Swimming performance trade-offs across a gradient in community composition in Trinidadian killifish (*Rivulus hartii*)

CHRISTOPHER E. OUFIERO,[1] MATTHEW R. WALSH, DAVID N. REZNICK, AND THEODORE GARLAND, JR.

Department of Biology, University of California, Riverside, California 92521 USA

Abstract. The impacts of predation and competition on life history, behavioral, and morphological traits are well established for many organisms, but effects on locomotor performance have received relatively little attention. We examined variation in sprint speed and critical swimming speed (U_{crit}, a measure of stamina) in the Trinidadian killifish (*Rivulus hartii*) across a gradient of ecological communities. *R. hartii* are located in (1) "high-predation" sites with large, piscine piscivores, (2) "*Rivulus*–guppy" sites with guppies, and (3) "*Rivulus*-only" sites with only *R. hartii*. *R. hartii* suffer higher mortality in high-predation sites. In *Rivulus*–guppy sites, population densities are reduced and growth rates increased compared with *Rivulus*-only sites, which likely represent indirect effects of guppy predation on young *R. hartii*. We show a significant negative relationship, suggesting a trade-off, between sprint speed and endurance; *Rivulus* from high-predation sites were faster sprinters but had reduced critical swimming speeds. This trade-off was also apparent in correlations of the nine population means. At the individual level, the correlation was weaker and only significantly negative when all nine populations (three from each site) were pooled and values were not corrected for body size. Sex had a significant effect on U_{crit}, with females having a lower U_{crit}, but sexes did not differ in sprint speed. Fish from high-predation sites also exhibited increased tail lengths and fineness ratios compared to sites without large predators. The two low-predation sites showed no statistical differences in locomotor performance or morphology.

Key words: critical swimming speed; predators; sprint speed; swimming performance; trade-offs.

INTRODUCTION

Multiple interactions within ecological communities, including competition for resources, territories, and mates—as well as predation—can impose strong selection on organisms. The effects of predators are particularly well documented, as they lead to phenotypic and genetic changes in a wide array of traits at the whole- and sub-organismal level (for reviews, see Havel 1987, Dodson 1989, Harvell 1990, Jürgens and Matz 2002, Langerhans 2006). In some cases, these changes likely improve Darwinian fitness. For example, Trinidadian guppies (*Poecilia reticulata*) from stream communities with relatively high predation intensity have evolved differences in coloration (Houde 1997, Magurran 2005), life histories (Reznick and Endler 1982, Reznick et al. 1990, Reznick and Bryga 1996), and morphology (Hendry et al. 2006) as compared to communities with relatively low predation. Locomotor performance is also tightly connected to aspects of Darwinian fitness and is thus likely to respond to changes in community composition, especially predation (Djawdan and Garland 1988, Irschick and Garland 2001, Oufiero and Garland 2007, Irschick et al. 2008,

Langerhans and Reznick 2009). However, the evolution of locomotor performance has received far less attention than life history or behavioral traits.

In environments with increased predation intensity, the direct consumption of prey by predators should lead to natural selection that favors increased escape abilities, potentially through an increase in burst or sprint performance (Irschick and Garland 2001, Oufiero and Garland 2007, Irschick et al. 2008). Such a result has recently been demonstrated from common garden studies of guppies and species of *Gambusia* (Ghalambor et al. 2004, Langerhans 2006, 2009). Conversely, as locomotor performance is also involved in finding food and mates (Garland and Losos 1994), species or populations inhabiting communities without predators may be selected for enhanced endurance capacities (see also Feder et al. 2010). However, any potential increase in escape or endurance ability may come at a cost. In particular, performance functions may trade-off with one another because of biomechanical, biochemical, or physiological interrelationships (Clobert et al. 2000, Vanhooydonck et al. 2001, Blake 2004, Dlugosz et al. 2009; but see Bennett et al. 1989). Consequently, populations from predator-rich communities may exhibit diminished endurance performance, and vice versa for populations from communities that lack predators.

The purpose of the present study was to compare the locomotor performance of Trinidadian killifish (*Rivulus*

hartii) from streams with varying predation intensity, and hence to evaluate trade-offs in locomotor function at the population and individual level. We compared the sprint speed and critical swimming speed (U_{crit}) of fish from sites with large piscivorous predators (e.g., *Crenicichla alta*), termed high predation (HP), to those with relatively weak predation. However, because *R. hartii* exhibits greater dispersal capabilities than other fish species in these streams (Gilliam et al. 1993, Fraser et al. 1999, Walsh and Reznick 2008), they are able to occupy sites with no other fish species present. Therefore, we extended our comparison to two low-predation sites that vary in community assemblage; *Rivulus*-only (R) sites, where *R. hartii* is the only fish species present, and *Rivulus*–guppy sites (RG), where *R. hartii* occurs with only *P. reticulata*. The two low-predation sites have the potential to differ in locomotor performance due to differences in field growth rate (Walsh 2009) that could result in trade-offs with locomotor abilities in general (Clobert et al. 2000, Billerbeck et al. 2001, Girard et al. 2002, Feder et al. 2010). These communities lack physical separation, but are distinct due to barrier waterfalls that truncate the distribution of some species (e.g., such predators as *C. alta*) but not others (e.g., *R. hartii*, *P. reticulata*). As a result, these communities contain similar physical habitats and do not differ in such environmental characteristics as water temperature or dissolved oxygen (Walsh and Reznick 2009).

