Among-individual behavioral responses to predation risk are invariant within two species of freshwater snails

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Abstract

Prey behavioral response to predation risk drives a range of ecological and evolutionary processes. Key to these effects is the degree to which conspecifics exhibit consistent individual differences in their response to risk or instead follow a mean population-level pattern. Here, we employed the behavioral reaction norm framework to quantify among-individual variation in average predator avoidance behavior (i.e., behavioral types) and the behavioral response to risk (i.e., individual plasticity) in two snail species (*Helisoma trivolvis* and *Physa acuta*) that differ in their vulnerability to predators. While both snail species exhibited substantial variation in behavioral types, individual plasticity in response to risk was remarkably invariant — both snail species increased avoidance behavior with increasing risk, but all conspecific individuals followed the population-level pattern (i.e., parallel reaction norms). Instead, individual snails differed in how they adjusted their behavior over the course of repeated behavioral assays (n = 12 per individual), with some exhibiting increased sensitization to risk cues and others habituation. We further show that among-individual behavioral variation, both in behavioral types and in individual responses to repeated assays, was sometimes correlated with physiological traits, providing potential mechanisms for the maintenance of this variation. In total, our results indicate that behavioral types and individual plasticity vary at different hierarchical scales (individual- vs. population-level, respectively) in freshwater snails, which has implications for species interactions and the evolution of predator avoidance behavior.

Key words: Animal personality, behavioral reaction norm, individual plasticity, predator-prey, state-dependence

INTRODUCTION

Prey modify their behavior in response to the presence of predators (Lima and Dill, 1990), with profound consequences for trophic dynamics (Schmitz et al., 2004), space use (Palmer et al., 2022) and species coexistence (Werner and Anholt, 1993). While early studies documented the population-level occurrence and costs of predator avoidance, focus has since shifted to among-individual differences in risk-taking, termed boldness (Beckmann and Biro, 2013; Brown and Braithwaite, 2004). At the individual level, predator avoidance can be separated into: (1) behavioral types, which describe the average level of risk-taking expressed over time or across risk levels; and (2) behavioral plasticity, describing behavioral change in response to increasing risk (Dingemanse et al., 2010). While the ecological importance of mean boldness differences among individuals has been established (e.g., (Griffen et al., 2012; Keiser et al., 2018; Moiron et al., 2020; Toscano et al., 2020)), the relevance of individual behavioral plasticity for predator-prey interactions remains understudied (but see (Dosmann and Mateo, 2014; Kim, 2016)). Here, we take an important first step by testing for consistent individual differences in average behavior and in behavioral plasticity within two prey species that differ in their vulnerability to predation.

The concept of behavioral reaction norms (BRN) provides a framework for parsing these complementary components of individual behavior, behavioral types and behavioral plasticity. Within a predator-prey context, the BRN framework can be experimentally applied by repeatedly assaying prey individuals' behavior across a risk gradient. Hierarchical modeling is then used to estimate two parameters that define an individual's reaction norm: an intercept, representing the stable component of the behavior (behavioral type), and a slope, representing the labile component of the behavior (behavioral plasticity) (Dingemanse et al., 2010). The repeated measures required for the BRN approach are crucial to proper measurement of the among-individual behavioral variation (Brommer, 2013; Dingemanse and Wright, 2020; Niemelä and Dingemanse, 2018a), though testing for individual differences in behavioral plasticity is particularly data-hungry (Martin et al., 2011). As a result, appropriate tests of persistent differences in individual plasticity are still relatively uncommon, and a failure to detect such differences may result from insufficient statistical power (e.g., from testing too few individuals: Martin et al., 2011).

We expect patterns of among-individual variation in behavioral types and behavioral plasticity to diverge between prey species that differ in their vulnerability to predation, particularly considering that behavioral traits are often heritable (Dochtermann et al., 2015). For example, in structurally well-defended prey species, selection on predator avoidance behavior should be weak, allowing for considerable among-individual variation in both behavioral types and plasticity. In the extreme case of near invulnerability, prey individuals should not show plasticity at all due to costs associated with predator avoidance (e.g., reduced feeding time; (Werner and Anholt, 1993)). In contrast, in prey species or populations that are highly vulnerable to predators throughout their ontogeny (e.g., due to weak structural defenses), individual avoidance responses might be canalized (i.e., reduced variation in behavioral types and behavioral plasticity) as an adaptive response to previous stabilizing selection (Charmantier et al., 2008; Kim, 2016; Reed et al., 2006). Here, individuals with weak behavioral responses should be removed by predators (Toscano, 2017). Last, effects of predator selection on among-individual variation in prey behavior might depend on the temporal dynamics of predation. If predation pressure varies over time, different behavioral strategies that are advantageous in low risk versus high-risk situations can be maintained via fluctuating selection (Le Cœur et al., 2015). While tests of such predictions are rare, comparative studies, including cross-species comparisons (Carter and Feeney, 2012; White et al., 2020) are necessary to understand the forces that maintain behavioral variation within populations and how predators shape trait variation within prey communities.

If among-individual variation in behavioral types and behavioral plasticity is present, these aspects of the behavioral phenotype could be associated with and maintained by intrinsic physiological or performance traits (i.e., state-dependence: (Niemelä and Dingemanse, 2018b; Sih et al., 2015)). For example, the "asset-protection principle" posits that prey individuals in good body condition take fewer risks to protect their prospects for future reproductive success (Clark, 1994), driving a positive relationship between body condition and predator avoidance. Alternatively, individuals in good condition are predicted to behave more boldly if good condition affords enhanced escape behavior (Luttbeg and Sih, 2010). Both mechanisms can involve positive feedbacks in which differences in physiological state reinforce behavioral differences among individuals (Sih et al., 2015).

