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# Plasticity constrained: over-generalized induction cues cause maladaptive phenotypes

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## ABSTRACT

The adaptive value of phenotypic plasticity depends upon the degree of match achieved between phenotype and environment. This match is governed in part by how well organisms discern environmental states (e.g. presence or absence of particular predators). To examine the specificity with which organisms use cues to induce defensive phenotypes, we raised freshwater snails, *Physella virgata*, with either molluscivorous or non-molluscivorous sunfish species (*Lepomis cyanellus*, *L. gibbosus*, *L. macrochirus*, *L. megalotis*, *L. microlophus*, *Micropterus salmoides*). Regardless of the predator class with which they were raised, snails exhibited two induced responses: (1) reduced growth and (2) development of rotund shells. Reduced growth typically results from predator avoidance behaviours and so is associated with a net survival benefit in environments containing molluscivores. Rotund shells increase crush resistance and so increase both handling time and rejection rates by molluscivorous sunfish. Despite the adaptive nature of these responses to molluscivorous species, responding unnecessarily to non-molluscivores is maladaptive. Growth reduction limits fecundity and prevents snails from attaining size refugia for most predators. Rotund shells increase vulnerability to shell-entry predators, which are typically more common than shell-crushing predators. Thus the induced responses entail costs but award no advantages in the presence of non-molluscivorous sunfish. Lack of specificity in use of cues (e.g. all sunfish treated alike) may be a major constraint on the evolution of phenotypic plasticity. Furthermore, these induced responses may produce trait-mediated indirect effects that cascade throughout the food web.

*Keywords*: evolutionary constraint, geometric morphometrics, inducible defences, multiple predators, phenotypic plasticity, shell morphology, trait-mediated indirect effects.

## INTRODUCTION

Research on environmentally contingent development (phenotypic plasticity) has burgeoned in the last few decades (Schlichting, 1986; Stearns, 1989; West-Eberhard, 1989; Scheiner, 1993). Early studies demonstrated that phenotypic plasticity is a trait under genetic control (Weis and Gorman, 1990; Scheiner, 1993; Via, 1993; Pigliucci, 1996) and is

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often an adaptation to fluctuating environments (Bradshaw, 1965; Levins, 1968; Scheiner, 1993; Gotthard and Nylin, 1995). More recently, the emphasis has shifted from simply documenting case after case of adaptive plasticity to addressing constraints on the evolution of phenotypic plasticity (Via *et al.*, 1995; DeWitt *et al.*, 1998) and effects of plasticity on community ecology (Chase, 1999; Relyea, 2000). Although these two topics are linked, we focus here on constraints. Constraints are presumed to exist because in their absence all traits in all organisms would exhibit perfect plasticity, which is clearly not the case.

Constraints on plasticity can involve costs or limits (DeWitt *et al.*, 1998). Costs of plasticity are indicated when a phenotype produced through flexible development confers lower fitness than the same phenotype produced by fixed development (Van Tienderen, 1991). Limits of plasticity are logistic problems that reduce the phenotype–environment matching ability relative to the theoretical maximum (DeWitt *et al.*, 1998). Costs and limits have received considerable theoretical attention in recent years (Via and Lande, 1985; Van Tienderen, 1991; Moran, 1992; Leon, 1993; Getty, 1996; Tufto, 2000), but empirical tests have only recently emerged (e.g. DeWitt, 1998; Scheiner and Berrigan, 1998; Donohue *et al.*, 2000; Weinig and Delph, 2001; Relyea, 2002; Van Buskirk, 2002).

Constraints on the evolution of plasticity may arise because flexible development can involve steps that fixed development does not (DeWitt *et al.*, 1998). For example, flexible development may require detection of a reliable cue to indicate the state of the environment. In this case, a receptor mechanism must exist in the organism to transduce environmental information into a vehicle that can interact with development (Stearns, 1989; Scheiner, 1993; Pigliucci, 1996). If a given cue is unreliable, or if the receptor is not specific enough, this will limit the phenotype–environment matching achieved by the organism.

