using regression lines from previous shell selection studies (Briffa & Elwood 2005, 2007), and calculated the proportional deviation from this (DOSM) for each crab’s actual shell mass (SM) as follows:

\[
DOSM = \left( \frac{|OSM - SM|}{OSM} \right)
\]
Since deviation from optimal shell mass might affect startle response duration (since it will determine the amount of protection afforded by retreating into the shell), DOSM was included as a covariate in the analysis. However, DOSM did not vary among size classes ($F_{2,47} = 2.1$, $P = 0.14$). We obtained two startle responses from the following number of males in each size class: A, $n = 17$; B, $n = 17$; C, $n = 16$, producing 100 observations across 50 individuals. Sample sizes for classes A and B were chosen to match the number of individuals in size class C that were obtained from the trawl (+1 in each case to bring $N$ up to 50 individuals). At the end of the experiment all male crabs were supplied with a new shell and returned to the sea.

In order to determine the effect of size class on startle response duration, we used a linear mixed effects model, estimated by restricted maximum likelihood (REML). The fixed predictors were size class (A, B or C), occasion (startle response 1 or 2) and proportional deviation from mass-specific optimal shell mass. We allowed individual specific random intercepts but did not attempt to fit random slopes across occasions as there were only two occasions. Initial analyses indicated that there was no interaction between size class and occasion so these were omitted from the final model (model 1). We also found that $\log_{10}$ transforming the startle response data improved the model fit, moving the normality of residuals and the homogeneity of variance in residuals across individuals closer to the underlying model assumptions (see Schielzeth et al. 2020). We confirmed that the single random effect in the model was justified by re-fitting the model using maximum likelihood estimation (ML) and comparing this refitted model to a linear model that contained only fixed effects. Significance testing for fixed effects was then assessed via $F$-ratios, using Satterthwaite's method to calculate the degrees of freedom. We further investigated the among-individual differences accounted for in the random intercept by calculating the repeatability of startle responses, for all data combined and for each size class separately.
Analyses were performed in R 3.6.1 (R Core Team 2019) using the packages lme4 (Bates et al. 2015), lmerTest (Kuznetsova et al. 2017) and rptR (Stoffel et al. 2017). Model code is provided in Appendix 1.

Results

Startle responses were unaffected by deviation from optimal shell size ($F_{1,46} = 0.01, P = 0.9$) but were greater on occasion 2 compared with occasion 1 ($F_{1,49} = 12.5, P < 0.0001$) and in large crabs in size class C compared with smaller crabs in size classes A and B ($F_{2,46} = 4.9, P = 0.012$) (Figure 1A, B, C; See table 1 for the model parameters). Post-hoc tests show that duration was not different between size classes A and B ($P = 0.93$) but for crabs in class C the duration was greater than for crabs in classes B ($P = 0.03$) and A ($P = 0.015$). While dividing crab masses into discrete size classes is an intuitive approach given the natural history of hermit crabs an alternative way of analysing the data would be to treat crab mass as a continuous predictor (Figure 1D). This approach yielded similar results, reported in Appendix 2 (model 2). It was evident (see Figures 1 and 2) that eight outlying data points were present in the data. To check that the patterns reported above were not driven by these observations, both models (1 and 2) were re-run with these data omitted, which returned qualitatively identical results (see Appendix 2). While it is clear that startle response duration varies with crab mass it is unclear whether this represents a discontinuous pattern of variation between size classes (i.e. Figure 1, A – C) or a pattern of continuous variation over the range of crab masses (Figure 1 D). In the latter case we would expect to see a significant positive slopes within each size class so to test this possibility model 2 was re-run separately for each size class. There was no correlation between mass and startle response duration for size classes A and C (A; $\beta = -0.12, F_{1,14} = 0.0008, P = 0.92$, C; $\beta = -0.01, F_{1,13} = 0.54, P = 0.47$) but within size class B a positive correlation was present ($\beta = 0.74, F_{1,13} = 5.87, P = 0.03$; Figure 2).
The random intercepts model where size class was used as a categorical predictor (model 1) provided a better fit with the data than an equivalent general linear model that lacked random intercepts (see Appendix 1.2), indicating significant among individual differences in startle response duration ($\chi^2_{1} = 28.24, P < 0.0001$; Figure 1A, B, C). To quantify these differences we calculated the repeatability of startle responses across the two observations, adjusted on proportional deviation from optimal shell size. For all size classes combined startle responses were highly repeatable ($R = 0.61, 95\% \text{ CIs} = [0.42, 0.77], P < 0.0001$). When repeatability was estimated specifically for each size class, there was a trend for much greater repeatability in the largest size class C compared with A and B on the basis of non-overlapping 80\% confidence intervals. However there was no significant repeatability difference on the basis of 95\% confidence intervals (A: $R = 0.40, 95\% \text{ CIs} = [0.01, 0.725], 80\% \text{ CIs} = [0.088, 0.625]$; B: $R = 0.41, 95\% \text{ CIs} = [0.01, 0.73], 80\% \text{ CIs} = [0.142, 0.676]$; C: $R = 0.88, 95\% \text{ CIs} = [0.70, 0.96], 80\% \text{ CIs} = [0.788, 0.942]; P < 0.0001$ in each case. Note that for size classes A and B, the lower 95\% CIs are constrained not to cross zero).