We evaluated these predictions regarding among-individual variation in predator avoidance behavior and the drivers of such variation using two freshwater snail species (*Helisoma trivolvis* and *Physa acuta)* that frequently coexist but differ in their susceptibility to shared predators (e.g., crayfish). *Helisoma* is relatively large and reaches a size refuge from predation, while *Physa* is smaller with a thinner shell that renders it vulnerable to predation throughout its ontogeny (Alexander and Covich, 1991; Chase, 2003; Turner and Chislock, 2007). To quantify among-individual differences in behavioral types and plasticity, we applied the same standardized boldness assay to both snail species across a common gradient of predation risk including chemical cues from both crushed conspecific snails and crayfish (*Faxonius limosus*) fed with snails. We further tested whether aspects of individuals' internal state, including body size, body condition and growth rate were related to aspects of individual behavior (i.e., state-dependence). Last, because the repeated assays on individuals that define the BRN approach could independently alter predator avoidance behavior through conditioning (e.g., sensitization or habituation), we tested for individual-level plasticity in response to the number of behavioral assays performed.

METHODS

Study system

Our study applied the BRN framework to explore individual-level responses to risk using two freshwater snail species, *Helisoma trivolvis* and *Physa acuta*. Freshwater snails have served as a model for predator-induced phenotypic plasticity (DeWitt, 1998), where waterborne chemical cues from injured conspecifics and predators (e.g., crayfish) elicit a range of adaptive phenotypic responses, including life-history, morphological and behavioral changes consistent with predation risk (Crowl and Covich, 1990; Dalesman and Rundle, 2010a, 2010b; Hoverman et al., 2005; Lakowitz et al., 2008; Rundle and Brönmark, 2001; Turner, 1996). Further evidence exists for snail behavioral adjustment to differences in risk type and magnitude (Hoverman et al., 2005; McCarthy and Fisher, 2000; Turner, 2008). However, these previous studies have explored the freshwater snail behavioral response to risk at the population level, for example, measuring the proportion of individuals in the population that exhibit avoidance behavior.

Animal collection and housing

Helisoma trials were conducted in July 2019 while *Physa* trials were conducted in July 2021. *Helisoma* (n = 90; mean initial length ± SD: 12.0 ± 0.8 mm) were collected from the South Brook Park River (Hartford, CT, USA) while *Physa* (n = 99; mean initial length ± SD: 7.1 ± 0.7 mm) were collected from Cedar Hill Cemetery Pond (Hartford, CT, USA). Given that species-specific trials were conducted in different summers and the two snail species were collected from different water bodies, we emphasize that the difference in predation vulnerability is only one factor that might influence behavioral differences between snail species. Size differences between the two snail species used in our study could also contribute to behavioral differences.

Accordingly, the comparative nature of our study should be considered in this context. The experimental procedures described below were applied to both snail species.

Snails were housed individually both before the start of the experiment and in between behavioral assays within small cylindrical containers (8 cm diameter × 8 cm height) filled with 200 mL of artificial pondwater (Klüttgen et al., 1994). Once per week throughout the duration of the experiment, we replaced the water in each container and fed snails with 1 g of dead, decaying leaves gathered from where snails were collected. This feeding regime amounted to *ad libitum* food conditions, similar to what snails would experience in the field. Containers were labeled with ID numbers to keep track of individual snails throughout the experiment.

Experimental design

To measure BRNs, we assayed individual snail behavior across a 4-level predation risk gradient. The behavior of each snail was measured three times at each risk level, yielding 12 behavioral assays per individual. Experiments lasted 3 weeks. Assays were conducted Tuesday-Friday each week (4-day periods), with one assay per snail per day to minimize conditioning. The order of predation risk levels that each snail received was completely randomized across all 3 weeks using a random number generator. Thus, in any one week, individual snails were not necessarily assayed in all 4 risk treatments. In total, our experimental design yielded 2268 independent behavioral measurements across both snail species. This experimental design, and particularly the number of risk levels and replication at each risk level, was informed by power analysis (Allegue et al., 2017) based on previous behavioral assays of *Helisoma*.

Predation risk gradient

We manipulated predation risk using water-borne chemical cues derived from a mix of freshly crushed conspecific snails and crayfish (*Faxonius limosus*) that had been feeding on crushed conspecific snails. Research shows that freshwater snails exhibit avoidance behavior in response to cues from both crushed conspecifics and crayfish themselves (Alexander and Covich, 1991; Dalesman and Rundle, 2010a, 2010b; Hoverman et al., 2005; Lakowitz et al., 2008; McCarthy and Fisher, 2000; Rundle and Brönmark, 2001; Turner, 1996), and our reason for combining these cues was to maximize the chance of seeing a behavioral response in both snail species. In particular, while response to crayfish cues could be dependent on differences in crayfish densities between the habitats where the two snail species snails were collected, crushed conspecifics provide a cue that is universally relevant. Chemical cues were prepared by completely crushing 150 mg of snails and mixing them with 710 mL of artificial pondwater from an aerated tank (30 cm length × 15 cm width × 20 cm height) that housed a single, large adult (9 cm) crayfish. This crayfish was fed *ad libitum* with crushed conspecific snails daily. To obtain different risk levels, we manipulated the concentration of chemical cues within the containers where behavioral assays were performed. The total volume within assay containers was held constant at 20 mL, which included a mixture of artificial pond water and varying volumes of chemical cues based on the level of risk: control - 0 mL, low risk - 4 mL, medium risk - 8 mL, high risk - 12 mL. Thus, our chemical cue concentration ranged from 0-60%.