In the present study, we examined predator-induced traits in freshwater snails to assess the specificity (or lack thereof) of environmental cues used to inform development about the nature of the environment. We measured phenotypic responses of the snail *Physella* (= *Physa*) *virgata* raised with six different sunfish species that vary in feeding habits. Specifically, we wished to determine whether the snails could discriminate between molluscivorous and non-molluscivorous species by producing appropriate phenotypic responses. Unnecessary induced responses that were costly but provided no compensatory benefits were considered to be constraints on the evolution of phenotypic plasticity. Finally, we addressed how over-generalized responses can create trait-mediated indirect effects.

## MATERIALS AND METHODS

### Snails and collection site

Approximately 150 *Physella virgata* were obtained from a pond in College Station, Texas, USA (30°36′N, 96°17′W). This pond was selected as our collection site because it contained all fish species used in the experiment that were native to the area. Pumpkinseed sunfish (*Lepomis gibbosus*), which do not occur in Texas or bordering states, were obtained from a commercial supplier (Mark Binkley, Columbus, Ohio, USA). These fish were included in the experiment as a positive control, as they are known to induce changes in morphology and growth rate in a congeneric snail (DeWitt, 1998). All other sunfish used in the experiment were collected from local ponds. Wild-caught snails were treated with antibiotic (erythromycin 25 $\mathrm{mg \cdot l^{-1}}$) for 3 days in group culture and fed Wardley's brand spirulina flakes *ad libitum* for a week. During this period, several hundred egg masses were generated. Hatchlings from these egg masses were moved using a small (size 0) paintbrush, 7–10 days later, into seven different environments as described below.

## Experimental design

$F_1$ snails were raised in seven treatments: negative control (no fish), bluegill sunfish (*Lepomis macrochirus*), green sunfish (*L. cyanellus*), longear sunfish (*L. megalotis*), redear sunfish (*L. microlophus*), positive control (pumpkinseed sunfish, *L. gibbosus*) and largemouth bass (*Micropterus salmoides*). The six sunfish used in the experiment differ in their molluscivory. Only two of the fish (redear and pumpkinseed) are considered to be molluscivores. Two 52-litre aquaria were established for each treatment and all tanks were systematically interspersed. A single fish (7.43 ± 1.74 cm standard length) was placed below a plastic grid in sunfish treatment tanks to allow flow-through of fish chemical cues while preventing physical contact between fish and snails. Snails were placed individually into ten cages above the grid in each tank. Each rearing cage was made of 300-ml plastic cups which had two ($35 \times 38$ mm) mesh windows (mesh size = 0.10 mm) to allow water exchange with the surrounding medium. The cages rested atop the grid such that approximately 1.5 cm of air space remained in each cage to permit snails to crawl out of the water. Crawling to the waterline or above the waterline is a typical anti-predator behaviour in physid snails (Snyder, 1966; Alexander and Covich, 1991b; Turner, 1995, 1997; DeWitt *et al.*, 1999; McCarthy and Fisher, 2000).

The experiment was conducted at 20–25°C with an 11L:13D photoperiod. We fed snails ground spirulina flakes *ad libitum*. Commercial food pellets and frozen bloodworms were fed twice a week to the lepomid sunfish. Since largemouth bass do not eat small, inanimate food, they were fed western mosquitofish (*Gambusia affinis*). We did not feed snails to the sunfish and no injured snail cues were presented; any induced responses were caused by cues of sunfish presence rather than cues of actual predation. Ten percent water changes were conducted every other week to maintain high water quality. Twelve snails died immediately after their transfer into treatments and were not included in our analyses. One snail was excluded due to obvious shell deformity, making our final sample size 127 snails.

## Morphometrics

After 42 days of rearing, snail shell images were captured with a video imaging system and measured using MorphoSys morphometric software (Meecham, 1993). Shells were placed aperture down and allowed to rest naturally on a stage below a video camera (Elmo model SE360S) equipped with a telecentric lens (Computar model TEC-M55). Video from the camera was captured at a resolution of 0.01 mm per pixel using a computer framegrabber board (PCVision Plus model AT OFG). We digitized shell outlines and nine landmarks for each shell (Fig. 2a) using MorphoSys software. Landmarks (LM) were digitized at the shell apex (LM 1), on sutures connecting the current and previous two whorls (left and right sides of the shell; LM 2–5), at the widest parts of the shell relative to the coiling axis (left and right; LM 6, 7), at the lower insertion of the aperture (LM 8) and at the farthest point from the apex (LM 9).