**Table 1**: Variance and parameter estimates for the mixed effects model of startle response durations.

| Random intercept     | Variance | SD    |
|----------------------|----------|-------|
| ID                   | 0.10974  | 0.3313|
| Residual             | 0.05297  | 0.2302|
| **Fixed effects**    |          |       |
| (Intercept)          | 0.84295  | 0.11381|
| Size class B         | 0.04503  | 0.12662|
| Size class C         | 0.38646  | 0.13313|
| Occasion 2           | 0.16303  | 0.04603|
| DOSM                 | 0.02646  | 0.28526|
Discussion

As in previous studies of hermit crabs, consistent between-individual variation in startle response duration was present. Here we found that as well as being present in the previously studied intertidal samples of hermit crabs, animal personality was also present in the sample of the larger-sized individuals of the subtidal population. Indeed the greatest repeatability value was found for those larger crabs in comparison with the smaller size classes. In addition to significant repeatability within each size class, we also found mean level differences in startle response duration between size classes. Size specific mean level boldness is seen in a range of study species, but the direction of the effect differs between examples. In several studies on insects (Gyuris et al. 2012, Niemelä et al. 2012, Stanley et al. 2017) and in gastropods (Dahirel et al. 2017) younger individuals were bolder than older ones, similar to the current results, but results in other taxa are more mixed. In killifish, Fundulus diaphanous, small individuals show the longest startle responses, possibly behaving cautiously because they are easy for predators to handle and therefore subject to high predation risk (Dowling & Godin 2002). In other examples, such as the poeciliid fish Brachyraphis episcosi (Brown & Braithwaite 2004) and perch Perca fluviatilis (Magnhagen & Borcherding 2008) it was the larger individuals that behaved more cautiously. In these vertebrate examples, boldness varies continuously with body size but the pattern of variation in the hermit crabs studied here was different. Only intermediate sized crabs in L. littorea shells showed continuous variation in startle response with mass but there were also clear differences across size categories, with the small and intermediate sized intertidal individuals being bolder on average (shorter startle response durations) than the large subtidal individuals in B. undataum shells.

