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Reinforcing abiotic and biotic time constraints facilitate the broad distribution of a generalist with fixed traits

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Abstract. Many species are habitat specialists along environmental gradients as a result of contrasting selection pressures, but others maintain broad distributions along such gradients. Phenotypic plasticity explains the persistence of some generalists, but not the broad distributions of species with fixed traits. We combined comparative and experimental data to investigate the role of multiple selection pressures on the distribution of a cased caddisfly (*Asynarchus nigriculus*) across a pond permanence gradient in the Mexican Cut Nature Preserve, Elk Mountains, Colorado, USA. Rapid development in this species facilitates the exploitation of short-duration vernal pools. Comparative data document that slowly growing individuals die from desiccation, suggesting an ongoing selection for rapid development. Surprisingly, development is as fast or faster in long-duration, autumnal ponds where emergence occurs long before drying, and overlaps with the appearance of beetle (*Dytiscus*) predators. In field experiments we found that the last two instars of beetle larvae pose a significant mortality threat to *Asynarchus,* but that threat declines after caddisfly pupation. In natural populations, the caddisflies pupate and emerge just as large beetle instars appear in the ponds. Experimental manipulation of caddisfly size suggests that rapid development in autumnal ponds will both facilitate intraguild predation on other caddisflies and reduce *Asynarchus* cannibalism. Both types of caddisfly interactions should have a positive feedback effect on rapid development via a protein supplement to their detrital diet. All of these biotic time constraints should select for rapid *Asynarchus* development in autumnal habitats, despite relaxed drying time constraints. *Asynarchus* did not display flexible antipredator responses to beetles (no changes in activity rates, morphology, or development), suggesting that the traits that lead to rapid development are fixed, regardless of habitat type and presence of predators. We propose that different, but convergent, selection pressures across different habitat types have led to fixed specialized traits that enable a broad distribution along this environmental gradient. These selection pressures are dependent on the relative phenologies of interacting species and appear to trump the trade-offs between other types of physical and biotic constraints across habitats.

Key words: antipredator traits; Asynarchus nigriculus; *caddisflies;* Dytiscus dauricus; *habitat generalist; hydroperiod; intraguild predation; Mexican Cut Nature Preserve, Elk Mountains, Colorado, USA; phenology; phenotypic plasticity; predator–permanence gradient; specialization.*

Introduction

Heterogeneous landscapes present an array of alternative habitats that can vary greatly in their suitability to an organism. Along many types of environmental gradients, species experience conflicting selection pressures because changes in physical stress are often accompanied by changes in the strength of biotic interactions such as herbivory (Rand 2002), competition (Lubchenco 1980, Crain et al. 2004), or predation (Menge and Sutherland 1987, Wellborn et al. 1996). There are two general types of adaptation that underlie patterns of distribution along such gradients. In some instances, closely related habitat specialists replace each other along environmental gradients, resulting in wholesale shifts in community composition (e.g., Lubchenco 1980, Wellborn et al. 1996). Landscapes where predictable selection pressures vary dramatically between coarse habitat patches are likely to select for the evolution of such specialists (Futuyma and Moreno 1988, Van Tienderen 1991, Tufto 2000, Alpert and Simms 2002). Alternatively, generalist species can persist in different habitats along environmental gradients as a result of (1) fixed intermediate traits that facilitate coexistence with different types of specialists (Levins 1968, McPeek 1996, McCauley 2008); (2) phenotypic plasticity, i.e., the developmental expression of alternative morphs to different environmental cues (West-Eberhard 1989, Nylin and Gotthard 1998); or

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(3) local adaptation of ecotypes via genetic differentiation (Hedrick 1986, Hays 2007). Although a few theoretical and empirical studies have explored the persistence of fixed-trait generalists across environmental gradients (McPeek 1996, McCauley 2008), most studies have focused on phenotypic plasticity, particularly in the context of inducible antipredator responses (Lima 1998, Tollrian and Harvell 1999, Hoverman and Relyea 2007).

Environmental gradients associated with drying in standing freshwater habitats (temporary → semipermanent → permanent without fish → permanent with fish) are ideal study systems in which to investigate mechanisms underlying the persistence of habitat generalists. Along this gradient, the replacement of habitat specialists in a wide range of taxa leads to shifts in community composition (Wellborn et al. 1996). In some cases, these replacements reflect trade-offs between traits selected for by drying time constraints (e.g., high activity and rapid growth) vs. traits that facilitate predator defense in permanent habitats (e.g., low activity) (Skelly 1995, Wissinger et al. 1999*b*, Stoks and McPeek 2006). In other cases, replacements occur between species with different antipredator strategies for coexisting with different predators (Werner and McPeek 1994, Stoks and McPeek 2006, Wissinger et al. 2006). Although species replacements are well documented, there are some species that persist as habitat generalists across drying gradients (McPeek 1996, De Block and Stoks 2005, McCauley 2008). In some systems, these habitat generalists contribute to a nested community pattern that reflects the sequential elimination of specialists in progressively shorter duration ponds (Schneider and Frost 1996, Wissinger et al. 2009). Identifying the mechanisms underlying the distribution and persistence of generalists that occur across the pond-permanence gradient, and environmental gradients in general, will require understanding spatiotemporal variation in the trade-offs that influence species coexistence (McPeek 1996).