Shell size was measured as the logarithm of area within shell outline. Gross shell shape was measured as the square root of the shell aspect ratio. The aspect ratio is the ratio of shell length to shell width. Shell length is the length of the long axis within the shell outline and shell width is the length of an axis halfway along, and perpendicular to, the long axis (see fig. 2 in DeWitt *et al.*, 2000). This shell shape measurement was used because previous work has shown it to be informative of the functional ecology of predator resistance (DeWitt, 1998; DeWitt *et al.*, 1999, 2000).

We used geometric morphometric methods (Rohlf and Marcus, 1993) to obtain detailed shell shape information. In general, geometric morphometric methods provide greater power than traditional (linear-distance) methods to address shape differences by retaining information about spatial covariation between landmarks (Rohlf and Marcus, 1993). Superimposed landmark configurations for all specimens were used to calculate uniform and partial warps using tpsRegr software (Rohlf, 1998). TpsRegr rotates, translates and scales landmark coordinates into alignment through generalized least squares superimposition (Bookstein, 1991) and re-expresses coordinates as bending energies using thin-plate-splines. This procedure allows for a comparison of shapes in a dimensionless, multivariate shape space. Affine components of shape variation (uniform spatial covariation in the x- and y-planes) were extracted and local deviations from this covariance suite were summarized as subsequent partial warps. Partial warps served as our shape variables and were entered into multivariate analyses as described in the next section. Since tpsRegr permits regression of shape onto multiple independent variables (i.e. treatment *and* centroid size), all visualizations depict shape differences corrected for multivariate allometry.

Thin-plate-spline visualizations of overall shell morphology suggested the need for a closer examination of shell aperture shape, so we performed geometric morphometric analyses of shell apertures. Using MorphoSys, we clipped the apertural region of shell contours by connecting points at the top (LM 5) and bottom (LM 8) of the aperture with a line and excising the apertural portion of the shell outline. From the midpoint of our excision line, we ran transects every 10° and digitized additional points at their intersections with the apertural outline (Fig. 2b). We used tpsRegr software with these points ($n = 19$) to visualize apertural shape differences between the control treatment and each of the sunfish treatments. Since apertural points have no true homology from one specimen to the next (*sensu* Bookstein, 1991), they are not true landmarks. Therefore, our use of this technique was heuristic and no statistical tests were conducted on these data.

## Statistical analyses

For each trait we fitted a general statistical model and then contrasted all sunfish treatments with the control. When sunfish effects were significant, we performed Tukey's test to establish whether effects were homogeneously or heterogeneously distributed across sunfish. When heterogeneous effects were noted, they were evaluated for correspondence with functional hypotheses about molluscivory. All analyses were conducted using JMP software (Version 4.04, SAS Institute Inc., Cary, NC).

Shell size (log shell area) and gross shape (square root aspect ratio) were analysed using analysis of variance (ANOVA). Sunfish environment was considered to be a fixed effect and tank effects (nested within environments) were considered to be random. Two assumptions were explicitly defined to clarify which type of mixed model analysis best fitted our experimental design (Hocking, 1985). First, we assumed that response variables may be correlated within tanks (i.e. tankmates might look alike) due to random but additive tank effects. Second, any such correlations within tanks are unlikely to differ systematically by treatment. Thus, this was a Model 1 mixed model (Hocking, 1985; for a discussion of biological applications, see Ayres and Thomas, 1990; Fry, 1992).

Shell size (log area) was initially used as a covariate in our analysis of aspect ratio. As in previous studies (e.g. DeWitt, 1998), however, shell size and aspect ratio were uncorrelated ($F_{1,112} = 0.64$, $P = 0.43$), so size was removed from the statistical model. Following Sokal and Rohlf (1995), mean squares in the analysis of aspect ratio were pooled because aspect ratio, as in previous studies (e.g. DeWitt, 1998), was not significantly influenced by tank effects ($F_{7,113} = 0.65$, $P = 0.7$). Pooling did not change the significance structure of the analysis.