Predator avoidance assay

For *Helisoma* and *Physa*, behavioral responses to chemical risk cues include hiding within a refuge that is the snail's shell as well as climbing or use of surface water (Goodchild et al., 2020;

Hoverman et al., 2005; McCarthy and Fisher, 2000; Turner, 2008). We focused on hiding behavior, and specifically the time to emerge from the shell and displace (full description below). Here, individual snails that emerge more quickly presumably accept a higher degree of risk (i.e., are bolder) (Ahlgren et al., 2015; Goodchild et al., 2020).

We conducted individual behavioral assays within clear plastic cups (4 cm diameter × 3 cm height) placed over a square grid. Prior to the assay, snails were picked up by hand, inverted (foot facing upwards) and tapped on the sides of the shell with tweezers, causing the snail to retreat into its shell. Once fully retreated (foot and antennae no longer visible outside of the shell), the snail was placed foot-side down in the assay cup in the center of a 2 × 2 cm grid cell. We started a timer upon snail placement and recorded the time it took for the snail to emerge, begin moving and eventually leave the 2 × 2 cm grid cell. These three time measurements were highly correlated (> 0.8), and we used latency to displace as our measurement of predator avoidance behavior. Note that in all four risk level treatments, snails were handled and tapped with tweezers to simulate a predation attempt. Thus, even the control treatment featured some degree of risk.

State measurements

In addition to behavioral data, we measured variables that characterize individuals' ontogenetic and physiological states, including body size, body condition and growth rate. We collected these data to test for relationships between state variables and aspects of among-individual variation in predator avoidance behavior (i.e., state-dependence). We measured snail total shell length (SL) both before the start (i.e., initial SL) and upon completion of behavioral trials (3 weeks later; i.e., final SL), and snail wet mass upon completion of behavioral trials. Snail wet mass was measured by gently drying snails with a paper towel and weighing to the nearest 0.01 mg.

Final snail SL was used as the measure of body size in our analysis, while body condition was defined as the residuals from a linear regression between the log-transformed wet mass and the log-transformed final SL (Jakob et al., 1996). Growth rate was calculated as (final SL - initial SL) / initial SL. Thus, body condition was based on wet mass with the effect of shell length removed, while growth was determined based on change in shell length.

Analysis

We used a multivariate modeling framework to test our predictions. For each snail species, we separately modeled: (1) the displacement latency time (i.e., predator avoidance behavior), (2) body size, (3) body condition, and (4) growth rate. The displacement model included the predation risk level and the number of the trials (i.e., repeated assays) as fixed effects. Individuals were further included as random effects in both the intercept and the slopes of displacement time in response to both fixed effects. We tested for a nonlinear relationship between displacement time and predation risk level by adding a squared term to the fixed effect part of the model. We log-transformed displacement time and excluded displacement times longer than 5 min from the analysis. These events were rare – 0.005% of observations for *Helisoma* and 0.02% for *Physa*.

Body size, body condition and growth rate models included these response variables and the intercept as fixed effects. We further added individuals as random effects in the intercept of these models, and explicitly set the residual variance to null. In these models, individuals had a single measurement and thus the residual variance was redundant to the among-individual variance at the intercept. This approach allowed us to compute correlations between among-individual variances across all four models (i.e., test for state-dependence).

We used a gaussian error distribution for all models. All variables were considered as continuous and were normalized (i.e., mean-centered and unit variance). We ran our analysis in R v4.1.2 (R Core Team, 2021) and fitted our models within a Bayesian framework using the R package brms (Bürkner, 2017). All model specifications, such as priors and warmup and sampling iterations, are available in supplementary material 1.

We conducted a model selection procedure on the displacement time model before fitting the multivariate model. We first selected the random effect structure followed by the fixed effect structure (see Tables S1, S2 in the supplementary material 2 for all model candidates). Models were compared using the expected log pointwise predictive density for a new dataset computed from an approximate leave-one-out cross-validation for Bayesian models using Pareto smoothed importance sampling (Vehtari et al., 2021, 2017), and using the model weight computed from a pseudo-Bayesian model averaging that is stabilized with Bayesian bootstrap (Yao et al., 2018).

We interpreted effect significance based on the credible intervals of posterior probability distributions for each parameter, where effects with 95% credible intervals that did not overlap with zero were considered significant. This approach provides an alternative to p-values, which are increasingly discouraged because they are potentially misleading and do not provide information on effect size or direction (Halsey, 2019). For the displacement time model, we also computed the marginalized coefficient of determination (R^2_{mar}; Nakagawa and Schielzeth, 2013), which is the proportion of the variance explained by the fixed effects, and the marginalized repeatability (R_{mar}; Schielzeth and Nakagawa, 2022), which is the proportion of the variance explained by the among-individual variation averaged across covariate gradients.

