

RESEARCH ARTICLE

Effects of intraspecific variation in reproductive traits, pectoral fin use and burst swimming on metabolic rates and swimming performance in the Trinidadian guppy (*Poecilia reticulata*)

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SUMMARY

There is considerable intraspecific variation in metabolic rates and locomotor performance in aquatic ectothermic vertebrates; however, the mechanistic basis remains poorly understood. Using pregnant Trinidadian guppies (*Poecilia reticulata*), a live-bearing teleost, we examined the effects of reproductive traits, pectoral fin use and burst-assisted swimming on swimming metabolic rate, standard metabolic rate ($\dot{M}_{O_{2std}}$) and prolonged swimming performance (U_{crit}). Reproductive traits included reproductive allocation and pregnancy stage, the former defined as the mass of the reproductive tissues divided by the total body mass. Results showed that the metabolic rate increased curvilinearly with swimming speed. The slope of the relationship was used as an index of swimming cost. There was no evidence that reproductive traits correlated with swimming cost, $\dot{M}_{O_{2std}}$ or U_{crit} . In contrast, data revealed strong effects of pectoral fin use on swimming cost and U_{crit} . *Poecilia reticulata* employed body-caudal fin (BCF) swimming at all tested swimming speeds; however, fish with a high simultaneous use of the pectoral fins exhibited increased swimming cost and decreased U_{crit} . These data indicated that combining BCF swimming and pectoral fin movement over a wide speed range, presumably to support swimming stability and control, is an inefficient swimming behaviour. Finally, transition to burst-assisted swimming was associated with an increase in aerobic metabolic rate. Our study highlights factors other than swimming speed that affect swimming cost and suggests that intraspecific diversity in biomechanical performance, such as pectoral fin use, is an important source of variation in both locomotor cost and maximal performance.

Key words: basal metabolic rate, energetics, gait transition, gravidity, life history, respiratory physiology.

Received 16 November 2012; Accepted 8 May 2013

INTRODUCTION

Intraspecific variation in metabolic rate and locomotor performance remain poorly understood in many taxa. Variation in physiological traits may be important, however, because it can be functionally significant and reflect behavioural or physiological trade-offs, where the costs or benefits of any phenotype are variable and may depend on internal and external factors (Williams, 2008; Biro and Stamps, 2010; Burton et al., 2011). For example, intraspecific variation in metabolic rate and locomotor performance may be associated with disruptive selection regimes leading to variation in foraging strategy and predator avoidance (Marras et al., 2010). Moreover, studies of physiological diversity may reveal the physiological basis of intraspecific variation in life history traits (Speakman, 2005; Arnott et al., 2006; Williams, 2012). Finally, phenotypic diversity may be indicative of genetic diversity and the degree to which a population can adjust to environmental change (Hayes and Jenkins, 1997; Bolnick et al., 2003; Sears et al., 2009).

Reproductive status may be a source of intraspecific variation in metabolic rate and locomotor performance. Elevated metabolic rate in relation to gravidity or pregnancy has been reported in many animals, including the eastern garter snake (*Thamnophis sirtalis*) (Birchard et al., 1984), mountain spiny lizard (*Sceloporus jarrovi*)

(DeMarco, 1993), yellowtail rockfish (*Sebastes flavidus*) (Hopkins et al., 1995), Korean rockfish (*Sebastes schlegeli*) (Boehlert et al., 1991), sailfin molly (*Poecilia latipinna*) (Timmerman and Chapman, 2003), striped surfperch (*Embiotoca lateralis*) (Webb and Brett, 1972) and European eelpout (*Zoarces viviparus*) (Skov et al., 2010). Several studies have demonstrated diminished locomotor performance caused by gravidity or pregnancy. Examples include the northern death adder (*Acanthophis praelongus*) (Webb, 2004), side-blotched lizard (*Uta stansburiana*) (Miles et al., 2000), short-horn sculpin (*Myoxocephalus scorpius*) (James and Johnston, 1998) and mosquitofish (*Gambusia affinis*) (Plaut, 2002; Belk and Tuckfield, 2010). Using the Trinidadian guppy (*Poecilia reticulata*), Ghalambor and colleagues provided evidence that pregnancy may constrain fast-start swimming performance employed to evade predators (Ghalambor et al., 2004). It has been suggested that diminished swimming performance in live-bearing pregnant fish may be attributed to metabolic constraints caused by the embryos (Plaut, 2002); however, to our knowledge such relationships have not been examined.