Geometric morphometric shape data were analysed using multivariate ANOVA (MANOVA). Shape parameters (i.e. $n = 14$ partial warps) from our nine digitized points were tested for variation attributable to sunfish treatments, tank effects nested within treatments and centroid size. Contrasts and *post-hoc* tests were performed using the major canonical variates from the MANOVA. All tests for treatment effects on shape included centroid size to control for multivariate allometry.

## RESULTS

Snails reduced growth and produced rotund shells in response to sunfish (Table 1). Growth contrasted markedly between sunfish treatments and the control (planned contrast, $F_{1,113} = 21.08$, $P < 0.0001$). Each sunfish species induced a similar growth reduction, apart from green sunfish, which induced a particularly pronounced effect (Tukey's test, $P < 0.05$; Fig. 1). Although aspect ratio varied by treatment, the contrast between sunfish and controls was not significant (planned contrast, $F_{1,120} = 2.13$, $P = 0.15$), probably because the amount of induction differed by sunfish treatment, although all sunfish seemed to induce the same qualitative response (see below).

Our geometric morphometric analysis also demonstrated shape differences across treatments (Wilk's $\lambda = 0.322$, $F_{84,558.17} = 1.50$, $P = 0.005$). Shell shape varied with size

**Table 1.** Rearing environment effects on shell size and shape: results of ANOVA assessing differences in size and shape of snail shells between treatments and tanks nested within treatments

| Dependent variable | Source | d.f. | MS | $F$ | $P$ | $F_{pool}$ | $P_{pool}$ |
|---|---|---|---|---|---|---|---|
| Shell size | Fish environment | 6 | 1.0429 | 9.38 | < 0.0001 | | |
| (log shell area) | Tank (fish environment) | 7 | 1.2425 | 11.18 | < 0.0001 | | |
| | Error | 113 | 0.1111 | | | | |
| Shell shape | Fish environment | 6 | 0.0033 | 3.59 | 0.003 | 3.66 | 0.002 |
| (square root | Tank (fish environment) | 7 | 0.0006 | 0.65 | 0.7 | | |
| aspect ratio) | Error | 113 | 0.0009 | | | | |

*Note*: Mean squares were pooled in the analysis of shell shape (square root aspect ratio) following Sokal and Rohlf (1995).

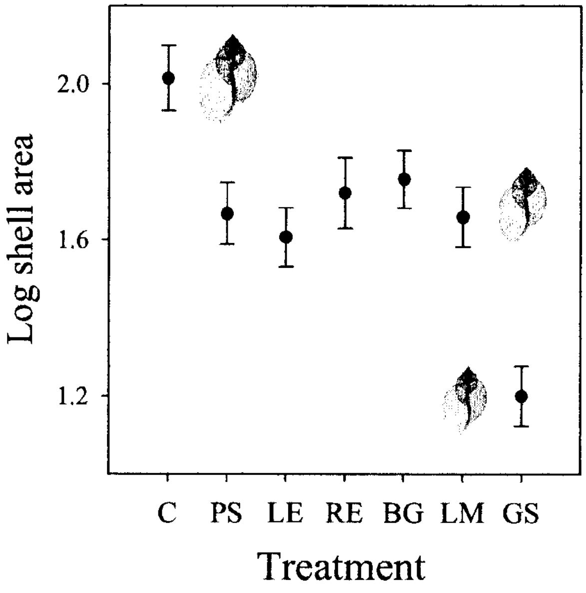

**Fig. 1.** The shell size of snails raised in the seven environments (mean ± standard error): control (C), pumpkinseed sunfish (PS), longear sunfish (LE), redear sunfish (RE), bluegill sunfish (BG), largemouth bass (LM) and green sunfish (GS).