For all three size-classes, startle responses increased, on average, between occasions one and two. This may represent a mean level pattern of sensitisation to the startle procedure.
In previous studies we found an approximately equal number of individuals that increased and decreased their startle response duration over repeated observations (see Stamps et al. 2012) but the current sample contained more individuals that increased the duration (see Figure 1). The differences in startle response between size classes were unlikely to have been influenced by occasion, however, or by physical constraints resulting from different shell morphologies. Responses were timed up to the point where the crab first contacted the substrate with its walking legs in order to make a righting attempt, rather than until the crab successfully righted its position. A previous study in which shell size was manipulated to be much too small (Briffa & Bibost 2009) demonstrated that the size of the gastropod shell relative to the size of the crab can influence startle response durations. In contrast, adequacy was far less variable in the present study based on naturally occupied shells, and there was no difference in POSM between size classes and no effect of POSM on startle response durations. There are two general explanations remaining for the longer startle responses in larger crabs. First, the pace of life syndrome hypothesis (POLS) implies that declining boldness with age would arise because bolder individuals with short startle responses may have lower early survival such that fewer of them attain large size. Second, there might be a developmental shift in boldness, with crabs becoming less bold with age. While it is not possible to directly distinguish between contributions from these two processes using the current cross-sectional data, the following can be noted: Short startle responses (i.e. high boldness) are still present in size class C, even though the mean duration is high. Furthermore, all of the startle responses in size class A are at the lower end of range seen across the whole dataset, whereas the longest startle responses are present in size class C. These observations seem less compatible with the first explanation, leaving the possibility of developmental change in startle response duration open for further investigation.
There are a number of reasons why, for hermit crabs, behaving more cautiously as size /age increases, could be beneficial. First, the shift in boldness could be due to differences between the intertidal and subtidal habitats. In the intertidal, hermit crabs frequently utilise cover from seaweed at the fringes of tide pools, in crevices and underneath loose stones. Sandy bottomed subtidal areas often lack such cover and general rugosity, perhaps elevating the level of predation risk (e.g. from predatory crustaceans or fishes) that hermit crabs are exposed to. Second, throughout growth hermit crabs must obtain new gastropod shells of increasing size, to maintain adequate protection. Those hermit crabs that have reached the largest size class however are less likely to require a change of shell, and larger shells are in any case less available as they already occupy shells at the upper range of available sizes. Thus, they will not need to allocate as much time to prospecting for new shells compared with smaller individuals. Given that boldness as measured with startle responses correlates with shell investigation behaviour (Mowles et al. 2012) it is likely that the chance of obtaining new shells correlates with boldness. Hence, the benefits of short startle responses would decline with crab size and age, such that an age related decline in boldness would be adaptive. Although the shift in boldness would be associated with access to a resource this possibility seems analogous to other situations where the requirement to perform risky behaviours such as dispersal declines with age.

The mechanisms underlying developmental personality change also warrant further investigation. One possibility is that developmental change occurs as a result of social learning from observing the behaviour of others. Social learning has not been directly demonstrated in hermit crabs but several studies have shown that isolation leads to changes in social behaviour in other crustaceans (e.g. Hemsworth et al. 2007, Duffield et al. 2015). If present, social learning could produce an increase in mean startle responses, if most individuals learned to act cautiously. In the current data we found (albeit tentatively) greater
repeatability within the largest size class. Although it is not possible to formally decompose variance components in the current data, the greater spread of startle responses in group C suggests that greater among individual variance might contribute to increased repeatability in older individuals, similar to the case of *G. campestris* crickets, although boldness increased rather than decreased across adulthood in that example (Fisher *et al.* 2015). This wider range of startle response durations compared to the smaller intertidal crabs, might reflect differences in experience and the opportunity to learn among individuals (e.g. see Frost *et al.* 2007). Alternatively, development of differences in behaviour might be driven by different experiences of social conflict (Bergmüller & Taborsky 2010). In animals that frequently engage in aggression (e.g. over gastropod shells in the case of hermit crabs), winner and loser effects could result in a form of niche specialization associated with distinct behavioural strategies. Indeed, startle responses in *P. bernhardus* can increase after fighting in individuals that have defended their gastropod shell from an attacker (Courtene Jones & Briffa 2014). Boldness and variation in boldness could also change during development independently of social experiences, as a result of other experiential differences and differences in developmental reaction norms (e.g. Stamps & Groothius 2010; Curley *et al.* 2013).