The impact of pregnancy on female performance could have important ecological and evolutionary consequences. For example, pregnant bighorn sheep (*Ovis canadensis*) spend less time in

optimal foraging areas, where the predation risk is highest, than females that have recently given birth (Berger, 1991). Such differences in behaviour may reduce the predation risk associated with diminished locomotor performance at the cost of resource acquisition. From an evolutionary point of view, cost of reproduction represents one of the most prominent elements in life history evolution (Stearns, 1989). Using free-ranging lizards, Miles and colleagues demonstrated that a decrement in performance is associated with current reproductive investment and represents a cost of reproduction expressed as diminished locomotor performance and lowered survivorship to next clutch (Miles et al., 2000).

Recent studies on the metabolic rates of swimming fish have included measurements of gait transitions occurring as a function of swimming speed (Korsmeyer et al., 2002; Jones et al., 2007; Cannas et al., 2006; Svendsen et al., 2010). A gait is 'a pattern of locomotion characteristic of a limited range of speeds described by quantities of which one or more change discontinuously at transitions to other gaits' (Alexander, 1989). However, as far as is known, no previous studies have investigated how intraspecific variation in fin use within a single gait affects swimming cost and maximal performance. Moreover, while previous studies have examined the metabolic rates associated with the transition from rigid-body, median or paired-fin (MPF) swimming to undulatory, body-caudal fin (BCF) swimming (Korsmeyer et al., 2002; Cannas et al., 2006; Svendsen et al., 2010), the energetics of the gait transition from steady BCF swimming to unsteady BCF swimming (i.e. burst-assisted) remain poorly understood (Farrell, 2007).

The objective of this study was to examine whether diversity in reproductive traits and swimming behaviour correlate with intraspecific variation in metabolic rates and maximal locomotor performance. Reproductive traits included reproductive allocation and pregnancy stage, the former defined as the mass of reproductive tissues divided by the total body mass. Swimming behaviour included use of the pectoral fins and gait transition from steady BCF swimming to unsteady BCF swimming (i.e. burst-assisted). To this end, we used *P. reticulata* Peters 1859 captured in Trinidad for swimming trials at increasing speeds.

Poecilia reticulata is a live-bearing species producing one litter every 3–4 weeks (Reznick and Yang, 1993). Reproductive allocation in female *P. reticulata* tends to vary with season (Reznick, 1989), resource availability (Reznick and Yang, 1993) and predation regime (Reznick and Endler, 1982). In terms of locomotion, *P. reticulata* is an acanthomorph fish (Chen et al., 2003) and as such, the pectoral fins are located relatively high on the body, at an approximately mid-dorsal position and relatively close to the centre of mass of the fish (Drucker et al., 2006). Compared with less derived fishes, the pectoral fins of acanthomorph fishes are generally associated with a wider range of motion and a correspondingly greater propulsor diversity (Drucker et al., 2006). Moreover, *P. reticulata* is a BCF swimmer that may switch to burst-assisted swimming (Pohlmann et al., 2001). Several studies have used Trinidadian *P. reticulata* to investigate factors causing intraspecific variation in relation to evolutionary ecology (Magurran, 2005), and *P. reticulata* is a key organism for empirical tests of theoretical life history models (Reznick et al., 1990; Reznick et al., 1996; Ghalambor et al., 2003). We used individual female *P. reticulata*, varying in reproductive traits, to document swimming metabolic rates, standard metabolic rate, swimming behaviour and prolonged swimming performance. Measurements of excess post-exercise oxygen consumption (Lee et al., 2003b) were included because individual variation in swimming performance might be related to processes associated with anaerobic rather than aerobic power production.