($F_{14,99} = 8.12$, $P < 0.0001$), indicating multivariate geometrical allometry, but tank effects on shape were not evident (Wilk's $\lambda = 0.403$, $F_{98,635.05} = 1.00$, $P = 0.48$). The major axis of variation (i.e. first canonical variate) separated sunfish treatments from the control (planned contrast, $F_{1,112} = 6.20$, $P = 0.014$). Thus, the major axis of phenotypic variation, which accounted for 32.8% of phenotypic variation, was associated with a general sunfish effect. Sunfish effects were not homogeneous, however, as pumpkinseed sunfish differed from all sunfish except bluegill, and bluegill differed from green and redear sunfish (Tukey's test, $P < 0.05$). Snails raised with green and redear sunfish exhibited the least difference from the control treatment. The rank order of sunfish treatments along canonical axis 1 was: pumpkinseed > bluegill > largemouth bass > longear > green > redear. Canonical axis 2 and subsequent axes explained less variance than the first and failed to discriminate between treatment levels.

We interpret canonical axis 1 as a multivariate geometrical analogue of our gross measure of shape (aspect ratio). The two variables were correlated by $r = 0.44$ ($P < 0.0001$). Thus sunfish induced rotund shells, but the nature of the change was not fully captured by the grosser shape measure of aspect ratio.

Thin-plate-spline visualizations and our statistical analysis both showed that sunfish induced a qualitatively similar response in the snails (Fig. 2a). A substantial portion of induced shell shape change seemed to be in the apertural region of shells (lower left of shell grids in Fig. 2a). Therefore, we generated thin-plate-spline transformation of our 19 apertural points using tpsRegr and again found similar shape change across sunfish treatments. When control snails were compared with those raised with sunfish, control