RESULTS

Behavioral reaction norms: Response to chemical risk cues

At the mean population level, both snail species responded to increasing concentrations of chemical risk cues by taking more time to displace from their shell refuge (*Helisoma*: mean effect [95% credible interval] 0.108 [0.063, 0.152], Table 1; *Physa*: 0.058 [0.003, 0.115], Table 2; Fig. 1). Given the positive effect of risk cue concentration on the time to displace from the shell refuge, we consider individual snails that took longer to displace as relatively shy hereafter.

We additionally detected persistent among-individual differences in mean predator avoidance within both snail species (i.e., behavioral types; intercepts in Fig. 1). While we could not statistically compare the degree of behavioral type variation between species, *Helisoma* individuals were widely dispersed in their average level of predator avoidance behavior (standard deviation of intercept [95% credible interval] 0.583 [0.492, 0.689], Table 1), while individual intercepts were more similar within *Physa* (0.433 [0.343, 0.533], Table 2). Further, the overall mean among-individual variation (marginalized repeatability) was larger in *Helisoma* (mean R_{mar} [95% credible interval] 41.3% [34.1%, 48.8%], Table 1) than in *Physa* (24.9% [18.1%, 32.4%], Table 2).

Despite the presence of mean population-level plasticity, neither snail species exhibited significant among-individual differences in the behavioral response to risk cue concentration (parallel slopes in Fig. 1). For both snail species, models containing random slopes in response to risk cue concentration performed considerably worse (*Helisoma*: Δ elpd = -0.707, weight = 0.359, *Physa*: Δ elpd = -1.46, weight = 0.205) than models without such random slopes (*Helisoma*: weight = 0.641, *Physa*: weight = 0.788; Tables S1, S2 in the supplementary material 2). Our model comparison further supported linear slopes in response to risk cue concentration for both snail species (supplementary material 2).

Behavioral reaction norms: Response to repeated assays

We further detected behavioral change in response to the 12 assays that each snail was subjected to within the BRN framework (Fig. 2). At the mean population level, *Helisoma* took more time to displace from their shell refuge (i.e., became relatively “shy” or sensitized) over repeated trials (mean effect [95% credible interval] 0.242 [0.173, 0.311], Table 1). *Physa* also showed a positive response to repeated trials, though the credible interval overlapped with zero (mean effect [95% credible interval] 0.031 [-0.047, 0.108], Table 2).

However, unlike the behavioral response to risk cue concentration, individual snails within both species responded differently to repeated assays (individual slopes included in the best-fitting models; *Helisoma*: Table 1, *Physa*: Table 2); some snails increased avoidance behavior while others decreased avoidance behavior (Fig. 2). *Helisoma* individuals mainly responded positively to the number of assays, whereas *Physa* individuals responded both positively and negatively (Fig. 2).

Trait correlations

For *Helisoma*, individual-level behavioral plasticity in response to repeated assays was weakly explained by behavioral types in predator avoidance (Fig. 3). We found that relatively bold *Helisoma* snails (i.e., those that took shorter to displace on average) became shyer (i.e., longer time to displace) over the course of the trials and vice versa. Thus, in *Helisoma*, differences in random intercepts became smaller over time in the lab, suggesting behavioral homogenization (i.e., “regression to the mean”).

Snail species further exhibited contrasting patterns of state-dependence, or individual-level correlations between behavioral and physiological traits. For *Helisoma*, both significant correlations involved among-individual differences in mean predator avoidance behavior, or behavioral types (Fig. 3). For example, we detected a strong positive correlation between behavioral types and growth rate in *Helisoma* (Fig. 3) whereby shy snails (i.e., those that took longer to displace on average) grew faster in shell length over the 3-week experimental duration than bold snails. Shy snails additionally had lower body condition than bold snails (Fig. 3).

In contrast to *Helisoma*, the only significant state-behavior correlation for *Physa* snails involved among-individual differences in the behavioral response to trial number (i.e., behavioral plasticity): individuals that grew faster over the 3-week experimental duration became less sensitive (i.e., took shorter time to displace) over repeated trials (Fig. 3). In addition, *Physa* snails that grew faster were generally larger and in lesser body condition (Fig. 3).

DISCUSSION

Our study applied the BRN framework to test for behavioral types and individual plasticity within two prey species that differ in their susceptibility to a shared predator. We broadly predicted that a well-defended snail species, *Helisoma trivolvis*, would exhibit more among-individual variation in predator avoidance behavior than a relatively vulnerable species, *Physa acuta*, due to stabilizing selection acting more strongly on *Physa* behavior. This hypothesis was partially supported in that *Helisoma* exhibited nearly 35% more variation in behavioral types than *Physa*. However, in both species, we failed to detect differences in the individual response to risk (i.e., individual plasticity). Instead, both snail species exhibited individual plasticity in response to the number of behavioral assays performed. This latter finding supports that our experimental design was powerful enough to detect subtle differences in individual plasticity and allows us to interpret the lack of differences in the individual risk response as biologically meaningful.

Behavioral reaction norms

While previous studies have tested for differences in boldness among functionally similar species (Ingley et al., 2014; Michelangeli et al., 2020; Nordberg et al., 2021) and populations of the same species from different environments (Brown and Braithwaite, 2004; Harris et al., 2020; Rasmussen and Belk, 2017), such studies have largely compared boldness averaged across individuals due to a lack of repeated measures data. Our study is unique in that we applied the BRN framework to derive a more complete picture of behavioral variation in terms of predator avoidance, including behavioral types and individual plasticity. While both species exhibited substantial variation in behavioral types, plasticity in the response to risk was indistinguishable among individuals. In both species, all conspecific snails increased avoidance behavior to the same degree with increasing risk. A recent study on *Physa* also failed to detect individual differences in the behavioral response to risk (Tariel et al., 2020), albeit with less experimental power (i.e., fewer individuals, risk levels and replicates) than the present study.