We predicted that reproductive allocation and/or pregnancy stage would correlate positively with metabolic swimming cost and negatively with prolonged swimming performance. Further, we predicted that standard metabolic rate would correlate positively with reproductive allocation and/or pregnancy stage. In terms of fin use, we predicted that fish extending their pectoral fins would experience increased drag and increased swimming cost, as hypothesised by previous studies (Webb, 1998; Weihs, 2002; Green and Hale, 2012). Inefficient fin use at increasing speeds could translate into decreased swimming performance. For example, if extending the pectoral fins causes a consistent increase in the swimming cost, a fish with extensive pectoral fin use at increasing speeds could exhibit inferior swimming performance, because the fish would reach the maximum metabolic rate at a relatively slow swimming speed. Finally, as a consequence of gait transition to burst-assisted swimming, either aerobic metabolic rate (i.e. oxygen consumption rate during exercise) should plateau, or the rate of increase, as a function of swimming speed, should decline because burst-assisted swimming is partly covered by anaerobic metabolism (Farrell, 2007).

MATERIALS AND METHODS

Experimental animals

A total of 18 female *P. reticulata* (mean \pm s.e.m. body mass = 0.296 \pm 0.009 g; total length = 3.0 \pm 0.0 cm) were captured using butterfly nets in the Naranjo River in Trinidad. The river is a low-predation tributary to the Aripo River. The mean current velocity at the collection site was 12.7 \pm 1.2 cm s⁻¹. In the laboratory, fish were kept in five identical holding tanks (30 l each) using filtered water originating from the Arima Valley. Each tank housed four to five individuals including one male. One air stone in each tank secured normoxic conditions. Each tank was cleaned and water was replaced every third day. Prior to experimentation, fish were acclimated to the laboratory for 2–3 weeks. Fish were fed daily on commercial flake food to satiation. Mean water temperature in the holding tanks was 25.8°C (range: 24.6–26.9°C). All fish holding procedures were identical for the five tanks.

Poecilia reticulata is a lecithotrophic species. Lecithotrophic means that there is no placental-like connection between the mother and young (Reznick and Yang, 1993), such that yolk stored in the egg is assumed to be the only source of embryo nutrition. Recent work within the Poeciliidae has indicated, however, that some mother-to-embryo nutrient transfer may occur in species thought to be lecithotrophic (Marsh-Matthews et al., 2005; Marsh-Matthews et al., 2011). To our knowledge, no attempt has been made to quantify any post-fertilization provisioning in *P. reticulata*, and it is not known to what degree there is a limited transfer of oxygen or small molecules.

Respirometry

A 0.170 l Bläzka-type swimming respirometer (Model SW10000; Loligo Systems Aps, Tjele, Denmark) was used to measure oxygen consumption rate (\dot{M}_{O_2} ; mg O₂ kg⁻¹ h⁻¹) as a function of swimming speed (U). The respirometer was submerged in an ambient tank (0.9 \times 0.35 \times 0.39 m) supplying water for the respirometer. Water temperature was maintained at 26.0°C (range: 25.9–26.1°C) using two cooling Peltier elements (IceProbe; Cool Works, San Rafael, CA, USA) and a submersible heater (50 W; AkvaStabil; Haderslev, Denmark). An air stone in the ambient tank maintained oxygen levels at >95% air saturation.

The inner dimensions of the cylindrical observation section in the respirometer were 26 \times 100 mm (diameter \times length). An impeller, placed downstream of the observation section, was driven by an

external electric motor that generated the re-circulating flow. Deflectors situated upstream of the observation section collimated the flow. To promote rectilinear flow and a uniform velocity profile in the observation section, water was passed through an upstream honeycomb (3 mm cell diameter) producing a micro turbulent flow. A grid (2×2 mm) in the downstream direction bounded the observation section. Water speeds in the observation section were measured using a laser Doppler anemometer consisting of a 4 W Ar-ion laser, a fibre probe and BSA data processors (Dantec Dynamics, Skovlunde, Denmark). The measurements were used to correlate water speed with voltage output from the external motor controller. Additional details have been published previously (Poulsen et al., 2012).

Polystyrene sheets covered the majority of the ambient tank to minimize any outside stimuli affecting the fish during the experiment. A small opening was used for behavioural observations. Fish were encouraged to swim in the most upstream part of the observation section using a darkening hide.