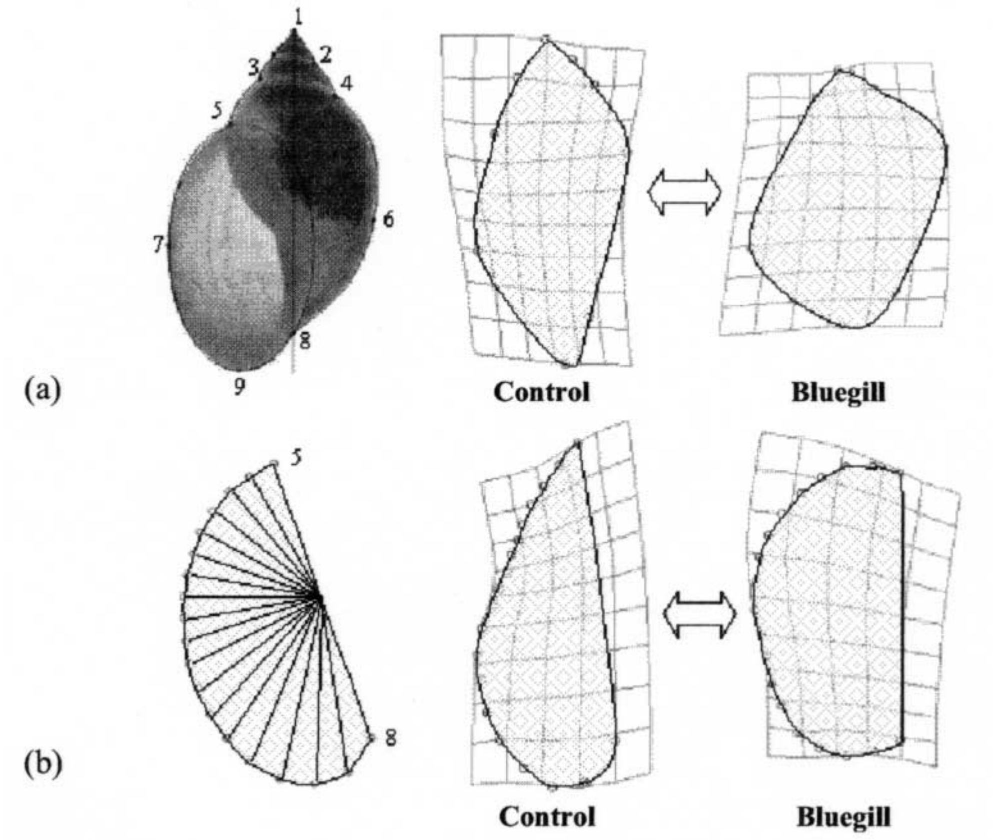

**Fig. 2.** Shell and aperture shapes produced in control and sunfish environments. Leftmost panels indicate landmarks used in our analysis. Middle and right panels illustrate thin-plate-spline transformations of control and sunfish-induced morphologies, respectively. The bluegill effect was selected as representative of the generalized sunfish response. (a) Sunfish effect on overall shell morphology. Sunfish induced rotund shells with mostly affine (uniform y dimension) changes and localized effects in the lower left quadrant (i.e. apertural region). (b) Sunfish effect on apertural region. This deformation grid illustrates a conversion from teardrop- to arch-shaped apertures induced by sunfish. All visualizations control for multivariate allometry and were produced using tpsRegr (Rohlf, 1998) with magnification of 10 × .

apertures appeared to be teardrop-shaped while sunfish-induced apertures appeared to be arch-shaped (Fig. 2b). Since transformation grids for all sunfish were similar, we displayed only the bluegill effect, which typified the shape change seen in all sunfish treatments.

## DISCUSSION

Our results show that *P. virgata* snails responded to all sunfish species by reducing growth and producing relatively rotund shells. These induced changes appear to be a generalized response to sunfish. Our expectations, based on adaptive hypotheses, were that responses should be weak or absent for non-molluscivorous sunfish species. Although the responses exhibited some quantitative differences between sunfish, the variation suggested no functional patterns. In fact, the patterns observed tended to be antithetical to adaptive hypotheses. If non-molluscivorous fish induce costly yet unnecessary phenotypes (as we describe below), the end-product of development can be considered an adaptive error. As errors, such induced phenotypes represent a constraint on the evolution of phenotypic plasticity (DeWitt *et al.*, 1998).

Reduced growth in these snails has both direct (fecundity limitations) and indirect (functional ecological trade-offs) costs associated with responding to over-generalized cues. The fecundity cost arises because smaller snails are less fecund (e.g. McMahon, 1975; DeWitt, 1991). Furthermore, for most predators of freshwater snails, prey vulnerability decreases with size (e.g. leeches: Townsend and McCarthy, 1980; crayfish: Alexander and Covich, 1991a; R.B. Langerhans and T.J. DeWitt, unpublished data; flatworms: Tripet and Perrin, 1994; mollusvicorous sunfish: R.B. Langerhans and T.J. DeWitt, unpublished data). So, unnecessarily reducing growth prevents snails from achieving the size necessary to reduce predation. Growth reduction of physid (*P. heterostropha*) snails raised with pumpkinseed sunfish has been observed previously and attributed to anti-predator behaviour (DeWitt, 1998). Physid snails crawl up vertical surfaces and out of the water in response to chemical cues of predation (Alexander and Covich, 1991b; Covich *et al.*, 1994; Turner, 1995; DeWitt *et al.*, 1999; McCarthy and Fisher, 2000). While out of the water, snails are safe from predatory fish (Turner, 1997) but cannot feed. Thus, while avoidance behaviours increase fitness in the presence of molluscivorous fish, they unnecessarily limit fecundity and disallow size refugia from possible predators in the absence of such fish.

The other trait in our study (shell shape) exhibited a similar pattern: a qualitatively similar response to all sunfish, where quantitative differences between sunfish species were mostly antithetical to adaptive hypotheses. Sunfish induced rotund shells, which appeared to result primarily from the induction of relatively arch-shaped apertures (Fig. 2). Based on architectural principles alone, we expect the arch conformation to better resist crushing. Furthermore, it has been shown empirically that rotund shells offer superior crush resistance and snails with rotund shells experience prolonged handling time and greater rejection rates by molluscivorous sunfish (DeWitt *et al.*, 2000). So there are clear adaptive benefits for the induced response in the presence of molluscivorous sunfish. In the absence of molluscivorous fish, such responses are unnecessary and probably maladaptive. Rotund shells make snails more vulnerable to shell-entry predators (e.g. crayfish, leeches, aquatic hemiptera) (DeWitt *et al.*, 2000; R.B. Langerhans and T.J. DeWitt, unpublished data). For these predators, which are ubiquitous in physid habitats, rotund shells (i.e. shells with broadly arching apertures) provide easy access to snail bodies. Snails raised in the absence of predators produced elongate (i.e. entry-resistant) rather than rotund shells (see also DeWitt, 1998). The production of elongate shells by default suggests that the usual environment for the snails may be composed primarily of shell-entry predators. Therefore, the unnecessary induction of rotund shells by non-molluscivorous sunfish is likely to entail costs in terms of increased vulnerability to alternative predators.