The pattern of increasing caution with size indicates the possibility of developmental changes in boldness in hermit crabs. The primary cause of this mean shift in boldness does not appear to be reduced survivorship in less bold individuals. The decline in boldness indicates that being more risk averse with increasing size class and age is beneficial, either because of higher risk levels in the subtidal environment or because the benefits of behaving in a risk prone way reduce with age. The proximate causes of this change could involve processes such as social learning and social conflict but these questions still need to be resolved. Lifetime studies on dumpling squid, *Euprymna tasmanica*, show that size specific boldness can be plastic and vary with habitat as well as developmental stage (Sinn *et al.*
Lifetime studies on hermit crabs would most likely need to be conducted in the laboratory due to the difficulties of long-term marking and recapturing crustaceans. Here, however, we show that the expression of a personality trait in a natural population of arthropods varies across size classes and appears to be subject to developmental change associated with a key life-history transition between habitats and associated resource requirements.

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Figure 1: Startle responses in occasion one and two for different sized hermit crabs. Box and whisker plots for size classes A ($n = 17$), B ($n = 17$) and C ($n = 16$), based on species of occupied gastropod shell (Littorina obtusata, L. littorea and Buccinum undatum respectively). Within each plot startle responses durations are indicated by black dots and the two responses from each individual are linked by black lines. Larger black dots denote outlying data points that were excluded from the supplementary re-analysis (see Appendix 2 for details). Part D shows the same startle responses plotted against crab mass ($N = 50$) with a fitted regression line (shaded area shows the standard error of the regression).
Figure 2: Intra-size class correlations between crab mass and startle response durations, for size classes A ($n = 17$), B ($n = 17$) and C ($n = 16$). Regression lines fitted for illustration (shaded area shows the standard error of the regression).
Appendix 1: R code

Variable codes

ID = hermit crab ID
Class = size class
Mass = crab mass
Occ = occasion
DOSM = deviation from optimal shell mass
Dur = startle response duration (s)

Packages

lme4, lmerTest, emmeans

1.1 R code for LMM using size class as a categorical predictor

model.1 <- lmer(log10(1+Dur) ~ Class + Occ + DOSM + (1|ID))

summary(model.1) #parameters
anova(model.1) #stats tests
emmeans(model.1, list(pairwise ~ Class), adjust = "tukey") #posthocs

1.2 R code for LMM using a crab mass as a continuous predictor

model.2 <- lmer(log10(1+Dur) ~ Mass + Occ + DOSM + (1|ID))

Further versions of this model were used to obtain estimates for the correlation between crab mass and duration within each specific size class.

A.data <- data[which(Class=='A'), ]
B.data <- data[which(Class=='B'), ]
C.data <- data[which(Class=='C'), ]
m2A<-lmer(log10(1+Dur) ~ Mass + Occ + DOSM + (1|ID), data= A.data)

m2B<-lmer(log10(1+Dur) ~ Mass + Occ + DOSM + (1|ID), data= B.data)

m2C<-lmer(log10(1+Dur) ~ Mass + Occ + DOSM + (1|ID), data= C.data)

1.2 R code for testing the random effects part of model 1

model.1.ML <- lmer(log10(1+Dur) ~ Class + Occ + DOSM + (1|ID), REML = F)

model.1.FEO <- lm(log10(1+Dur) ~ Class + Occ + DOSM)

anova(model1.ML, model1.FEO)

1.3 R code for estimating repeatability

A.data <- data [which(Class=='A'), ]

B.data <- data [which(Class=='B'), ]

C.data <- data [which(Class=='C'), ]