Oxygen partial pressure in the respirometer was measured using fibre optic sensor technology (PreSens, Regensburg, Germany). Intermittent-flow respirometry was applied in accordance with a previous study (Steffensen, 1989). The respirometer was fitted with an inlet port and a standpipe outlet, through which the volume of water in the respirometer could be replaced with a computer-actuated pump. The software AutoResp (Loligo Systems Aps, Tjele, Denmark) was used to control the flush (240 s), wait (120 s) and measurement (360 s) phases. These settings provided one \dot{M}_{O_2} measurement per 12 min. Preliminary trials demonstrated that the R^2 associated with each \dot{M}_{O_2} measurement was always >0.95, similar to previous studies (Claireaux et al., 2006; Svendsen et al., 2012). The oxygen content never fell below 18.4 kPa. Standard equations were used to calculate \dot{M}_{O_2} (Svendsen et al., 2010). Water in the ambient tank was recirculated through a loop consisting of a separate mechanical filter (Pick-up 2006; Eheim, Deizisau, Germany) and a UV sterilizer (UV-10000; Tetra Pond, Melle, Germany). Between experimental runs, the entire setup was cleaned using a chlorine solution, flushed repeatedly, and refilled with water from the same source as used for the fish holding.

Fish for experiments were starved for 24 h prior to respirometry to ensure a post-absorptive state. Fish mass (to nearest 0.001 g), length, depth and width (all to nearest 0.5 mm) were determined for pre-experimental calculation and correction of the solid blocking effects, ranging from 2.2 to 4.2%. Calculations of solid blocking effects followed Bell and Terhune (Bell and Terhune, 1970).

Each *P. reticulata* was introduced to the working section and given at least 8 h (overnight) to acclimate while swimming at 2 BL s^{-1} (total body lengths per second). Preliminary trials demonstrated that 2 BL s^{-1} was the minimum swimming speed that secured positive rheotaxis (i.e. upstream orientation of the anterior body part). After the acclimation period, fish maintained a low \dot{M}_{O_2} , even when exposed to a few stepwise increases in the swimming speed. Occasionally, the acclimation period was extended to meet this criterion. Subsequently, each individual fish was exposed to progressive increments in the swimming speed of 0.5 BL s^{-1} every 12 min until fatigue. \dot{M}_{O_2} was measured at each swimming speed. Preliminary trials demonstrated that the critical (maximum) swimming speed (U_{crit}) was $9\text{--}17 \text{ BL s}^{-1}$. The speed increment (0.5 BL s^{-1}) was chosen to ensure an adequate number of \dot{M}_{O_2} measurements (>12) at increasing speeds in individual fish. This type of data was required because we aimed at describing the relationship between U and \dot{M}_{O_2} using an equation representing each individual fish. Maximum \dot{M}_{O_2} ($\dot{M}_{O_2\text{max}}$) was estimated as the

highest \dot{M}_{O_2} measured during the swimming protocol (McKenzie et al., 2003).

Immediately after fatigue, the swimming speed was returned to 2 BL s^{-1} (acclimation speed), following Lee and colleagues (Lee et al., 2003b). Using this swimming speed, \dot{M}_{O_2} was measured for 1 h to quantify any excess post-exercise oxygen consumption (EPOC) (Lee et al., 2003b). Levels of background respiration were estimated from blank runs and used to correct the \dot{M}_{O_2} measurements following Jones et al. (Jones et al., 2007).

Swimming behaviours and critical swimming speed

Behavioural data were collected during the swimming trials, similar to a previous study (Swanson et al., 1998). During the measurement phase (6 min) of the respirometric loop (12 min), time spent swimming with extended pectoral fins, with caudal undulation and using burst-assisted swimming were recorded. Use of each behaviour was recorded over a 1 min time interval (i.e. 3 min in total). These data were collected during each 12 min interval, starting at the acclimation speed (2 BL s^{-1}) and ending at fatigue. End point values were the percentages of time allocated to these swimming behaviours at each swimming speed (Korsmeyer et al., 2002; Webb and Fairchild, 2001). The values were used to calculate the average fin and gait use (% of time) during the complete swimming trial for the individual fish. Finally, the gait transition speed [U_{STmax} (Peake, 2008)] from steady to unsteady swimming (i.e. burst-assisted) was recorded as the highest swimming speed without unsteady swimming.