We predicted that fish from HP sites would exhibit a greater sprint speed and a lower U_{crit} compared to the two low-predation sites (R and RG). *R. hartii* from HP localities suffer a 2× higher mortality rate per 60 days compared with *Rivulus*-only localities (J. F. Gilliam and D. F. Fraser, *personal communication*); therefore, if this increased mortality is at least partly related to higher predation intensity, then it should lead to natural selection favoring increased escape ability, as compared with the communities that lack large piscivores (RG and R localities). We also predicted divergence in locomotor performance between the RG sites compared to R sites. Population densities are reduced in RG sites and field growth rates are increased (>3× greater in RG sites; Walsh 2009), as compared with *Rivulus*-only sites, likely because guppies prey upon very young *R. hartii*. This increase in growth may trade-off with locomotor abilities, resulting in decreased locomotor performance of fish from the RG sites (Billerbeck et al. 2001). However, because *P. reticulata* prey upon young *R. hartii*, there might be an increase in locomotor performance in RG sites due to selection favoring enhanced escape performance, which may be maintained throughout their life. We also compared body size among the sites, and tested for differences in traits that may be important for sprinting. Specifically, we predicted that HP fish would have an increased tail length and a lower fineness ratio (defined as the standard length of the fish divided by its body depth; Webb 1975), which would serve to increase thrust production.

MATERIALS AND METHODS

Rivulus hartii ($N = 178$, ranging in size from ~54 to 69 mm in total length) were collected from all three community types from each of three independent river drainages (Aripo, Guanapo, and Quare Rivers) in the Northern Range Mountains of Trinidad (Appendix A). One of each *Rivulus* community type was sampled from these rivers (high predation [HP], *Rivulus*–guppy [RG], *Rivulus*-only [R]). Each high-predation locality specifically contained the piscivorous *Crenicichla alta* and/or *Hoplias malabaricus*. Guppies (*P. reticulata*) and *R. hartii* were the only fish species present in *Rivulus*–guppy communities, except in the Guanapo River, where the catfish *Rhamdia sebae* is also present. For each focal river, the *Rivulus*–guppy and *Rivulus*-only sites were located in tributaries to the main river (containing the high-predation community) above barrier waterfalls that prevented the upstream movement of large predatory species. Fish were captured with seines and hand nets. The fish were brought back to our research facilities in Trinidad, maintained in group tanks for 10–14 days, and fed flake food. Fish were then transported to the University of California, Riverside, separated into male/female pairs in 2.5-gallon aquaria (1 L = 0.26 gallons), and fed ad libitum for two to three months. Both adult males and females were tested (the effects of sex were tested in final analyses). Fish were fasted for at least 24 hours prior to swimming measurements to reduce the possibility that food in the gut and/or ongoing digestive processes might affect performance (e.g., Garland and Arnold 1983, Huey et al. 1984).

Sprint speed was measured using methods similar to Nelson et al. (2002), who chased fish along a "racetrack" lined with 12 sets of photocells linked to a computer that recorded maximum speed. Fish were first acclimated (15 minutes) to the entire racetrack, which measures 91 cm long × 5.5 cm wide × 15.8 cm high. The 12 pairs of photocells (LS10 light screen; Banner Engineering Corporation, Minneapolis, Minnesota, USA), each 5 cm apart, spanned the middle 60 cm of the racetrack beginning 18 cm from the starting side to allow the fish room to accelerate. Aquatic plants and pieces of clay pots were placed at one end to offer a refuge toward which the fish would sprint. After the acclimation, fish were gently ushered towards the start end of the racetrack. The fish was then startled by hitting a small fish net directly behind the fish against the bottom of the track (the net did not make contact with the fish). If a fish did not sprint all the way to the refuge, then the net was lifted from the water (to avoid triggering the photocells) and again hit against the bottom of the track behind the fish. This procedure was repeated three or four times per individual, with ~30 seconds between trials. As the fish sprinted along the track and broke the infrared light beams emitted from the photocells, the timing of the break of the beams was recorded in custom computer software (Serrace.exe; T. Garland, Jr., and P. E. Midford, *unpublished software*). The program then

calculated the maximum sprint speed of the individual. We used the single highest speed attained over 10 cm (over a total of three consecutive photocells) from any one of the three or four trials as a measure of a fish's maximum sprint speed. Sprint speed was measured at room temperature, which was recorded for each trial and was 23.62°C ± 1.53°C (mean ± SD). Temperature did not significantly affect sprinting performance, so it was not included in the final analyses.

Critical swimming speed (U_{crit}) was used as a measure of the fish's endurance capacity and was measured in the setup and procedures outlined in Oufiero and Garland (2009). Critical swimming speed is a ramped velocity test with predetermined velocity increments over a set time; and can be defined as the maximum speed a fish can attain over a set time period (Brett 1964, Beamish 1978). In short, fish were acclimated to a flow tunnel for 15 minutes at a low flow velocity (~3 cm/s) to orient them to the direction of flow. The flow tunnel holds approximately 55 L of water and is 119.5 cm long × 15.3 cm wide × 18.3 cm high. The area where the fish was tested is 12 cm long × 15.3 cm wide × 11.5 cm high. This smaller area was obtained by placing grating throughout the working area of the flow tunnel, which also served to attain laminar flow. After the 15-minute acclimation the velocity of the flow was increased 4.5 cm/s every 5 minutes until the fish could no longer maintain position in the flow and was pressed up against the back grating. A fish was considered exhausted when it could not remove itself from the grating after tapping on the sides of the tunnel and gently prodding it with a net. Critical swimming speed is expressed as a velocity (in cm/s), using the calculation in Brett (1964) and Beamish (1978). Critical swimming speed was measured at room temperature, which was recorded for each trial and was 22.89°C ± 1.38°C. Temperature did not significantly affect U_{crit}, so it was not included in the final statistical analyses.