The purpose of this study was to investigate the mechanisms that enable the cased caddisfly, *Asynarchus nigriculus,* to inhabit different types of temporary alpine ponds (Appendix A). *Asynarchus* can complete its life cycle in vernal pools that dry in <60 days (Wissinger et al. 2003) by (1) hatching before ice-out in spring; (2) developing rapidly as a result of high activity levels and aggression, including cannibalism (Wissinger et al. 1999*b*, 2004); and (3) investing minimally in case construction, relative to permanent habitat species (Wissinger et al. 2006; see Plate 1). In experimental studies, these traits increase the vulnerability of *Asynarchus* to salamander predators in permanent ponds, and to beetle (*Dytiscus dauricus*) predators in long-duration autumnal ponds (dry in autumn in most years) (Wissinger et al. 1999*b*, 2006). *Asynarchus* only occurs in permanent ponds without salamander predators, but curiously does coexist and is often abundant in autumnal habitats with beetle predators (Wissinger et al. 2003). Moreover, *Asynarchus* does not extend larval development in autumnal ponds beyond that observed in vernal habitats, despite relaxed time constraints. This is nonintuitive, given that extending larval growth should have direct, positive effects on adult survival and fecundity, especially considering that adults do not feed (see reviews by Peckarsky et al. [2001] and Jannot et al. [2007]), and that *Asynarchus* larvae do not alter their "fast lifestyle" behavioral traits (Sih 1987) or exhibit risk-sensitive foraging (Lima 1998).

Thus, we hypothesized that the rapid development of this species in autumnal ponds is related to one or more biotic time constraints associated with intra- and interspecific interactions including (1) reduced time of exposure to beetle predation, (2) reduced cannibalism, or (3) increased opportunity for intraguild predation on confamilials (e.g., *Limnephilus* spp.) that co-occur with *Asynarchus* in autumnal ponds (Wissinger et al. 1996, 2003). To test these hypotheses, we compared the phenologies of *Asynarchus* larval development in adjacent vernal and autumnal habitats and to that of *Dytiscus* beetles in the autumnal ponds. We then conducted microcosm experiments to determine size-specific vulnerability of *Asynarchus* to *Dytiscus* predation, and the effects of relative body size on cannibalism and intraguild predation by *Asynarchus* on *Limnephilus*. Finally, we reared *Asynarchus* in mesocosms with and without beetles to investigate whether the success of *Asynarchus* in autumnal habitats could be explained by beetle-induced phenotypic plasticity in development or foraging behaviors.

Methods

Study site and natural history

This study was conducted in the Mexican Cut Nature Preserve in the Elk Mountains, Colorado, USA. The preserve includes 60 ponds spanning a broad permanence gradient (inundation period range: 40–365 open-water days after snow melt) within an area of 40 ha (for a detailed habitat description, see Wissinger et al. 1999*a*, 2003). Nearest neighbor distances between ponds are typically 10–20 m, and high rates of recolonization across the study site after population crashes suggest that adult caddisfly dispersal among ponds is high (S. A. Wissinger, *unpublished data*). Of the five species of caddisflies at the Mexican Cut, *Asynarchus* is the only species that persists in vernal habitats. These vernal pools contain no known predators or competitors of *Asynarchus*. Autumnal ponds typically support a more diverse food web than vernal pools, with *Dytiscus* beetle larvae as the top predator (final instar body length ~40 mm, density 2.6 ± 0.6 individuals/m^2, mean ± SE) (Wissinger et al. 1999*a*). Odonates and other large-bodied predatory invertebrates are absent from autumnal ponds at the Mexican Cut, and hatchling and metamorphic salamanders in autumnal ponds feed almost exclusively on zooplankton (Whiteman et al. 1994, Wissinger et al. 1999*a*). Thus, *Dytiscus* is the only

major predator of *Asynarchus* in these ponds. *Dytiscus* is rare in permanent ponds due to predation by paedomorphic salamanders. The confamilial competitor and intraguild prey of *Asynarchus*, *Limnephilus externus*, is common in autumnal ponds, and can reach high densities in early instars (50/m^2; Wissinger et al. 1996, 2003). *L. externus* is able to complete development in autumnal ponds, but consistently lags 1–2 instars behind *Asynarchus* due to delayed egg hatching in spring.

Comparative phenologies of caddisfly and beetle development

We assessed the rate and timing of *Asynarchus* development from weekly samplings of four autumnal basins (ponds 6, 8, 10, 11), one permanent basin (9), and five vernal ponds (7, 13, 15, 21, 22) beginning a week after snow melt (15 June 2006) until pupation ended (17 July 2006). The vernal pools selected were known to have annual populations of *Asynarchus* that are regularly constrained by drying time (Wissinger et al. 1999*a*, 2003). The permanent pond (9) typically has few salamanders as a result of winterkill, and therefore supports an invertebrate community similar to that in autumnal basins. Each pond was sampled on each date by taking four nondestructive, quantitative, 1-m sweeps with a 30 cm wide D-net (1-mm mesh). Sampling was restricted to littoral vegetation and benthos adjacent to pond edges where caddisfly densities were highest. Larvae were sorted into instars in the field, with voucher specimens collected to confirm field-identified instar categories (body mass and instar metrics are given in Wissinger et al. 2003). Pupae attached to the substrate were counted in the sample area before sweeps were taken. Emergence periods for each pond were estimated by repeatedly (every 2–3 days) counting and removing pupal exuviae that accumulated on emergent boulders placed near the edge of the ponds. In autumnal ponds, *Dytiscus* phenology was inferred from 10 0.33-m^2 quantitative sweeps taken when the first-instar larvae appeared on 22 June, and continuing weekly until beetle pupation began on 24 July 2006. The three larval instars of the beetles were easily identified in the field.