The equation provided by Brett was used to calculate U_{crit} (Brett, 1964). Oufiero and Garland demonstrated that the U_{crit} protocol yields critical swimming speeds that are repeatable for individual *P. reticulata*, indicating that they represent actual measures of organism performance (Oufiero and Garland, 2009).

Reproductive traits

Immediately after the swimming trial, fish were euthanized using an overdose of MS-222 and preserved in 6% formaldehyde. Wet and dry reproductive allocation (RA) were quantified using methods similar to those of Reznick (Reznick, 1983). Briefly, embryos and associated reproductive tissues were separated from female somatic tissue. Stage of embryonic development (i.e. pregnancy stage) was determined morphologically following standard procedures (Haynes, 1995). Development ranged from stage 0 (an egg with yolking ova) to stage 50 (fully developed embryos, ready to be born). Wet masses of the reproductive and somatic tissues were measured using a Mettler AE163 analytical balance (Mettler-Toledo, Columbus, OH, USA) and recorded to the nearest 0.00001 g. Subsequently, the tissues were air dried for 24 h at 600°C and weighed again following the same procedure. RA was calculated as the mass of reproductive tissues divided by the total body mass.

Data analysis

As indicated, we aimed at describing the relationship between U and \dot{M}_{O_2} using an equation representing each individual fish. Previous studies have used power, exponential and polynomial models to describe the relationship between U and \dot{M}_{O_2} (Korsmeyer et al., 2002; Arnott et al., 2006; Tudorache et al., 2011). Webb recommended that a certain model should not be assumed, but rather a model should be used that best describes the available data (Webb, 1993). Accordingly, we examined various models before determining the most appropriate model. Using the model for individual fish, \dot{M}_{O_2} was extrapolated to zero swimming speed to estimate standard metabolic rate ($\dot{M}_{O_2\text{std}}$), following previous studies

(Brett, 1964; Arnott et al., 2006). The model was also used to estimate metabolic swimming cost in individual fish, expressed as the slope of the relationship between U and \dot{M}_{O_2} .

To test the predictions of this study, reproductive traits and pectoral fin use (considered the independent variables) were correlated with swimming cost and $\dot{M}_{O_{2std}}$, both derived from the identified model, as well as U_{crit} (considered the dependent variables). Reproductive traits and pectoral fin use were not manipulated experimentally. Instead, the analyses relied on *post hoc* intraspecific variation resulting from differences among individuals. To test our predictions, linear least square regression was used to examine whether the reproductive traits correlated positively with $\dot{M}_{O_{2std}}$ and swimming cost and negatively with U_{crit} . In terms of pectoral fin use, we tested whether this variable correlated positively with swimming cost and negatively with U_{crit} . To assess such relationships further, we also tested for a negative correlation between U_{crit} and swimming cost.

The final objective of this study was to test the prediction that gait transition from steady BCF to unsteady BCF swimming would cause either \dot{M}_{O_2} to plateau, or the rate of increase, as a function of swimming speed, to decline. To examine this prediction, we compared \dot{M}_{O_2} before and after transition to burst-assisted swimming at increasing speeds using a sign test.

Because the five fish holding tanks were identical, maintained in an identical fashion and kept in the same room, we have no reason to believe that the different tanks affected the fish differently. Therefore, tank origin was not included in any analyses.

Estimates of $\dot{M}_{O_{2std}}$ and $\dot{M}_{O_{2max}}$ were used to estimate the metabolic scope (MS). The MS was defined as the difference between $\dot{M}_{O_{2std}}$ and $\dot{M}_{O_{2max}}$, following Farrell and Richards (Farrell and Richards, 2009). The speed at which fish transitioned from steady to unsteady BCF swimming (U_{STmax}) and the simultaneous

\dot{M}_{O_2} measurements were used to partition the MS into the proportion attributed to steady swimming and the proportion attributed to unsteady swimming.