Only two of the six sunfish species in our study (redear and pumpkinseed sunfish) are specialized molluscivores. These fish possess a pharyngeal mill that enables a primarily molluscivorous diet (Lauder, 1983; Osenberg and Mittelbach, 1989; Huckins, 1997). The other sunfish used in our study (largemouth bass, longear, green and bluegill sunfish) only rarely eat snails and lack a pharyngeal mill (Etnier, 1971; Werner and Hall, 1977; Keast, 1978; Mittelbach, 1984; Butler, 1989; Nowlin and Drenner, 1999). Largemouth bass, in particular, seldom if ever eat snails in nature (Anderson, 1984; Mittelbach *et al.*, 1995; Olson, 1996; Huskey and Turingan, 2001), even when lack of prey fish forces them to adopt an invertebrate diet (Schindler *et al.*, 1997). In rare instances where small numbers of snails are ingested by non-specialized sunfish, the snails are extremely small and are swallowed whole (e.g. Gerking, 1962). Rotund shells increase crush resistance but do not affect the shell-swallowing mode of predation, since the snails are too small to gape limit predators. Therefore, induced (i.e. rotund) shell shapes in these cases are still non-adaptive at best.

To be adaptive, costly defences should only be assumed when compensatory benefits exist. Yet in the present study, snails suffered reduced growth (hence reduced fecundity), were denied size refugia and produced entry-prone shells in response to non-molluscivorous fish. Snails suffered the greatest growth reduction in response to green sunfish (Fig. 1), which is among the fishes least likely to eat snails. *Physella virgata* exhibited the greatest differences in shell shape for pumpkinseed and bluegill sunfish. Bluegill sunfish are primarily planktivorous (Keast, 1978; Mittelbach, 1984) and pumpkinseed sunfish, although molluscivorous, are not native within the snail's range. Native redear sunfish – the sister species of pumpkinsed sunfish (Mabee, 1993) and a more voracious molluscivore (Huckins, 1997) – induced the lowest magnitude of shape change. If snails could discriminate and adaptively adjust responses to various sunfishes, the strongest response should have been induced by redear sunfish.

The apparent inability of snails to respond differentially to molluscivorous sunfish suggests that either (1) species-specific sunfish cues do not exist or (2) specific cues exist but snails lack specific receptors for the cues. The net effect of generalizing perception of environmental cues is that organisms cannot effectively discriminate between non-threatening and threatening environments. Thus, information on the state of the environment is noisy, at best. Information reliability has often been seen as an important limiting factor for phenotypic plasticity (Levins, 1968; Moran, 1992; Getty, 1996; Leon, 1993; DeWitt *et al.*, 1998; Tufto, 2000) but, to our knowledge, until now has never been demonstrated empirically for developmental plasticity.

Responses to non-molluscivorous sunfish could be adaptive if the scent of those sunfish could serve as an indirect cue to the additional presence of molluscivorous species. Indirect cues can be useful when sufficiently correlated with threatening environments. Von Elert and Pohnert (2000) demonstrated a possible example with behavioural plasticity of *Daphnia*. *Daphnia* (a zooplankton) responded in their study to both planktivorous shad (dangerous to *Daphnia*) and piscivorous pike (harmless to *Daphnia*). Since pike generally co-exist with planktivorous fish – planktivores are a staple food for pike – the presence of pike scent could indirectly indicate planktivore presence to *Daphnia*. In contrast, in the present snail–sunfish system, the scent of non-molluscivorous species such as largemouth bass or bluegill sunfish is likely to be negatively correlated with the presence of molluscivorous sunfish. Largemouth bass negatively impact other sunfish species through predation, and all sunfish species compete with one another as juveniles (Osenberg *et al.*, 1994). Bluegill in particular negatively impact other lepomid sunfish (Osenberg *et al.*, 1994). Furthermore, in the watershed from which snails in this study originated, redear sunfish (the native molluscivore) occurs relatively infrequently. The presence of any given non-molluscivorous sunfish is typically associated with the absence of redear sunfish (R.B. Langerhans and T.J. DeWitt, personal observations). Thus the sign of correlation between non-molluscivorous and molluscivorous sunfishes, to the extent it exists, is opposite to that which would make it a useful indirect predictor for snails.