Although the repeatability of sprint speed and U_{crit} was not tested in *R. hartii*, previous studies on two species of poeciliids in the same experimental setup and protocol revealed that both measures are significantly repeatable over various time scales (Oufiero and Garland 2009; C. E. Oufiero and T. Garland, *unpublished data*).

All fish were euthanized and measured when the swimming performance measures had been completed. We measured each fish's total length (TL, from the most anterior tip to the tip of the caudal fin), standard length (SL, from the most anterior tip to the point where the caudal fin inserts into the body), and body depth (BD, the deepest portion of the fish) to the nearest 0.01 mm using digital calipers, as well as the mass of the fish to the nearest 0.001 g on an electronic balance. After measurements, fish were preserved in 5% formalin.

We tested for site and sex effects using both absolute and relative swimming performance. For the former, we used an ANCOVA with river, site, sex, and site × sex as

fixed effects, and standard length as a covariate (results with TL, BD, or mass as a covariate yielded similar results, so for simplicity we only present results with SL as a covariate). For the latter, we used an ANOVA of body lengths (bl, from standard length) per second, again with river, site, sex, and the site × sex interaction as fixed effects. To obtain least-squares means for the 18 site × sex × river subpopulations, we added the site × sex × river interaction term to the above models.

Because river cannot be clearly defined as either a "fixed" or "random" effect (cf. Newman et al. 1997), recent studies on the *R. hartii* system have treated river as a fixed effect and not included interactions with other factors or covariates in the model (Walsh and Reznick 2010). We adopted that approach here. In the present study, the alternative approach of treating river as a random effect (while still excluding interactions with river) led to similar results for the statistical significance of the site effect. Another alternative approach is to treat site and sex as fixed effects, river as a random effect, the river × site interaction as a random effect, and standard length as a covariate. In this type of model, degrees of freedom for testing the site effect are 2 and 4 in the omnibus comparison of all three sites. In the present study, that approach led to similar results for the statistical significance of the site effects, albeit with larger *P* values, in part because of the greatly reduced degrees of freedom.

Because we had a priori predictions regarding the differences among sites, we used planned comparisons (a priori contrasts) to test for specific differences among the sites (HP vs. R and RG; R vs. RG). Analyses were performed in SAS Procedure Mixed (SAS version 9.2; SAS Institute 2008). We $\log_{10}$-transformed critical swimming speed values to improve normality of residuals, and used this for cm/s analyses, but not for bl/s analyses.

To examine the potential trade-off between performance measures, we computed the correlation between sprint speed and $\log_{10}(U_{crit})$ both among populations, using least-squares means as mentioned above, and at the individual level. We used both Pearson's correlation and Spearman's rank correlation with a two-tailed test (see Ruxton and Neuhäuser 2010). For the correlation at the individual level, we examined (1) the absolute (raw value) relationship between sprint speed and $\log_{10}(U_{crit})$, (2) the relationship between the residual values obtained from the ANCOVA model described above, and (3) the relative value (bl/s). We also examined these relationships within each population type (site: HP, RG, and R).

We evaluated differences among the sites in the various size measurements through analysis of variance (ANOVA) with a priori contrasts in SAS Procedure Mixed. We also examined two traits that may be important for sprinting abilities, using ANCOVAs with mass as a covariate. We examined the fineness ratio, an index of shape, which is calculated by SL/BD (Webb 1975). A low ratio is indicative of shapes that are short

and fat, high fineness ratios are shapes that are long and slender. Fish from HP sites should have a lower fineness ratio because they are expected to be deeper bodied to increase sprint performance. We also compared tail length (TL − SL), with the expectation that fish from HP sites will have longer tails, thereby increasing surface area and thrust. We used a priori contrasts to test the hypothesis that HP fish had a lower fineness ratio and longer tails.

RESULTS

Sprint speed.—Our results demonstrate a divergence in sprint speed between communities that differ in the presence (HP sites) vs. absence of large predators (R and RG sites), but no difference between the two low-predation sites (Fig. 1A, Table 1). *R. hartii* from high-predation sites had significantly increased sprint speeds when expressed as cm/s and as bl/s (see Table 1). High-predation fish had ~11% higher sprint speeds for cm/s and ~31% higher for bl/s. Standard length was a significant predictor of sprint speed and there was no significant effect of river, sex, or the site × sex interaction (see Table 1).

Critical swimming speed (U_{crit}).—*R. hartii* from high-predation sites had significantly reduced critical swimming speeds than their counterparts from the other two types of communities (R and RG sites) when expressed as cm/s (Fig. 1B), but not when expressed as bl/s (see Table 1). We also found no difference in U_{crit} between the two low-predation sites (Table 1). Sex had a significant effect in both models, with males tending to swim faster than females (Table 2). There was also a significant river effect when critical swimming speed was expressed as cm/s, due to the Aripo river tending to be faster (~3% increase compared to the overall mean) and the Guanapo tending to be slower (~3% decrease compared to the overall mean; Aripo, 1.2766 ± 0.019 [mean ± SE]; Guanapo, 1.2020 ± 0.017; Quare, 1.2382 ± 0.018). The site × sex interaction was significant for bl/s, reflecting a much greater difference in U_{crit} between males and females in the two low-predation sites compared to the high predation site (Table 2).