To detect EPOC, we compared the individual pre-exercise \dot{M}_{O_2} with the first post-exercise \dot{M}_{O_2} using a paired *t*-test after examining the assumptions of a normal distribution of data and homogeneity of variance. Both data sets were collected while the fish was swimming at 2 BL s^{-1} (acclimation speed). If post-exercise \dot{M}_{O_2} was significantly higher than pre-exercise \dot{M}_{O_2} , it was considered evidence of EPOC and anaerobic power production, following a previous study (Svendsen et al., 2010).

The free statistical software R (R Development Core Team, 2011) was used for statistical analyses. The R package nlme (Pinheiro et al., 2011) was used to fit models. Results were considered significant at $P < 0.05$. All values are reported as means \pm s.e.m. unless otherwise noted.

RESULTS

The behavioural data showed that *P. reticulata* employed the caudal fin for swimming (i.e. BCF swimming) at all speeds (data not shown). In contrast, use of the pectoral fins and burst-assisted swimming varied with swimming speed (Fig. 1). As swimming speed increased, the use of the pectoral fins decreased; however, there was no distinct threshold speed at which fish discontinued using the pectoral fins (Fig. 1). In fact, two individuals used the pectoral fins at all swimming speeds (Fig. 1).

Most fish (15 out of 18) employed burst-assisted swimming at the highest swimming speeds (Fig. 1). Burst-assisted swimming was less variable than use of the pectoral fins. All fish that started using burst-assisted swimming continued doing so throughout the remaining swimming trial (Fig. 1). The average U_{STmax} from steady swimming to unsteady swimming (i.e. burst-assisted swimming) was

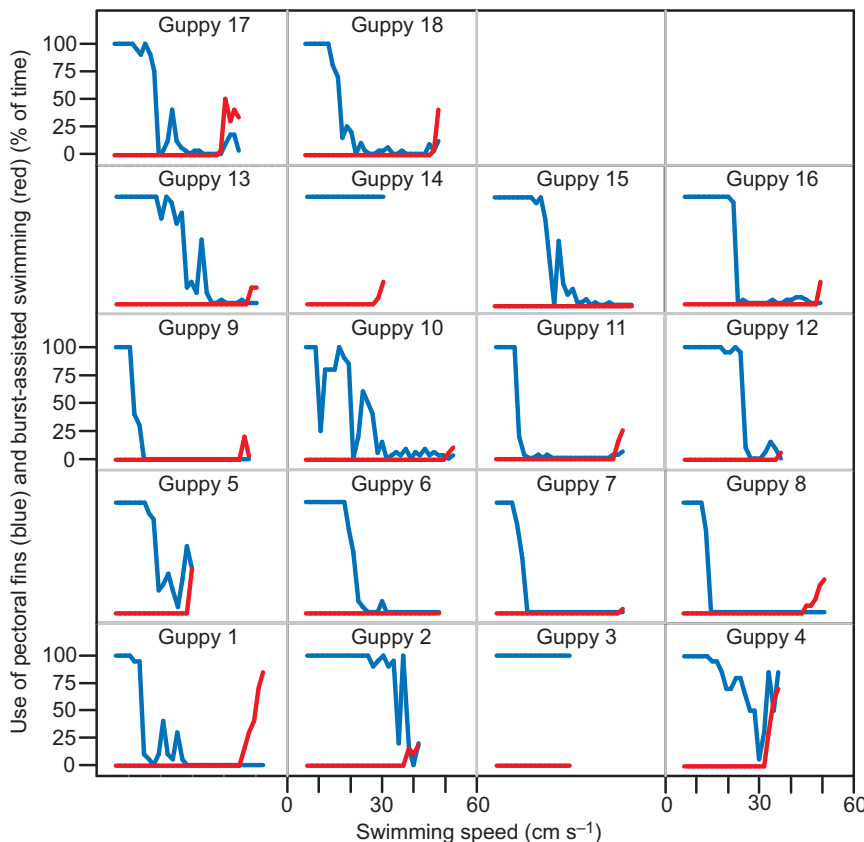


Fig. 1. Use of pectoral fins and burst-assisted swimming (% of time) in 18 individual *Poecilia reticulata* as a function of swimming speed (U ; cm s^{-1}). Pectoral fin use is indicated using a blue line, whereas burst-assisted swimming is indicated using a red line (unsteady swimming). The caudal fin was employed at all swimming speeds (i.e. body-caudal fin swimming). Each fish is represented by an average of 28 measurements of pectoral fin use and burst-assisted swimming.