We tested differences in the development rate of caddisfly larvae between five vernal and five long-duration basins (i.e., four autumnal ponds and pond 9) with repeated-measures ANOVA on mean larval stage. Permanence was tested between ponds, and date and permanence × date interaction were tested within ponds. Mean larval stage was calculated by assigning each individual from a sample to categories 1–7, with 1–5 assigned to the five larval instars, and prepupae and pupae assigned 6 and 7, respectively. Caddisfly prepupae were late fifth instars that had added stones to their detrital cases in preparation for pupation but had yet to seal the ends of their cases and affix them to the substrate (Wissinger et al. 2003). Difference in mean caddisfly emergence date (in Julian days) between vernal and autumnal ponds was analyzed using a t test with pond means as replicates.

Size predation experiment with beetles and Asynarchus *caddisflies*

To determine how the relative sizes of beetles and caddisflies affected predation rates, we conducted survival trials by manipulating different size combinations of the two species. Experiments were conducted in plastic microcosms (W × L × D = 39 × 54 × 11 cm) to which detritus (1000 cm^3) (without caddisflies and beetles), and a large cobble (long axis ~20 cm) were added to mimic pond substrate. Microcosms were housed in a portable field shelter. We added 15 fourth-instar, fifth-instar, or prepupal (one-half to three-fourths of the case covered in stones) *Asynarchus* caddisfly larvae to tanks 30 min prior to the addition of one first-, second-, or third-instar *Dytiscus* larva that had been starved for 20 h. We also included a no-beetle control treatment for each caddisfly stage. Caddisfly synchrony within ponds and the natural time lag of beetle phenologies necessitated collecting caddisflies from ponds at relatively high elevations (3400–3800 m) at our study site, and beetle larvae from a pond at a lower elevation (2900 m). We ran two replicates of each of the 12 treatments over two days, yielding a total of four replicates of each treatment. Experiments began in mid-afternoon and continued for 19 h overnight, with survival being assessed the following morning.

Mortality rates were corrected for cannibalism by subtracting mean mortality in control tanks from that in beetle tanks. We tested the effect of beetle instar and caddisfly stage on untransformed mean caddisfly mortality using two-way ANOVA with time as a randomized block. Initial exploration of block interactions for caddisflies and beetles revealed no significant interactions, so the final model included only the block main effect. Scheffé's post hoc comparisons were used to compare treatment means.

Nonconsumptive effects of beetle predators on caddisflies

To investigate the degree of flexibility in behavioral and developmental responses of *Asynarchus* larvae to *Dytiscus* larvae, we reared caddisfly larvae in large plastic wading pools (1.5 m^2) with and without beetles. Tanks contained snowmelt water maintained to a level of 15 cm, 3000 cm^3 of detritus collected from a beetle-free experimental mesocosm, and two large rocks added for pupation substrate. We collected third-instar caddisfly larvae from an autumnal pond (pond 55; see Wissinger et al. 2003) before beetles appeared, and we added 40 of the caddisflies to each mesocosm on 21 June 2006. The next day one third-instar beetle larva housed inside a perforated floating container (two-way guppy breeder; Lee's Aquarium and Pet Products, San Marcos, California, USA; L × W × D = 14 × 8 × 8 cm) with a twig and several sedge leaves, was added to half of the 14 tanks. Identical floating containers without beetles were

added to the control treatments. Beetles were fed *Asynarchus* larvae introduced at a rate of 12 individuals every two days for the duration of the experiment. Tanks were checked every 2–3 days for pupae, and when found, pupae were transferred to individual emergence chambers (Wissinger et al. 2004) floating in a wading pool nearby. Emerged adults were hand-collected and frozen. Their sex was determined, right-wing length measured (±0.05 mm) using an ocular micrometer, and thorax (with legs and head attached), abdomen, and wing mass were determined (±0.001 mg) after drying at 60°C for 24 h by weighing on a Cahn C-31 microbalance (Cahn, Cerritos, California, USA).

Twice during the experiment (12 and 18 July), we conducted 5-min focal animal observations of fifth-instar larvae during mid-morning to assess foraging behavior in the presence and absence of predator cues. We recorded time spent moving and the number of encounters with conspecifics. Seven observers worked simultaneously in different tanks, alternating observations between treatments until two caddisfly larvae in each tank had been observed. Mean time spent moving and number of encounters were analyzed with randomized-block ANOVA and Poisson GLMs, respectively. No observer bias or block × predator interaction was found; thus, final models included the block main effect only. Mean emergence date and adult size metrics (total mass, thorax mass, and forewing length) were analyzed by two-way ANOVA and MANOVA, respectively, with tank means (for both male and females) as replicates.

Because *Asynarchus* larvae may be unable to detect nonmechanical predator cues, we conducted additional behavioral trials with unconstrained *Dytiscus* beetles in microcosms identical to those used in the size–predation experiments. We introduced 10 fifth-instar caddisfly larvae collected from an autumnal pond (pond 8) to each of the 12 microcosms 30 min prior to the introduction of one third-instar beetle to half of the tanks. Caddisflies were acclimated for 30 min before trials began. Two observers worked simultaneously on predator and control tanks. Focal-animal sampling followed the same protocol as before, but with 10-min samples. One focal animal was observed in each tank during each trial, yielding six independent replicates of each treatment.

Cannibalism and predation experiments with caddisflies

We knew that *Asynarchus* larvae engaged in cannibalism and intraguild predation (IGP) on *Limnephilus externus* (Wissinger et al. 1996), but how the proclivity for these interactions varied as a function of relative size was not known. We assessed size-specific cannibalism rates by manipulating instar combinations of *Asynarchus* in microcosms (30 ×16 cm plastic storage containers containing 2.5 cm of spring water with detritus placed on insect screen cut to fit the base of the chambers) housed in a portable field shelter at the Rocky Mountain Biological Laboratory (Crested Butte, Colorado, USA). We added 10 prey individuals to each container and allowed them to acclimate for 30 min before we introduced 10 potential cannibals. Six replicates of the 10 pairwise size combinations of second- through fifth-instar larvae were randomly allocated to containers. We collected animals from populations at different elevations to obtain the various size combinations. In a second experiment, we manipulated instar combinations of *Asynarchus* and *Limnephilus* in microcosms to investigate size-specific predation rates throughout *Asynarchus* larval development. The experimental venue and protocol were the same as in the cannibalism experiment. For both experiments, data were analyzed using nonlinear regression with treatment means as replicates.