Correlation between sprint speed and critical swimming speed.—We found a significant negative correlation between sprint speed and $\log_{10}(U_{crit})$ among the nine populations for both females and males (Fig. 2). This negative relationship was also statistically significant at the individual level, but only for raw values and not for body-size-corrected values or the relative values (Fig. 3). Similar results at the individual level were obtained when the sites were analyzed separately (Appendix B); however, most correlations were nonsignificant.

Tail length, fineness ratio, and body size.—Fish from HP sites had increased tail lengths (Table 1, Fig. 4A). Our results for fineness ratio were opposite of what we predicted (Table 1, Fig. 4B): HP fish had increased fineness ratios, suggesting they have a more elongate and slim body. River did not have a significant effect on
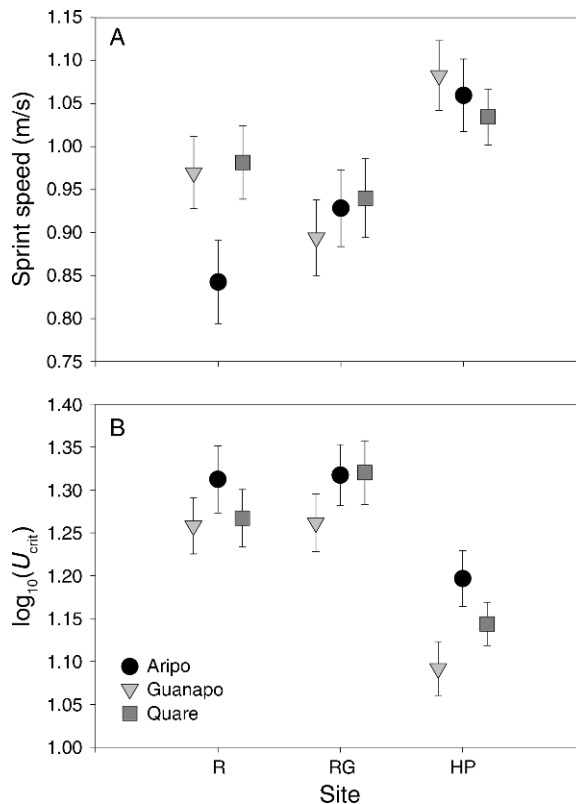

FIG. 1. (A) Sprint speed (m/s) and (B) $\log_{10}$ of critical swimming speed (U_{crit}), for the three sites (*Rivulus* only [R], *Rivulus* and guppy [RG], and high predation [HP]) in each of the three rivers. Points represent least-squares means (±SE) from an ANCOVA in SAS Procedure Mixed (see *Methods*). The effect of site was significant, with HP sites being faster sprinters, but having a lower U_{crit} compared to the two low predation sites; consistent with predictions. There was no significant difference between the low-predation sites (R and RG).

either trait, but females had shorter tails (Tables 1 and 2). Finally, we found a significant effect of site for all measures of body size; HP fish were smaller for all size measures, and males were smaller than females at the two low-predation sites (Table 2). River had a significant effect only for SL; and the site × sex interaction was significant for all size measures except BD (Appendix C). The significant site × sex interaction reflects male and female fish from the high-predation sites being similar in size, whereas females tended to be larger in the low-predation sites (Table 2).

DISCUSSION

Our results demonstrate a divergence in swimming performance between high- and low-predation sites (Fig. 1), as well as an apparent trade-off between maximum swimming speed and stamina. The trade-off is apparent at the level of population means (Fig. 2), but less so in the analysis of variation among individuals within a population. In the latter case, the results were significant

TABLE 1. Results of ANOVAs and ANCOVAs for swimming performances, tail length, and fineness ratio from SAS Procedure Mixed.

Trait	River			Site			Sex			Site × sex		
	F	df	P	F	df	P	F	df	P	F	df	P
Sprint speed (cm/s)	0.60	2, 164	0.5477	4.01	2, 164	0.0199	0.30	1, 164	0.5817	0.68	2, 164	0.5056
Sprint speed (body lengths/s)	1.19	2, 165	0.3081	39.78	2, 165	<0.0001	0.40	1, 165	0.5298	0.02	2, 165	0.9827
U_{crit} (log$_{10}$[cm/s])	4.45	2, 163	0.0131	11.06	2, 163	<0.0001	12.77	1, 163	0.0005	2.10	2, 163	0.1260
U_{crit} (body lengths/s)	3.05	2, 164	0.0501	1.12	2, 164	0.3301	23.31	1, 164	<0.0001	3.43	2, 164	0.0347
Tail length (mm)‡	1.19	2, 165	0.3068	27.29	2, 165	<0.0001	7.70	1, 165	0.0062	0.60	2, 165	0.5507
Fineness§	1.35	2, 165	0.2618	4.78	2, 165	0.0096	0.71	1, 165	0.3992	5.07	2, 165	0.0073

Note: A priori contrasts between high predation (HP) and the two-low predation sites (*Rivulus* only [R] and *Rivulus*–guppy [RG]), and between the two low-predation sites (R vs. RG), are also reported.