$40.85 \pm 1.79 \text{ cm s}^{-1}$, equivalent to $13.48 \pm 0.59 \text{ BL s}^{-1}$. This measure included the maximum recorded steady swimming speed of three individuals that did not perform burst-assisted swimming (Fig. 1). The mean U_{crit} was $44.99 \pm 1.84 \text{ cm s}^{-1}$, equivalent to $14.89 \pm 0.66 \text{ BL s}^{-1}$. There was no significant relationship between fish total length and U_{crit} ($P > 0.1$, $R^2 < 0.16$).

In terms of metabolic rates, \dot{M}_{O_2} as a function of U in individual fish was best described by the exponential function:

$$\dot{M}_{\text{O}_2} = a \exp(Ub), \quad (1)$$

where a is the \dot{M}_{O_2} at zero speed (i.e. $U=0$) and b is the rate of increase in the \dot{M}_{O_2} as a function of U . The equation has been used to describe relationships between \dot{M}_{O_2} and U in a number of studies (Brett, 1964; McKenzie et al., 2003; Arnott et al., 2006). Eqn 1 provided an estimate of the $\dot{M}_{\text{O}_2\text{std}}$ (i.e. a) and swimming cost (i.e. b) measured at increasing U in individual fish.

Maximum likelihood estimation was performed on the logarithm of \dot{M}_{O_2} . The $\log \dot{M}_{\text{O}_2}$ was described by the following linear random coefficient model for the i th observation on the j th fish:

$$\log \dot{M}_{\text{O}_2ij} = a'_j + U_i b_j + e_{ij}, \quad i = 1, \dots, n_j, j = 1, \dots, 18. \quad (2)$$

The fish-specific intercepts a'_j and slopes b_j were assumed to follow normal distributions with means μ_a and μ_b and variances σ_a^2 and σ_b^2 . Moreover, a correlation between a'_j and b_j , ρ , was allowed. An AR1 (autoregressive of order 1) autocorrelation was allowed in the residuals using the equation:

$$e_{ij} = \phi e_{i-1,j} + \varepsilon_{ij}, \quad (3)$$

where ϕ is the autoregressive parameter and ε_{ij} are the uncorrelated residuals. The ε_{ij} are assumed to be independently and identically normally distributed with mean zero and variance σ_ε^2 . The \dot{M}_{O_2} at zero U is $a_j = \exp(a'_j)$ for the j th fish, while b_j is the rate of increase in \dot{M}_{O_2} as a function of swimming speed for the j th fish. Thus, a_j and b_j represent $\dot{M}_{\text{O}_2\text{std}}$ and swimming cost, respectively, for the j th fish.

Measurements of \dot{M}_{O_2} in relation to U in individual fish are plotted in Fig. 2. Data indicated that the rate of increase in \dot{M}_{O_2} , as a function

of U , was lower at speeds when burst-assisted swimming was not employed (steady swimming) than at speeds when burst-assisted swimming was employed (unsteady swimming) (Fig. 2). Consequently, the parameters in Eqn 1 were estimated using observations with steady swimming only (Figs 1, 2).

The mean $\dot{M}_{\text{O}_2\text{std}}$ was $\exp(\mu_a) = \exp(5.76) = 318.05 \text{ mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$. The 95% confidence interval was $294.01 - 344.05 \text{ mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$. The average rate of increase in the \dot{M}_{O_2} as a function of U was 0.0262 (Fig. 2). Estimates of the parameters for Eqns 1–3 are provided in Table 1. The average $\dot{M}_{\text{O}_2\text{max}}$ was $1270.69 \pm 40.50 \text{ mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$. Body mass correlated weakly with $\dot{M}_{\text{O}_2\text{std}}$ and $\dot{M}_{\text{O}_2\text{max}}$ in a positive and negative fashion, respectively, but none of the relationships were significant ($P > 0.05$).