Results

Comparative life histories in vernal and autumnal ponds

Asynarchus developed rapidly and synchronously in vernal ponds, progressing from second-instar larvae to adults in less than 50 days during spring and early summer (Fig. 1). On average, only 21% ± 2% (pond mean ± SE) of larvae were larger or smaller than the modal instar on each sampling date. Despite this rapid development, emergence in three of the four ponds was constrained by pond drying. Inspection of pupae remaining in ponds after drying revealed that 21.0% ± 0.06% (pond mean ± SE) of individuals that reached the pupal stage perished from desiccation before emergence occurred. Although autumnal ponds dried two months later than vernal pools, caddisfly larval development was actually faster in them than in vernal ponds (Fig. 1; split-plot ANOVA: pond effect, $F_{1,8} = 26.93$, $P < 0.001$), with a divergence in growth over time (date × pond interaction, $F_{3,24} = 3.09$, $P = 0.046$) resulting in an earlier transition to prepupal and pupal stages in autumnal ponds (Fig. 1). However, mean emergence date did not differ significantly between autumnal and vernal ponds ($t_9 = 0.225$, $P = 0.83$). *Dytiscus* larvae were observed only in the autumnal ponds, where their developmental phenology lagged behind that of caddisflies (Fig. 1). Much of caddisfly larval development in autumnal ponds occurred before beetles had reached the second larval instar. By this time, the caddisflies were adding stones to their cases in the prepupal phase, and then pupated and emerged before beetles developed into large, final (third) larval instars.

Size-specific vulnerability of caddisflies to beetle predation

Predation by *Dytiscus* larvae on *Asynarchus* increased dramatically with predator size (Fig. 2; beetle effect, $F_{2,26} = 95.11$, $P < 0.001$), and varied with caddisfly stage (caddisfly effect: $F_{2,26} = 25.73$, $P < 0.001$; caddisfly × beetle interaction: $F_{4,26} = 12.69$, $P = 0.024$). Predation by first-instar beetle larvae was negligible, but second-instar beetles attacked and killed fourth- and fifth-instar caddisfly larvae (Fig. 2). Caddisfly larvae of all sizes were most vulnerable to third-instar beetles, and fourth- and

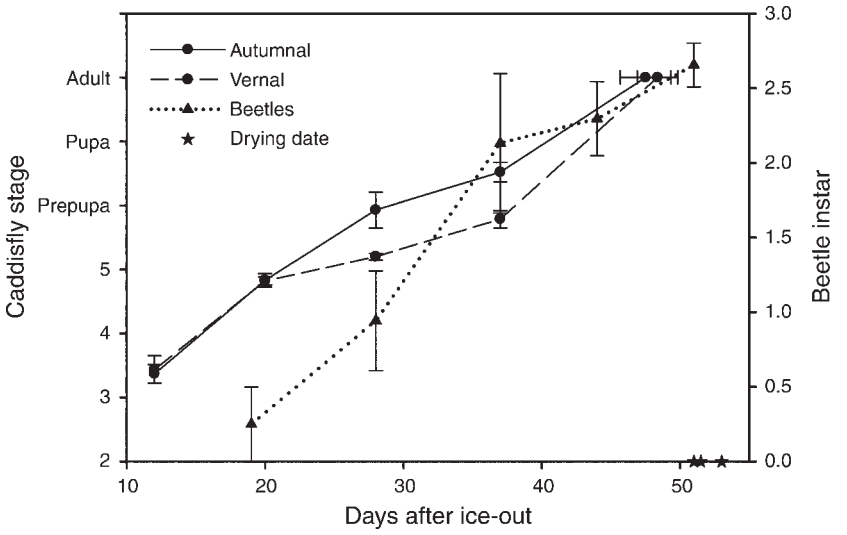

FIG. 1. The developmental phenology of the caddisfly *Asynarchus nigriculus* in vernal (wet only in spring) and autumnal (dry late in summer) ponds and of larvae of the beetle *Dytiscus dauricus*, which are only present in autumnal ponds. Caddisfly stages are second through fifth instar, prepupa, pupa, and adult. Prepupae were defined as fifth-instar larvae that have begun to add stones to their cases in preparation for pupation. The drying date of the vernal pools studied is shown on the *x*-axis line (stars). Ice-out occurred on 3 June 2006. Error bars represent ±SE, with pond means as replicates. Horizontal error bars indicate ±SE of mean adult emergence date.

fifth-instar larvae had significantly higher mortality than prepupae (Fig. 2).

Nonconsumptive effects of beetle predators on caddisflies

Male *Asynarchus* emerged significantly earlier than females across all treatments (sex effect: $F_{1,24} = 73.25$, $P < 0.0001$; Fig. 3), and the presence of beetle larvae had no effect on time to emergence of either sex (beetle effect: $F_{1,24} = 0.051$, $P = 0.82$; beetle × sex: $F_{1,24} = 0.003$, $P = 0.95$; Fig. 3). Emerging males had longer wings and lower total mass and thoracic mass than did females (univariate ANOVAs on significant MANOVA: $P < 0.0001$; Appendix B), However, beetle presence had no effect on these size metrics for either sex (MANOVA: $P > 0.78$; Appendix B).