† Standard length was used as a covariate for sprint speed and critical swimming speed (U_{crit}); mass was used as a covariate for tail length and fineness (see *Methods*).

‡ TL – SL, where TL is total length (from the most anterior tip to the tip of the caudal fin) and SL is standard length (from the most anterior tip to the point where the caudal fin inserts into the body).

§ SL/BD, where BD is body depth (the deepest portion of the fish).

only for raw values (Fig. 3). Finally, we found no difference in the swimming abilities between *R. hartii* from communities in which they were the only fish species present (R) and where they co-occurred with guppies (RG). Although some studies have demonstrated a negative relation between sprint and endurance-swimming capability at the population level in fish (Langerhans 2006, 2009), a relationship at the individual level has not previously been observed (Reidy et al. 2000, Chappell and Odell 2004, Claireaux et al. 2007). The differences we found in swimming performance between high-predation communities and the localities that lack larger predators support the predictions that predator-induced mortality favors an increase in sprint or escape abilities, possibly at the expense of other locomotor functions.

Several sources may contribute to the differences in swimming performance between communities with and without large predators. First, overall body condition may vary, with fish in better condition having increased swimming performance (Martinez et al. 2003). However, adding an index of body condition (mass/length^3) to the ANCOVA models in Table 1 indicated that it did not statistically affect either measure of performance, and the P values for the other main effects were only slightly altered (results not shown). Moreover, a simple regres-

sion of swimming performance on condition index was not significant for sprint speed ($\beta = -0.83$, $R^2 = 0.007$, $F_{1, 171} = 1.182$, $P = 0.279$), and significantly negative for log$_{10}$(U_{crit}) ($\beta = -0.175$, $R^2 = 0.031$, $F_{1, 171} = 5.369$, $P = 0.022$), which is opposite to the prediction. Prior work has demonstrated large genetic differences in life history traits between *R. hartii* from high-predation and *Rivulus*-only communities (Walsh and Reznick 2008), indicating local adaptation to these differences in predation in other traits. Furthermore, swimming performances are heritable in other fish (Nicoletto 1995, Garenc et al. 1998). In addition, training effects on sprint speed and endurance appear to be minor in fish (Davison 1997), so it is unlikely that the differences in performance that we observed could be attributable to differences in what the fish experienced before they were collected. For these reasons, the consistent results across three independent river drainages in the present study suggest that the differences in swimming performance in *R. hartii* between high-predation sites and those that lack predators are likely genetic in origin. However, in order to demonstrate this, fish bred and reared under common garden conditions would need to be compared (e.g., see Garland and Adolph 1991).

Although we found substantial differences among sites for both sprint performance and U_{crit} (Fig. 1), we

TABLE 2. Least-squares means (±SE) from SAS Procedure Mixed analyses as shown in Table 1 (see *Materials and methods*), for males and females from the three population types (sites).

Trait	High predation		*Rivulus* only	
	Male	Female	Male	Female
Sprint speed (cm/s)	1.0550 ± 0.0316	1.0301 ± 0.0335	0.9202 ± 0.0355	0.9721 ± 0.0398
Sprint speed (body lengths/s)	21.2170 ± 0.5873	20.9385 ± 0.6055	16.4459 ± 0.7274	16.1677 ± 0.7274
U_{crit} (log$_{10}$[cm/s])	1.1675 ± 0.0232	1.1391 ± 0.0246	1.3048 ± 0.0267	1.2362 ± 0.0301
U_{crit} (body lengths/s)	3.3521 ± 0.1856	3.0501 ± 0.1941	3.7337 ± 0.2332	2.8227 ± 0.2383
Tail length (mm)	9.1835 ± 0.1607	8.9598 ± 0.1708	7.9898 ± 0.1902	7.5392 ± 0.2138
Fineness	5.2628 ± 0.0522	5.0975 ± 0.0555	4.9300 ± 0.0618	5.0596 ± 0.0695
Total length (mm)	56.9786 ± 1.1715	55.8500 ± 1.2258	65.2794 ± 1.4724	72.8322 ± 1.4724
Standard length (mm)	48.2157 ± 1.0850	47.3992 ± 1.1352	57.2918 ± 1.3636	64.3542 ± 1.3636
Body depth (mm)	9.0746 ± 0.2660	9.2392 ± 0.2783	11.6308 ± 0.3343	12.9248 ± 0.3343
Mass (g)	2.1398 ± 0.1850	1.9868 ± 0.1935	2.9723 ± 0.2325	4.3079 ± 0.2325

TABLE 1.   Extended.

Covariate†			HP vs. (R and RG)			R vs. RG		
F	df	P	F	df	P	F	df	P
9.04	1, 164	0.0031	7.82	1,164	0.0058	0.24	1,164	0.6264
	1, 165		79.39	1,165	<0.0001	0.32	1,165	0.5703
0.26	1, 163	0.6076	21.37	1,163	<0.0001	0.71	1,163	0.4014
	1, 164		1.23	1,164	0.2698	1.04	1,164	0.3088
63.22	1, 165	<0.0001	54.56	1,165	<0.0001	0.03	1,165	0.8564
17.15	1, 165	<0.0001	9.51	1,165	0.0024	0.03	1,165	0.8582

also detected consistent differences among rivers for U_{crit} (Table 1) and one river that differed from the others for sprint performance only at the *Rivulus*-only site (Fig. 1A). (As explained in the Methods, we did not formally test for site × river interactions.) Further research will be required to determine if these river differences are stable over time and/or what aspects of the environment (e.g., food availability, current) or population demography (e.g., age structure: see Oufiero and Garland 2009) may lead to consistent or idiosyncratic differences among rivers.