Why does mean predator avoidance behavior vary among individuals while plasticity does not in the freshwater snail species studied here? One possibility is that stabilizing selection acts more strongly on plasticity than behavioral types, thereby eroding among-individual variation in behavioral plasticity (Becker et al., 2022; Kim, 2016). Alternatively, stabilizing selection may act on individual plasticity (Toscano, 2017) while diversifying selection acts on behavioral types (Dingemanse et al., 2010). Stabilizing selection on individual plasticity but not behavioral types could occur because plasticity is exhibited in the presence of predators, while behavioral types instead could be considered a mean, background level of individual behavior. Additionally, selective forces on predator avoidance behavior likely change over time (e.g., seasonally) (Palmer et al., 2022) and over the ontogenetic growth trajectory of snails (Alexander and Covich, 1991), though our study provides a temporal snapshot of among-individual behavioral variation in adult snails. Finally, population-level behavioral responses to risk in our study were statistically significant but somewhat weak, which could limit the potential for individual differences in plasticity if all individuals respond to risk in the same direction.

Regardless of the reason, the lack of individual differences in the behavioral response to risk limits the possibility that individual plasticity impacts the predator-prey interactions that these snails engage in, or that behavioral types and individual plasticity covary in meaningful ways (Mathot et al., 2012). Still, our findings have important implications for how boldness is measured. While boldness has been measured using a variety of assays under different contexts, studies most often measure boldness as time to emerge from a refuge under a single level of risk (Beckmann and Biro, 2013). However, this approach could be misleading if individuals shift their behavior differently across levels of risk (i.e., exhibiting individual differences in plasticity) and thus change in rank order of boldness. In the two freshwater snail species we studied, repeated assays at any level of risk adequately capture boldness.

While we failed to detect individual differences in the behavioral response to risk, individual snails did differ in their response to repeated behavioral assays. On average, both snail species increased time to displace from the shell refuge over the course of the experiment (i.e., became shyer or more sensitive), but individual snails exhibited divergent responses to repeated assays (see also (Rodríguez-Prieto et al., 2011)). In *Helisoma*, the direction (i.e., the slope) of such individual plasticity was inversely related to behavioral types, where initially shy snails became more bold, suggesting behavioral homogenization. While our study was conducted with field collected snails and thus conditioning to the lab might be expected, a recent study using lab-reared *Helisoma* showed a similar pattern: shy snails became bolder over repeated assays, though bold snails showed no evidence of conditioning (Goodchild et al., 2020). We intentionally designed our experiment to avoid such conditioning effects by conducting a single behavioral observation per individual snail per day, with 3-day breaks after every 4 experimental days. Unfortunately, such repeated measurements across environmental gradients are a core feature of the BRN framework and unavoidable in BRN applications (Dingemanse et al., 2010). Our finding of conditioning across both snail species suggests that future studies could consider holding animals in the field, rather than the lab, between behavioral assays, or designing experiments that allow separating the effects of experimental treatments and conditioning, as we did here.

Patterns of state-dependence

Our study additionally detected differences between snail species in relationships between individual behavior and physiological traits (i.e., state-dependence), including growth rate and body condition. For *Helisoma*, we found that shy snails (those that took longer to displace) grew faster over the 3-week experimental duration but were also in lesser body condition at the end of the experiment. Key to interpreting these effects is how we measured growth rate and body condition. While snails that grow faster might be expected to do so by adding mass, we measured growth rate as change in shell size and body condition as the residuals from a length-mass regression, thus removing the effect of shell size. Accordingly, the finding that shy snails grew faster could suggest that shy snails prioritize shell growth over tissue mass to enhance protection while remaining in their shells longer (i.e., adaptive growth: (Irie and Iwasa, 2005)). These results are in accord with a recent study on *Helisoma* snails where slow-emerging individuals invested more in morphological shell defense than bold snails (Goodchild et al., 2020). Additionally, the lower body condition of shy snails could reduce available energy for rapid escape behaviors (Luttbeg and Sih, 2010), reinforcing this pattern of state-dependence.

In contrast, for *Physa* snails, among individual differences in mean predator avoidance (i.e., behavioral types) were unrelated to physiological traits. Instead, individuals that grew faster became less sensitive (i.e., took less time to displace) with repeated trials. While the mechanism behind this effect is unclear, the finding that physiological traits can affect the individual trajectory of conditioning to the lab environment is novel, and again suggests that studies applying the BRN framework should be aware of the potential for behavioral changes driven by individual traits unrelated to experimental manipulations.

CONCLUSION

While behavioral change in response to predation risk is well-studied (Lima and Dill, 1990; Palmer et al., 2022; Schmitz et al., 2004; Werner and Anholt, 1993), few studies have explored whether among-individual responses to heightened risk match or differ from those observed at the population level. Such studies are necessary to establish the role of individual plasticity in ecological and evolutionary processes, but require powerful experimental designs capable of parsing among- vs. within-individual sources of behavioral variation (Martin et al., 2011). Our BRN application provides new insight into the behavioral landscape of predator avoidance in a classic model system, showing that while behavioral types vary considerably in freshwater snails, individual differences in the response to risk are absent and instead match those observed at the population-level. These findings highlight that behavioral types and plasticity can manifest at different organizational scales, and are likely governed by different forms of selection. We propose that new studies applying the BRN framework while directly manipulating predation risk are necessary to establish whether individual plasticity is a context-dependent phenomenon or ubiquitous like behavioral types (Bell et al., 2009).