During observation trials, the activity rates of late-instar *Asynarchus* (Fig. 4A; randomized-block ANOVA, beetle effect: $F = 0.033$, $P = 0.857$) and number of encounters between larvae (with beetle: 0.43 ± 0.14 encounters, mean ± SE; control (no beetle): 0.14 ± 0.10; Poisson GLM, $P = 0.095$), did not differ significantly

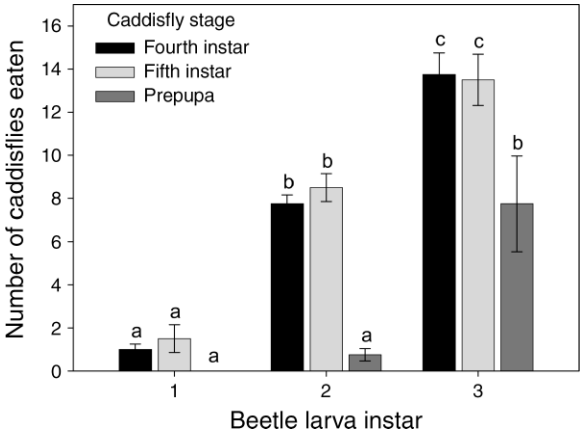

FIG. 2. Per oapita predation by three instars of *Dytiscus* on three developmental stages of *Asynarchus* in a 19-h microcosm experiment. Different lowercase letters indicate significant differences ($P < 0.05$) based on Scheffé's post hoc comparisons. Predation rates (number of caddisflies eaten; mean ± SE) are adjusted for stage-specific cannibalism rates in *Dytiscus*-free treatments (mean ± SE: fourth instar, 0.25 ± 0.25; fifth instar, 0; prepupa, 0.75 ± 0.25).

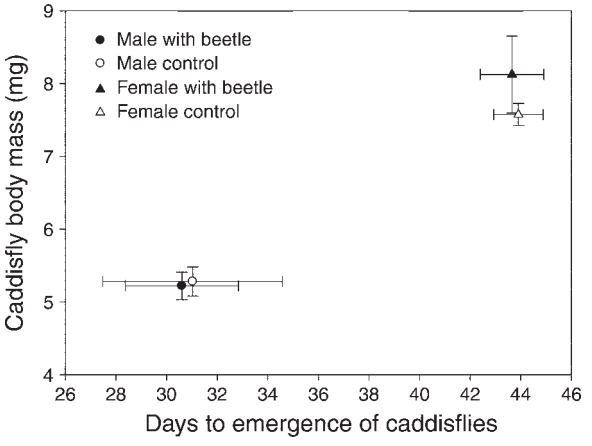

FIG. 3. Mean adult body mass and mean time of emergence for male and female *Asynarchus* reared in mesocosms with and without (control) a caged third-instar *Dytiscus* beetle larva as a non-consumptive predator. Error bars indicate ±SE. Days until emergence were calculated from the beginning of the experiment, when early third-instar larvae were added to the tanks.

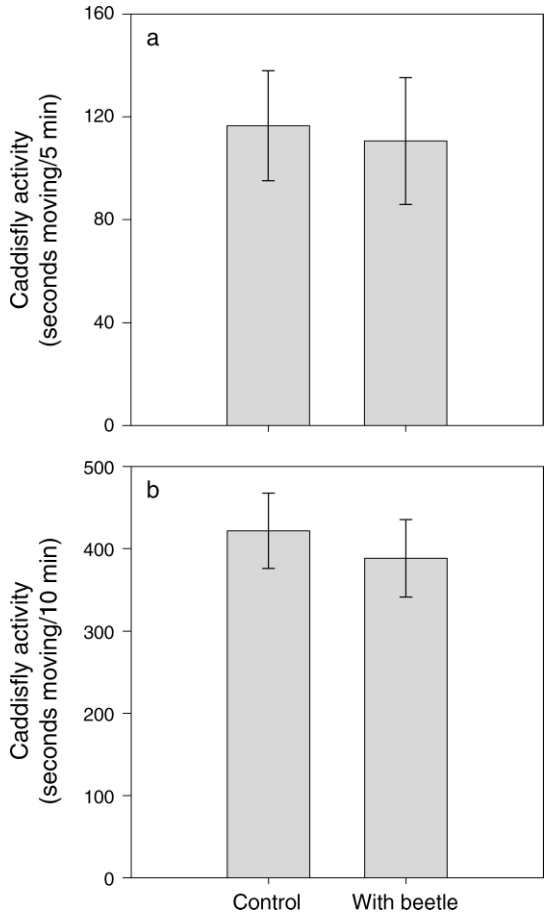

FIG. 4. Activity rate (mean ± SE) of *Asynarchus* fifth-instar larvae exposed to (A) nonconsumptive predator cues from caged *Dytiscus* in mesocosms (activity rate = seconds of movement per 5-min focal animal study) and (B) predator cues from uncaged, unmanipulated *Dytiscus* (activity rate = seconds of movement per 10-min focal animal study) in microcosms.

between treatments with and without caged beetles. There was no difference in caddisfly activity level (Fig. 4B; $t_5 = 0.317$, $P = 0.757$) or the number of encounters between individuals (with beetle: 1.5 ± 0.43 encounters, mean ± SE; control: 1.0 ± 0.37 encounters; $\chi^2 = 0.60$, $P = 0.439$) when exposed to the full suite of potential cues from unconstrained beetle larvae (chemical, visual, acoustic, and tactile). Moreover, in the trials with unconstrained beetles, caddisflies were regularly observed to collide with feeding beetles or even to crawl between the open mandibles of the predators. In summary, we found no evidence for plasticity in caddisfly development or for risk-sensitive activity levels or foraging behaviors in response to the presence of beetles.