Our results (Fig. 1A) and similar studies (O'Steen et al. 2002, Ghalambor et al. 2004, Walker et al. 2005, Langerhans 2009) suggest that increased sprint speed may improve Darwinian fitness in sites with high predation. However, direct evidence that a faster escape response in natural aquatic communities with predators actually improves fitness is virtually nonexistent.

Similarly, the increased critical swimming speed in the sites that lack predators could result from selection favoring increased endurance in such environments, which may enhance foraging ability, mating success, or dispersal abilities (Garland and Losos 1994, Garland 1999, Feder et al. 2010). *R. hartii* in low-predation sites shift their habitat use to more open water (Fraser and Gilliam 1992), and this behavioral response could expose them to higher current and hence favor an increase in U_{crit}. Yet, to our knowledge, no studies have examined the fitness advantages of an increased endurance capacity of fish in a natural setting. Recent work has begun to explore the relation between U_{crit} and

survival in seminatural aquatic systems (Claireaux et al. 2007), and work in terrestrial systems has demonstrated that increased endurance is beneficial for certain aspects of fitness (review in Irschick et al. 2008, but see Jayne and Bennett 1990). However, although we found a significant difference in critical swimming speed between high- and low-predation sites, with length represented as a covariate, this difference was not significant when U_{crit} was expressed as body lengths per second (Table 1), suggesting that this difference is potentially caused by differences in the size of the fish from different communities.

We found a lack of divergence in swimming performance between the two low-predation sites (Fig. 1, Table 1), which is not entirely surprising. Although some studies have found a negative effect of growth on swimming performance (Kolok and Oris 1995, Billerbeck et al. 2001), at least one other found no effect (Royle et al. 2006). Also, the differences in growth between *Rivulus*–guppy and *Rivulus*-only sites are environmental in origin (Walsh 2009), suggesting they have the same potential for growth; when *R. hartii* are transplanted from a *Rivulus*-only locality to a *Rivulus*–guppy locality immediately downstream, their growth rate accelerates to match that of the residents. The similarity in their potential for growth may explain the lack of difference in swimming performance as well as size and shape characters between the two low-predation sites (Table 2, Figs. 1 and 4). Additionally, the lack of difference in swimming performance between these two low-predation sites suggests that predation by *P. reticulata* on young *R. hartii* is not causing a divergence in adult swimming performance.

Swimming performance is a complex phenotype, with many levels of biological organization affecting how an organism performs in its environment (Arnold 1983, Garland and Losos 1994, Oufiero and Garland 2007, Feder et al. 2010). In the present study, the only measured candidate traits for predicting swimming performance (other than body size) are tail length and fineness ratio. Our results indicate that HP fish have longer tails (Tables 1 and 2, Fig. 4A), which, in principle, could account for their greater sprint speed. However, when tail length is added as an additional covariate to the ANCOVA model predicting sprint speed in Table 1, it does not have a statistically

TABLE 2.   Extended.

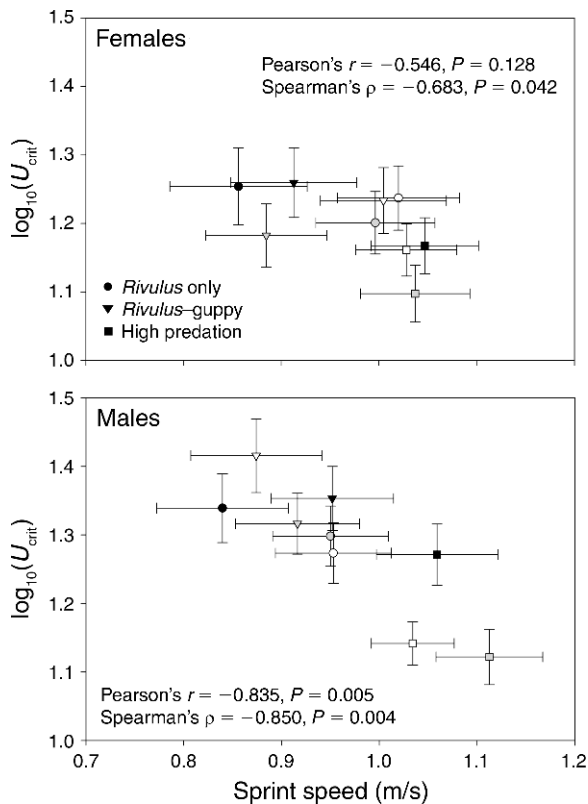| *Rivulus*–guppy	
Male	Female
0.9187 ± 0.0372	0.9388 ± 0.0388
16.1433 ± 0.7578	15.6337 ± 0.7268
1.3578 ± 0.0276	1.2281 ± 0.0291
4.2325 ± 0.2433	2.8104 ± 0.2379
8.0327 ± 0.1951	7.4266 ± 0.2039
4.9247 ± 0.0634	5.0872 ± 0.0663
66.1089 ± 1.5023	71.3879 ± 1.4713
57.9121 ± 1.3913	63.3721 ± 1.3626
11.8691 ± 0.3411	12.7713 ± 0.3340
3.3138 ± 0.2372	4.1485 ± 0.2323