So what is an organism to do when sensing one cue (or similar cues that cannot be distinguished), where producing a costly response is adaptive for some sources of the cue and not others? In plants, the ratio of red to far-red wavelengths (R:FR) indicates shading caused by neighbouring plants. If shading is caused by conspecifics or other plants of similar height, then stem elongation allows a plant to overtop its neighbours and obtain more light. In other environments, neighbours are too tall to overtop, so responding to the cue is futile. If such futile responses are also costly, then the response is maladaptive. Weinig (2000) demonstrated that in most environments velvetleaf (*Abutilon theophrasti*) respond to foliar shading with vertical stem elongation. However, velvetleaf cannot overtop corn. Populations of velvetleaf in cornfields have reduced elongation responses to foliar shade. Similarly, forest populations of *Impatiens capensis* do not respond to shading cues but field populations do (Dudley and Schmitt, 1996; Donohue *et al.*, 2000). Even in natural field environments, these plants often experience light competitors that cannot be overtopped. Nevertheless, plasticity *has* evolved, suggesting that benefits achieved when responses are advantageous outweigh costs of futile responses.

The present system is largely analogous to that of the plants discussed above. Here snails apparently perceived a generalized ‘sunfish’ cue. Response to the cue is good in some cases but bad in others. Without a specific cue, snails sometimes end up responding inappropriately – that is, they make adaptive errors. Such errors will not prevent the evolution of plasticity as long as their costs are outweighed by benefits of correctly cued responses. In populations of snails that never experience molluscivorous fish, the cue is not informative of risk (just as when foliar shade cues are meaningless in forests) and local adaptation for reduced plasticity would be expected to evolve. Gene flow is excessive for physid snails (Dillon and Wethington, 1995), which reduces the chance of local adaptation, so the magnitude of differentiation between populations is an interesting question in this system. So far, snails from populations lacking molluscivorous fish have not been assessed for predator-induced plasticity.

Besides constraints and population differentiation, a recent focus in plasticity research is how induced responses affect community-level interactions (Chase, 1999; Relyea, 2000; Agrawal, 2001). The functional ecology of physid snails and their predators suggests the induced responses observed in this study will produce trait-mediated indirect effects that may cascade throughout the food web. That is, responses to non-molluscivorous fish (rotund shells and reduced growth) should lead to increased predation by crayfish and other ubiquitous shell-entry predators. The subsequent increased mortality (and decreased resource consumption) of snails will probably affect several trophic levels, including both predators and algal resources (Vaughn *et al.*, 1993; Gelwick, 2000; Bernot and Turner, 2001). Although several studies have documented trait-mediated indirect effects for behaviour (arguably a plastic trait; e.g. Turner and Mittelbach, 1990; Werner and Anholt, 1996; Peacor and Werner, 1997, 2001; Schmitz *et al.*, 1997; Bernot and Turner, 2001), our results support the emerging emphasis on the potential of life-history and morphological plasticity in creating these indirect effects (Raimondi *et al.*, 2000; Relyea, 2000).

Although predator-induced morphologies can greatly reduce predation (reviewed by Havel, 1987; Tollrian and Harvell, 1999), unreliable or over-generalized cues can lead to costly phenotypic errors. Phenotype–environment mismatches that result from responding inappropriately to the environment are likely to be a major constraint on the evolution of phenotypic plasticity (DeWitt *et al.*, 1998). Yet theoretical models show that plasticity can evolve despite severe constraints (Van Tienderen, 1991; Leon, 1993; Getty, 1996; Tufto, 2000). The present study provides direct empirical evidence that plasticity can evolve even when such constraints operate. Thus the benefits of a correctly cued response must outweigh the costs of unnecessarily responding to imperfect cues.

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