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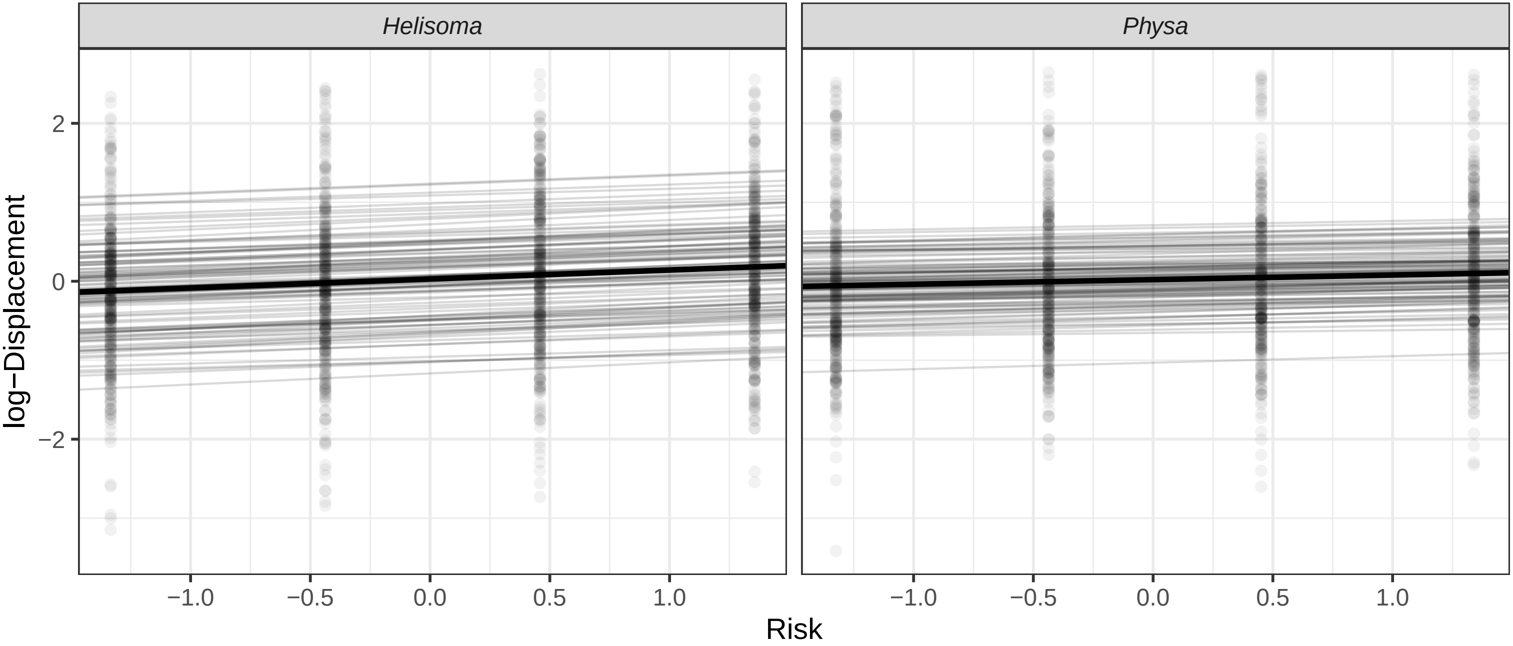
Figure captions

Fig. 1. Log displacement time of both snail species in response to the level of risk. The log-displacement and risk were normalized (i.e., mean centered and unit variance). The black centered line is the population mean effect and the gray lines are the effect of each individual. Points are the observed data.

Fig. 2. Log displacement of both snail species in response to the trial number. The log-displacement and trial were normalized (i.e., mean centered and unit variance). The black centered line is the population mean effect and the gray lines are the effect of each individual. Points are the observed data.

Fig. 3. Posterior distributions of the correlations between each individual-specific parameters computed from the multivariate model in both snail species. Multivariate models included the displacement latency time (referred to here as "Shyness"), body size (referred to here as "Size"), body condition, and growth rate (referred to here as "Growth"). Points are the mean values, thick lines are the 50% credible intervals, and thin lines are the 95% credible intervals.