#All data

rep.All<-rpt(log10(1+Dur) ~ DOSM + (1 | ID), grname = "ID", data = data, datatype = "Gaussian", nboot = 1000, npermut = 0)

#A

rep.A <-rpt(log10(1+Dur) ~ DOSM + (1 | ID), grname = "ID", data = A.data, datatype = "Gaussian", nboot = 1000, npermut = 0)

#B
rep.B <- rpt(log10(1+Dur) ~ DOSM + (1 | ID), grname = "ID", data = B.data, datatype = "Gaussian", nboot = 1000, npermut = 0)

#C

rep.C <- rpt(log10(1+Dur) ~ DOSM + (1 | ID), grname = "ID", data = C.data, datatype = "Gaussian", nboot = 1000, npermut = 0)

#To change the CIs from the 95% default to 80%, add CI = 0.8 to the formula

Appendix 2: Results of supplementary analyses

Table A2.1 Model using mass as a continuous predictor. The second and third columns show variance and parameter estimates, the remaining columns report the results of significance testing.

| Random intercept | Variance | SD  | \(\chi^2\) | df | \(P\)     |
|------------------|----------|-----|------------|----|----------|
| ID               | 0.11812  | 0.3437 | 30.95     | 1  | <0.0001  |
| Residual         | 0.05297  | 0.2302 |

| Fixed effects    | Estimate | SE  | \(F\)     | df | \(P\)    |
|------------------|----------|-----|----------|----|---------|
| (Intercept)      | 0.85934  | 0.09944 |
| Mass             | 0.01041  | 0.00440 | 5.60   | 1.47| 0.022   |
| Occasion         | 12.54    | 1.49 | 0.0009   |
| Occasion 2       | 0.16303  | 0.04603 |
| DOSM             | 0.15442  | 0.28674 | 0.29   | 1.47| 0.60    |
Table A2.2 Model using size class as a categorical predictor omitting outliers. The second and third columns show variance and parameter estimates, the remaining columns report the results of significance testing.

| Random intercept | Variance | SD   | $\chi^2$ | df | $P$   |
|------------------|----------|------|----------|----|-------|
| ID               | 0.003233 | 0.05686 | 14.8     | 1  | <0.0002 |
| Residual         | 0.002570 | 0.05070 |          |    |       |

| Fixed effects    | Estimate | SE   | $F$      | df | $P$   |
|------------------|----------|------|----------|----|-------|
| (Intercept)      | 0.238685 | 0.021360 |          |    |       |
| Size class       |          |       | 3.8484   | 2, 43.5 | 0.03 |
| Size class B     | 0.003886 | 0.023609 |          |    |       |
| Size class C     | 0.064200 | 0.025659 |          |    |       |
| Occasion         |          |       | 17.3     | 1, 44.9 | <0.0002 |
| Occasion 2       | 0.044161 | 0.010617 |          |    |       |
| DOSM             | 0.051625 | 0.054473 | 0.90     | 1, 43.1 | 0.35 |
Table A2.3 Model using mass as a continuous predictor omitting outliers. The second and third columns show variance and parameter estimates, the remaining columns report the results of significance testing

| Random intercept | Variance       | SD    | $\chi^2$ | df   | P     |
|------------------|----------------|-------|----------|------|-------|
| ID               | 0.003214       | 0.05670 | 1        | 14.9 | <0.0002 |
| Residual         | 0.002581       | 0.05081 |          |      |       |

| Fixed effects    | Estimate       | SE    | F        | df   | P     |
|------------------|----------------|-------|----------|------|-------|
| (Intercept)      | 0.236          | 0.018 |          |      |       |
| Mass             | 0.002          | 0.0007 | 6.65    | 1, 43.5 | 0.013 |
| Occasion         |                |       | 17.1     | 1,44.6 | <0.0002 |
| Occasion 2       | 0.044          | 0.011 |          |      |       |
| DOSM             | 0.074          | 0.052 | 1,43.8   | 2.02 | 0.16  |