Size-specific cannibalism and intraguild predation among caddisflies

Cannibalism increased with size disparity between cannibals and victims (Fig. 5A). Cannibalism rates of 15% occurred between individuals of the same size due to "mob" cannibalism (Wissinger et al. 2004), and increased dramatically to a peak of 100% mortality of second-instar larvae when exposed to fifth-instar cannibals (Fig. 5A). Observations made during the trials revealed that one-on-one cannibalism (as opposed to mob cannibalism) became increasingly common with the proportional size difference between cannibal and victim.

Intraguild predation of *Asynarchus* on *Limnephilus* was negligible when the proportional size differences of potential prey and predators were less than 0.5 (Fig. 5B). The size differences on the lower asymptote of Fig. 5B corresponded to combinations of larvae that were the same instar, or differed by one instar (fifth on fourth, fourth on third, third on second instars). Above that lower threshold, IGP increased significantly with proportional difference in size (Fig. 5B). IGP was highest for fifth-instar *Asynarchus* preying on second-instar Limnephilus, and intermediate when *Asynarchus* were

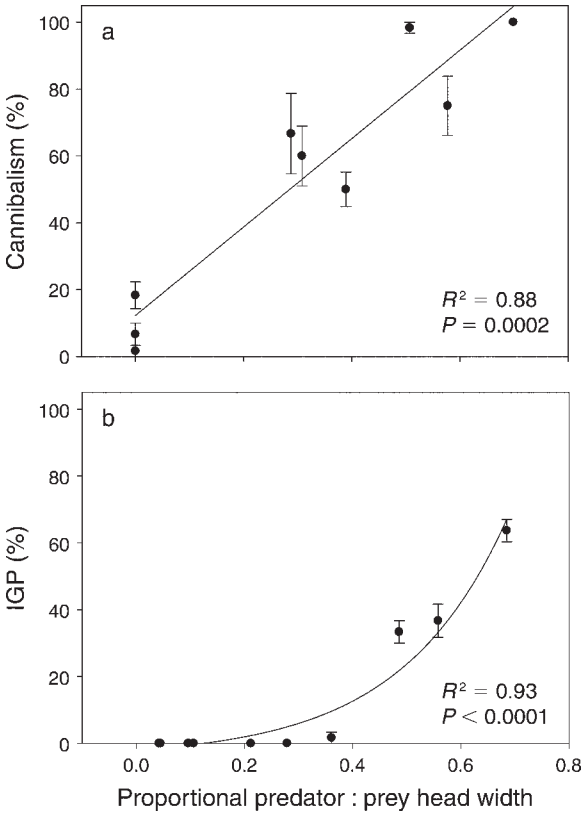

FIG. 5. Size-specific *Asynarchus* (A) cannibalism and (B) intraguild predation (IGP) on *Limnephilus externus* in microcosm experiments. Cannibalism is the percentage of *Asynarchus* individuals eaten by other *Asynarchus*, and IGP is the percentage of *Limnephilus* individuals eaten by *Asynarchus*. Error bars indicate ±SE, $n = 6$ independent replicates of each treatment; R^2 and P values are from (A) linear and (B) nonlinear regressions, respectively. Regression equations are: (A) $y = 12.31 + 132x$; (B) $y = 1.02 + 451^x$.

two instars larger (fifth on third and fourth on second) than *Limnephilus* (Fig. 5B).

Discussion

Convergent, reinforcing time constraints across habitat types

The results of our experiments, combined with comparative data from natural populations, provide evidence for a suite of reinforcing selective pressures that simultaneously operate on *Asynarchus* across different types of temporary habitats. Rapid development facilitated by minimal investment in antipredator defense and dietary supplementation through cannibalism and intraguild predation enables *Asynarchus* to complete development in short-duration vernal pools (Wissinger et al. 2004, 2006). In this study we found that although many larvae in vernal pools complete development before drying, slowly growing individuals die from desiccation, indicating that there is ongoing selection for traits that lead to rapid development in those populations.

Not surprisingly, these fast lifestyle traits increase vulnerability to predators (Wissinger et al. 1999*b*, 2006), including *Dytiscus* beetles in autumnal ponds, which were voracious predators on *Asynarchus* in our experiments. We found that *Asynarchus* development in these ponds was as fast or faster in than in vernal pools (Fig. 1) and field experiments with *Asynarchus* and *Dytiscus* beetle larvae suggest that this rapid development in autumnal ponds is driven by several biotic, rather than abiotic, time constraints. Comparing the phenologies of beetles and caddisflies in the ponds to the results of size- and stage-specific rates of predation in microcosm experiments illustrates that even a short time lag (2–7 days) in caddisfly development relative to that of the beetles would substantially increase larval mortality in autumnal ponds (Fig. 2). Nearly all individuals of this caddisfly species emerge from autumnal ponds just before the beetles reach the size at which they become effective predators, and most caddisflies that have not emerged by that time pupate in stone cases that reduce beetle predation (Wissinger et al. 2006).

In addition to minimizing beetle predation, rapid development in long-duration habitats facilitates *Asynarchus* intraguild predation on other species of caddisflies. Early hatching and rapid development of *Asynarchus* leads to a one- to two-instar size advantage over *Limnephilus* spp. (Wissinger et al. 2003). Experimental data confirm that this developmental size advantage facilitates intraguild predation, IGP (also see Wissinger et al. 1996), and previous experiments document the fitness benefits by providing a protein supplement to a predominately detrital diet and eliminating potential competitors (Wissinger et al. 1996, 2004). Maintaining a two-instar size advantage is crucial for *Asynarchus* IGP; i.e., there is a sharp decline in predation on *Limnephilus* spp. between a two-instar and one-instar size difference. This type of developmental priority effect for facilitating asymmetrical interference competition and IGP is similar to that in other guilds of competing predators (Wissinger 1992, Padeffke and Suhling 2003).