Helisoma
Physa
log−Displacement
2
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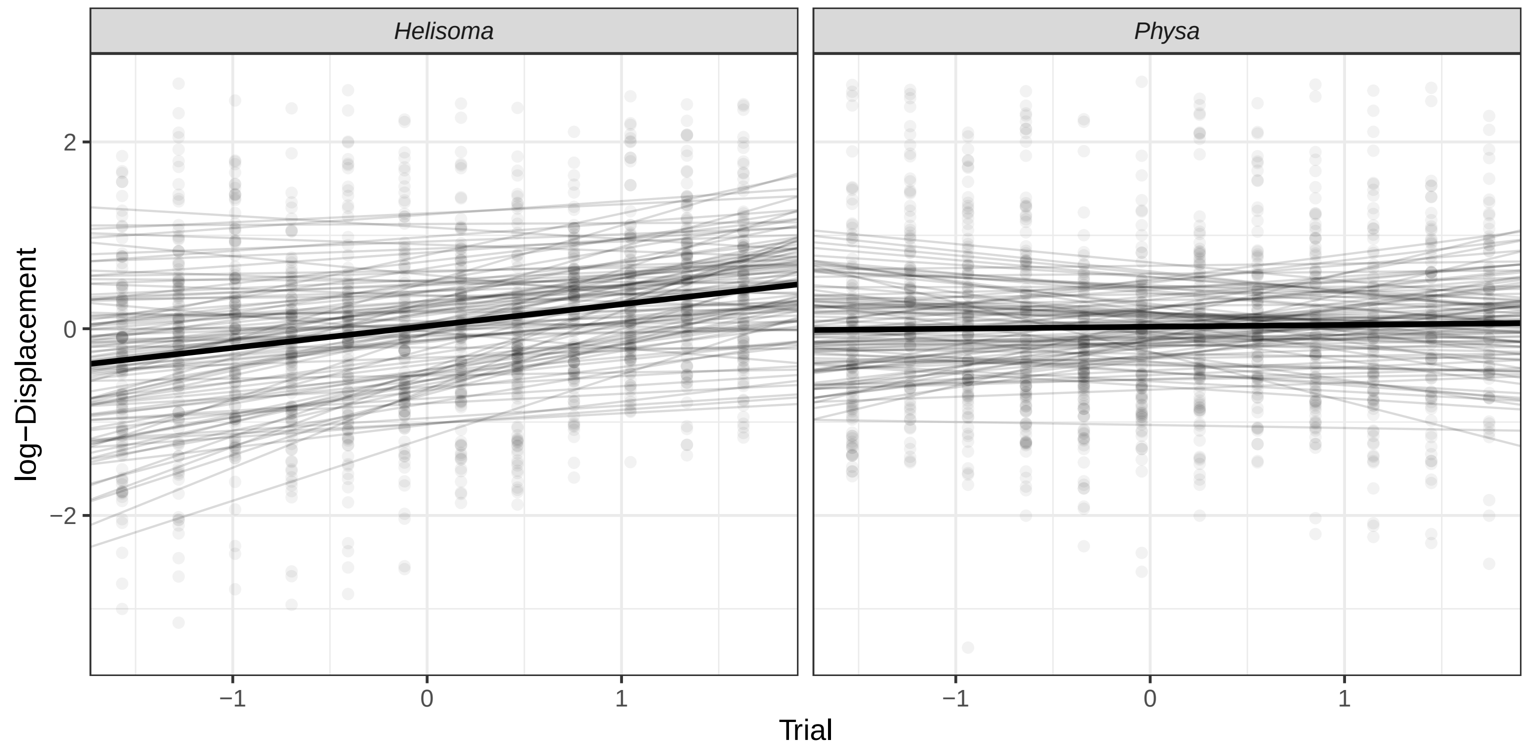
Helisoma
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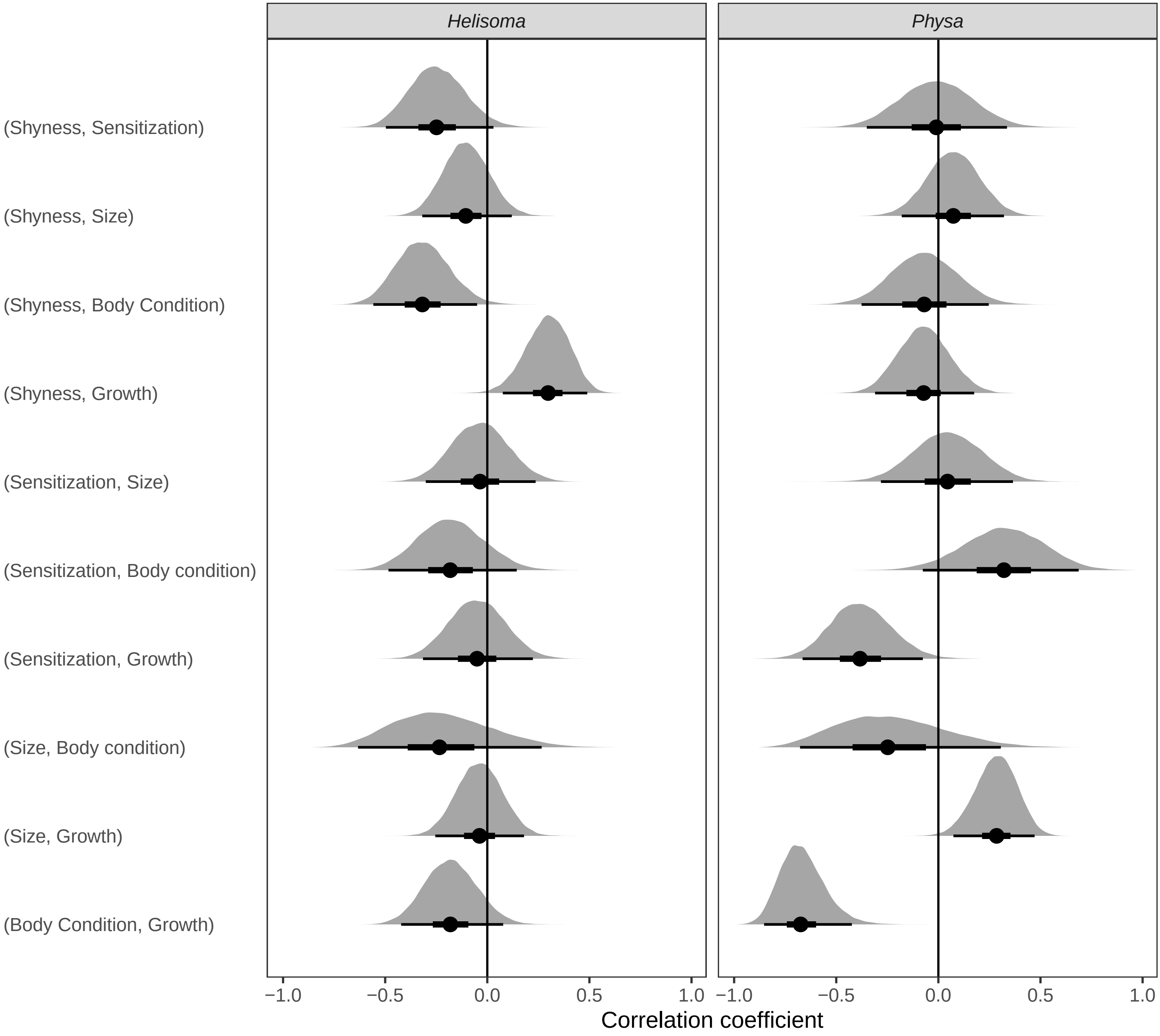
Helisoma
Physa
(Shyness, Sensitization)
(Shyness, Size)
(Shyness, Body Condition)
(Shyness, Growth)
(Sensitization, Size)
(Sensitization, Body condition)
(Sensitization, Growth)
(Size, Body condition)
(Size, Growth)
(Body Condition, Growth)
−1.0
−0.5
0.0
0.5
1.0
−1.0
−0.5
0.0
0.5
1.0
Correlation coefficient