FIG. 2. Relation between sprint speed and $\log_{10}(U_{crit})$ for the nine population least-squares means (see *Methods*). Results are shown separately for females and males with both Pearson's and Spearman's correlations reported. The relationship was significantly negative for both females (Spearman's $\rho = -0.683$, two-tailed $P = 0.042$) and males (Pearson's $r = -0.835$, two-tailed $P = 0.005$; Spearman's $\rho = -0.850$, two-tailed $P = 0.004$), suggesting a trade-off between these aspects of swimming performance. Symbols denote sites (circles, *Rivulus*-only sites [R]; triangles, *Rivulus*–guppy sites [RG], squares, high-predation sites [HP]); shading denotes river (black, Aripo; white, Guanapo; gray, Quare).

significant effect ($P = 0.7149$), and the effects of site ($P = 0.0226$) and standard length ($P = 0.0034$) remain significant, as does the a priori contrast of HP vs. (R and RG) sites ($P = 0.0064$). Therefore, some morphological or physiological characteristic other than tail length must account for the higher performance of HP fish. Similar results (not shown) were obtained when tail length was added to the ANCOVA model predicting U_{crit}. Fineness ratio was also not a significant predictor of either sprint speed or U_{crit} when added to the ANCOVA models, and again did not substantially alter the P values shown in Table 1. Further physiological and morphometric analyses are needed to determine the mechanisms underlying site, river, and sex differences in swimming performance (e.g., Kolok 1999, Guderley and Couture 2005).

The difference between the sexes was most pronounced in critical swimming speed and body size measures. Sex did not have a significant effect on sprint
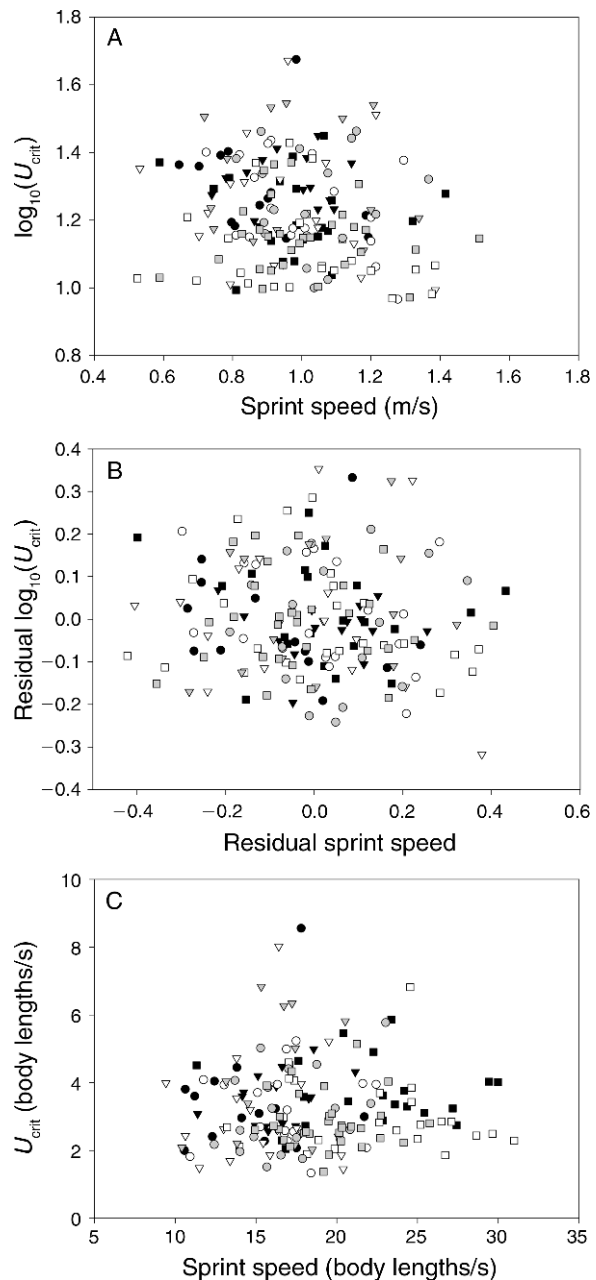

FIG. 3. Plots of the relationship between (A) raw sprint speed and $\log_{10}$ critical swimming speed (U_{crit}), (B) residual sprint speed and $\log_{10}(U_{crit})$ from the full ANCOVA model (see *Statistical analysis*), with standard length (SL) as a covariate, and (C) relative sprint speed and U_{crit}. Symbols denote sites, and shading denotes river, as in Fig. 2. The correlation is significantly negative for the raw values (A, Pearson's $r = -0.168$, two-tailed $P = 0.029$), but not for the residuals (B, Pearson's $r = -0.096$, two-tailed $P = 0.212$) or the relative values (C, Pearson's $r = 0.0152$, two-tailed $P = 0.844$; see Appendix B).