Cannibalism is likely to exert a third biotic selection pressure on *Asynarchus* development. Cannibalism occurs at relatively low rates among same-sized *Asynarchus* via mob attacks on vulnerable individuals (e.g., those with damaged cases; Wissinger et al. 2006). The results of our experiments reveal that vulnerability to cannibalism increases dramatically with size disparities among conspecifics, as a result of both mob and one-on-one attacks. A two-instar difference in size resulted in an almost 10-fold increase in cannibalism compared to that among same-sized larvae. Similar thresholds for cannibalism based on relative size have been observed in many species (Polis 1981, Wissinger et al. 2010), including other aquatic insects (Wissinger 1992). Theory predicts that when such size thresholds for cannibalism occur within cohorts, there should be selection for the type of rapid and synchronous development that we observed (Crowley and Hopper 1994). We do not know whether *Asynarchus* cannibalism rates differ between habitats across the permanence gradient as a result of difference in food quality (e.g., Inkley et al. 2008), but we do know that they are not density dependent (Wissinger et al. 1996) or affected by experimental drying (S. A. Wissinger and H. S. Greig, *unpublished data*). Regardless, it is clear that cannibalism has the potential to be an important selective force in autumnal ponds (~20% mortality; Wissinger et al. 1996).

In summary, despite the relaxed time constraints associated with drying in autumnal habitats, there are at least three potential selection pressures for rapid development in *Asynarchus*: decreased vulnerability to beetle predation, a size advantage that facilitates IGP on *Limnephilus*, and reduced vulnerability to cannibalism. Additional experiments will be needed to determine whether these effects interact; i.e., slow growth early in development that leads to decreased protein supplementation from IGP could increase subsequent vulnerability to beetle predation.

Adaptive and nonadaptive explanations for life history traits

One implication of reinforcing biotic and drying time constraints in autumnal and vernal habitats, respectively, is that it appears to have led to the evolution of fast lifestyle traits that are not flexible to predation risk. We found no evidence for plasticity in antipredator behaviors or activity rates of *Asynarchus* in response to the presence or absence of beetles. Despite their vulnerability to beetle predation, there were no differences in development rate or size at emergence between caddisflies reared with and without beetles (Fig. 4), and caddisflies developed and emerged at the same rate in beetle-free autumnal ponds as they did in ponds with beetles (H. S. Greig and S. A. Wissinger, *unpublished*



PLATE 1. (Top) A final-instar *Asynarchus nigriculus* caddisfly larva with a case built from small fragments of detritus. (Bottom) Cannibalism among same-sized *Asynarchus* larvae occurs through mob attacks on injured individuals. Photo credits: Angus McIntosh.

data). These results are consistent with previous work showing that *Asynarchus* larvae from other populations do not modify their behavior, morphology, or activity levels in response to the presence of predatory salamanders (Wissinger et al. 1999*b*, 2006).

The selection pressures for rapid development in both vernal and autumnal habitats are likely to be predictable between years. Over the past 20 years, the sequence of drying among ponds and the lengths of the wet phase during summer have been remarkably constant (Wissinger et al. 1999*a*). Although the densities of beetle predators fluctuate from year to year, they are always present in the same ponds each year, and the phenologies of beetles and *Limnephilus* respond similarly to interannual variation in snow melt (Wissinger et al. 2003). Given the predictability and convergence of selection pressures across habitats, it is perhaps not surprising that fixed developmental strategies have evolved in the landscape context of adjacent vernal and autumnal ponds. This is consistent with both theoretical and empirical results suggesting that highly predictable environments should lead to simple adaptive responses (Hairston 1987) and fixed phenotypes (Levins 1968, Tufto 2000, Alpert and Simms 2002).

There are several non-mutually exclusive alternative hypotheses that could also explain the fixed life history and antipredator traits that we observed in *Asynarchus*. Firstly, the development of plastic antipredator traits could be constrained by historical factors, such as phylogenetic inertia or founder effects in isolated populations that impair the ability of this species to adapt to novel, current selection pressures (Sih et al. 2000, Caudill and Peckarsky 2003). We have no reason to suspect that any of the taxa involved (caddisflies, beetles, salamanders) have recently invaded these habitats, making phylogenetic inertia unlikely. Further-

more, co-occurrence patterns at our main study site are representative of those observed across dozens of other study sites in the region (distance between sites > 10 km; Wissinger et al. 2003); thus, the patterns that we observed probably are not a result of unique founder genotypes. Finally, phenotypic plasticity to both predators and drying has been observed in other limnephilid species (e.g., Otto and Svensson 1980, Nislow and Molles 1993) suggesting that phylogenetic constraints on the plasticity of these traits are not characteristic of this group of taxa on the whole.

Secondly, the spatial proximity of ponds at the Mexican Cut, combined with high adult dispersal rates, could lead to indiscriminate gene flow between populations across pond types (De Block et al. 2005) that constrains local adaptation (Lenormand 2002), thus providing an alternative explanation for the absence of risk-sensitive foraging. However, asymmetrical gene flow from temporary ponds to autumnal ponds that could constrain the evolution of antipredator responses to *Dytiscus* (as in Storfer and Sih 1998, Caudill and Peckarsky 2003), is unlikely in our system. *Asynarchus* occur in densities 2–3 times higher in autumnal ponds than vernal ponds (Wissinger et al. 1996, 2003), and autumnal ponds are, on average, three times larger than vernal pools (Wissinger et al. 1999*a*). Therefore, if anything, gene flow may be biased from autumnal to vernal habitats.