Table 1. *Helisoma* multivariate model output. Each column represents a response variable model. Estimates are the mean of the effect posterior distribution with ± the standard error and the [95% credible interval]. Fixed effects include the intercept, the risk level, the trial number, and the log-transformed body size. The random effects include the among-individual standard deviation at the intercepts, the among-individual standard deviation in the effect of the trial number, and the residual standard deviation. The marginalized coefficient of determination (R^2_{mar}) is the proportion of the variance explained by the fixed effects, and the marginalized repeatability (R_{mar}) is the proportion of the variance explained by the among-individual variation averaged across the gradient of the number of trials. All variables are normalized, i.e., mean centered and unit variance, prior to model fitting.

	Helisoma (n = 90)			
Parameter	log-Displacement	Size	log-Mass	Growth rate
Fixed effects				
Intercept	-0.022 ± 0.064 [-0.148, 0.104]	-0.001 ± 0.073 [-0.144, 0.141]	-0.003 ± 0.046 [-0.092, 0.086]	0.003 ± 0.073 [-0.141, 0.146]
Risk	0.108 ± 0.023 [0.063, 0.152]			
Trial	0.242 ± 0.035 [0.173, 0.311]			
log-Size			0.971 ± 0.088 [0.800, 1.147]	

Random effects				
sd(Intercept)	0.583 ± 0.051 [0.492, 0.689]	0.984 ± 0.082 [0.839, 1.160]	0.403 ± 0.053 [0.308, 0.517]	0.987 ± 0.083 [0.840, 1.163]
sd(Trial)	0.261 ± 0.035 [0.196, 0.332]			
sd(Residuals)	0.728 ± 0.018 [0.694, 0.763]			
R^2_{mar}	6.8% [3.9%, 10.2%]			
R_{mar}	41.3% [34.1%, 48.8%]			

Table 2. *Physa* multivariate model output. Each column represents a response variable model. Estimates are the mean of the effect posterior distribution with ± the standard error and the [95% credible interval]. Fixed effects include the intercept, the risk level, the trial number, and the log-transformed body size. The random effects include the among-individual standard deviation at the intercepts, the among-individual standard deviation in the effect of the trial number, and the residual standard deviation. The marginalized coefficient of determination (R^2_{mar}) is the proportion of the variance explained by the fixed effects, and the marginalized repeatability (R_{mar}) is the proportion of the variance explained by the among-individual variation averaged across the gradient of the number of trials. All variables were normalized, i.e., mean centered and unit variance, prior to model fitting.

	Physa (n = 99)			
Parameter	log-Displacement	Size	log-Mass	Growth rate
Fixed effects				
Intercept	0.019 ± 0.052 [-0.082, 0.124]	0.014 ± 0.072 [-0.128, 0.155]	0.000 ± 0.041 [-0.080, 0.081]	0.003 ± 0.070 [-0.136, 0.141]
Risk	0.058 ± 0.028 [0.003, 0.115]			
Trial	0.031 ± 0.039 [-0.047, 0.108]			
log-Size			0.977 ± 0.084 [0.814, 1.146]	

Random effects				
sd(Intercept)	0.433 ± 0.048 [0.343, 0.533]	1.003 ± 0.081 [0.856, 1.175]	0.358 ± 0.054 [0.259, 0.472]	0.978 ± 0.079 [0.837, 1.146]
sd(Trial)	0.260 ± 0.049 [0.162, 0.357]			
sd(Residuals)	0.876 ± 0.023 [0.833, 0.921]			
R^2_{mar}	0.6% [0.0%, 1.7%]			
R_{mar}	24.9% [18.1%, 32.4%]			