speed, similar to results found in Atlantic cod (*Gadus morhua*; Reidy et al. 2000), and also did not have a significant effect on tail length or fineness ratio. Our results show that males tend to be smaller than females
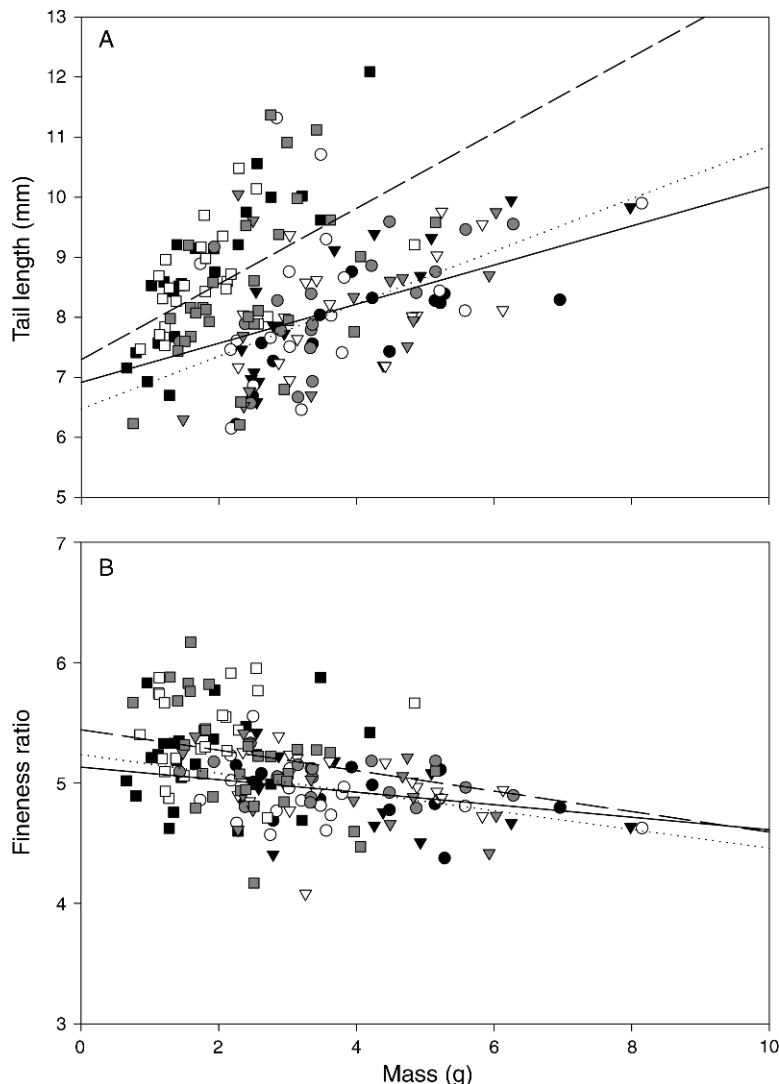
FIG. 4. Plots representing (A) tail length (total length − standard length) and (B) fineness ratio (standard length/body depth) as a function of mass. Symbols denote sites, and shading denotes river as in Fig. 2. Lines represent least-squares regressions for each site: solid line, R; dotted, RG; dashed, HP. There were significant differences between the HP sites and two low-predation sites (R and RG). HP tended to have higher fineness ratios and longer tails, but there was no statistical difference between the two low-predation sites (R and RG).

(Table 2), which may be related to fecundity selection on females if the differences in body size are maintained throughout their lifespan. In addition, males tend to have increased critical swimming speeds compared to females, for both absolute (cm/s) and relative (bl/s) values (Table 2). Several studies have demonstrated the effects of pregnancy on swimming performance in livebearing fish (Plaut 2002, Ghalambor et al. 2004), although Reidy et al. (2000) found no effect of sex on U_{crit} in adult *G. morhua*. The difference in U_{crit} between the sexes may be due to the effect of reproductive state of the female; however, *R. hartii* are egg-laying fish and the percentage of body mass that is allocated to reproduction in females varies from 4.5% to 6.9% compared to males, for which it is likely to be

considerably less (Walsh and Reznick 2009). There may also be selection for increased endurance in males related to finding mates or sexual selection; however, the intensity of sexual selection for this trait is not known in this species.

We found a significant effect of site for all measures of body size (Appendix C); males and females from HP sites were consistently smaller than fish from RG or R sites. This difference may be an artifact of high-predation sites consisting of larger proportions of smaller individuals in natural streams (due to higher mortality rates), and likely does not reflect a genetic divergence in body size (Rodd and Reznick 1997). *R. hartii* from high-predation and *Rivulus*-only sites do not

differ in rates of growth on controlled levels of food availability in the lab (Walsh and Reznick 2008).

In summary, our results demonstrate that *R. hartii* from high-predation sites are capable of higher sprint speeds but have poorer stamina than *R. hartii* from sites with lower predation. The replication of these differences across three river drainages suggests that fish in high-predation environments have evolved faster sprint speeds, compared to their low-predation counterparts. The apparent trade-off between acceleration and stamina was most evident in comparisons among population means (Fig. 2). Variation among individuals within populations yielded weaker and sometimes non-significant correlations (Fig. 3). More experiments are warranted to determine the genetic basis of these swimming performance differences and their ecological relevance.

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APPENDIX A

Map of collection sites (*Ecological Archives* E092-013-A1).

APPENDIX B

Correlations of sprint speed and critical swimming speed (U_{crit}) (*Ecological Archives* E092-013-A2).

APPENDIX C

ANOVA models for the difference in body size measurements (*Ecological Archives* E092-013-A3).