Thirdly, ontogenetic or interpopulation differences in antipredator plasticity (e.g., Relyea 2002, Brodin et al. 2006) could explain the fixed traits we observed. Although we used multiple *Asynarchus* populations in our study, each experiment was conducted with a single population, and only fifth-instar individuals were observed in behavioral trials. Nevertheless gene flow between the adjacent ponds at the Mexican Cut should minimize interpopulation differences in antipredator traits. The two populations used in experiments (pond 8 and 55) were on different topographical benches within the Mexican Cut Reserve (3400 and 3450 m above sea level, respectively) and are among the most likely to exhibit interpopulation differences at our study site, yet they both exhibited fixed traits. Similarly, ontogenetic shifts are an unlikely explanation for not observing plasticity. A comparison of the relative phenologies of *Asynarchus* and *Dytiscus* indicate that *Asynarchus* larvae that are smaller than fifth instar are rarely, if ever exposed to beetle predation (Fig. 1).

Seasonal variation in biotic interactions and the persistence of habitat generalists

The life histories of many organisms, especially those with complex life cycles, are influenced by time constraints imposed by seasonal environments (Rowe and Ludwig 1991). Most studies have focused on abiotic factors, such as drying and flooding in freshwater habitats (Leips et al. 2000, Lytle and Poff 2004) and the onset of winter in terrestrial environs (Taylor 1980, Berner and Blanckenhorn 2006). In the autumnal ponds at our study site, constraints of pond drying are relatively weak and selection for rapid development is replaced by biotic time constraints. The biotic time constraints that we observed for *Asynarchus* are similar to those of copepods (*Diaptomus sanguineus*), which diapause before fish become active in spring (Hairston 1987). In both cases, it is the relative phenology of prey–predator development and activity that creates a time-specific change in life history events, driven by the seasonal onset of the threat of predation and availability of prey.

Biotic time constraints can also arise through seasonal variation in competition and herbivory, and are likely to occur in a broad range of habitats where environmental fluctuations produce phenological differences among interacting species. For example, understory herbs rapidly vegetate, flower, and set fruit soon after snow melt, but before competition for light occurs when the canopy closes in deciduous forests (Kudo et al. 2008). Similarly, rapidly developing desert annuals can coexist with strong competitors by exploiting temporal windows of weak competition after seasonal rainfall events (Chesson and Huntly 1997). In both cases, biological time constraints in less disturbed or long-duration environments should be similar to those imposed by abiotic constraints in frequently disturbed or short-duration environments. This potentially trumps the trade-off between disturbance and biotic interactions, as the fast developer–poor competitor/predator-avoider niche is available in adjacent, contrasting habitats. Under this scenario, which we observed in our study, rapidly developing species with fixed traits can persist across a range of habitat types, provided dormant stages or ontogenetic habitat shifts maintain the species persistence during unfavorable conditions (Caceres 1997, Chesson and Huntly 1997).

McPeek (1996) argued that understanding the trade-offs that organisms experience in contrasting habitats is key to understanding how some species maintain broad distributions. He found that generalists and specialists can coexist if convergent niche space across two habitat types eliminates evolutionary trade-offs. Our results indicate that this niche space can be generated across habitats by seasonally variable species interactions that result in parallel biotic and abiotic selection. We do not know of other comparable studies and propose this as a novel mechanism for the persistence of habitat generalists across environmental gradients. Furthermore, considering changes in the structure of food webs over time, as well as across habitats, provides additional understanding of species distributions across environmental gradients; especially in habitats with pronounced seasonality.

Conclusions

Many studies have documented the behavioral, developmental, physiological, and morphological phe-

notypic plasticity that prey exhibit in predator-free and predator-present habitats (Lima 1998, Tollrian and Harvell 1999). Such flexibility facilitates broad distributions across environmental gradients with contrasting selection pressures. This study was motivated by the puzzling absence of flexibility in *Asynarchus* to the presence or absence of drying time constraints and to predation risk by both vertebrate and invertebrate predators. Our results indicate that multiple biotic time constraints in autumnal habitats (reduced beetle predation, reduced cannibalism, increased opportunity for intraguild predation) reinforce, rather than trade off with traits that facilitate the timely completion of development in habitats that dry out. This result emphasizes that seasonal shifts in food web interactions can create a temporal niche for a species with fixed traits that has a selective advantage in an alternative habitat type for different reasons.

Coexistence mediated by the phenological relationships between a species and their predators, competitors, or mutualists is likely to have a widespread influence on species distributions, especially in habitats with pronounced seasonality (Caceres 1997, Chesson and Huntly 1997). However, both phenological relationships between species and seasonal changes in physical habitat (i.e., drying) are vulnerable to anthropogenic influences such as climate change (Memmott et al. 2007). Thus, an understanding of how phenological relationships affect species distributions will be necessary to predict the effects of human-induced changes in climate on species distributions across heterogeneous landscapes.

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APPENDIX A

Photographs of a representative vernal, autumnal, and permanent pond from the Mexican Cut Nature Preserve, Elk Mountains, Colorado, USA (*Ecological Archives* E091-060-A1).

APPENDIX B

A table of results from MANOVA and univariate ANOVAs testing the effect of nonconsumptive *Dytiscus* beetle presence and caddisfly sex on size at emergence of *Asynarchus* caddisflies (*Ecological Archives* E091-060-A2).