After completing the swimming trial, fish were dissected and RA_j (i.e. fish-specific RA) and fish-specific pregnancy stage were quantified as described above. Measurements showed that both wet and dry RA varied between individuals (Table 2). Likewise, the pregnancy stages varied between individuals (Table 2). Wet and dry RA_j and fish-specific pregnancy stage were related to \hat{b}_j (i.e. estimated fish-specific steady swimming cost), \hat{a}_j (i.e. estimated fish-specific $\dot{M}_{\text{O}_2\text{std}}$) and $U_{\text{crit},j}$ (i.e. fish-specific U_{crit}). The tests revealed no significant relationships (all $P > 0.1$). These findings indicated that steady swimming cost, $\dot{M}_{\text{O}_2\text{std}}$ and U_{crit} did not correlate with the reproductive traits.

The same tests were carried out using average pectoral fin use instead of the reproductive traits. These tests revealed that steady swimming cost (i.e. \hat{b}_j) correlated positively with the average pectoral fin use ($P < 0.001$, $R^2 = 0.56$; Fig. 3). Hence, *P. reticulata* that spent more time with extended pectoral fins had increased steady swimming costs (Fig. 3). There was no correlation between \hat{a}_j and the average pectoral fin use, indicating that $\dot{M}_{\text{O}_2\text{std}}$ and average pectoral fin use were unrelated ($P = 0.42$).

There was a negative correlation between the average pectoral fin use and $U_{\text{crit},j}$ ($P < 0.0001$, $R^2 = 0.70$; Fig. 4). Hence, *P. reticulata* that spent more time with extended pectoral fins had a low U_{crit} (Fig. 4). There was no correlation between average pectoral fin use and $\dot{M}_{\text{O}_2\text{max},j}$ (fish-specific $\dot{M}_{\text{O}_2\text{max}}$), or between $U_{\text{crit},j}$ and $\dot{M}_{\text{O}_2\text{max},j}$.

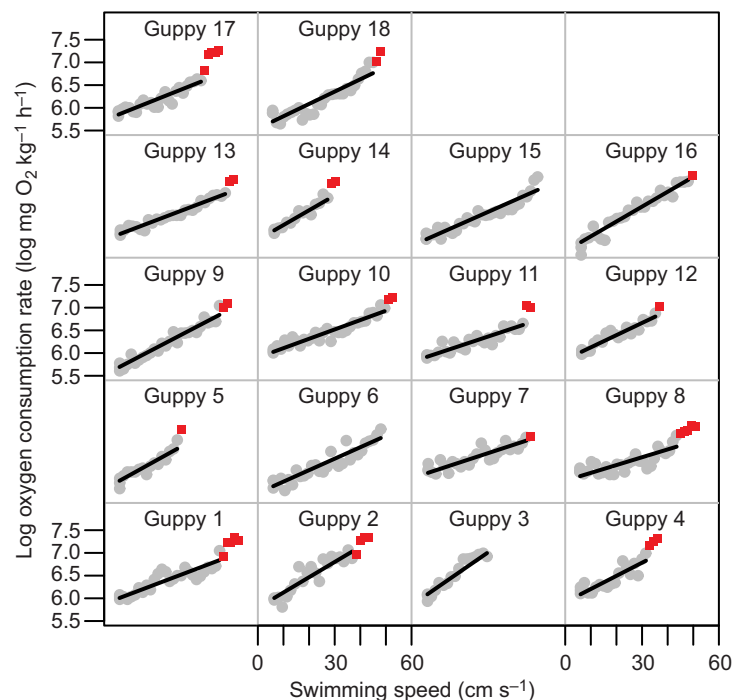


Fig. 2. Oxygen consumption rate (\dot{M}_{O_2} ; $\text{mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$) as a function of swimming speed (U ; cm s^{-1}) in 18 individual *P. reticulata*. Grey symbols represent \dot{M}_{O_2} when no burst-assisted swimming occurred (steady swimming), whereas red symbols represent \dot{M}_{O_2} when burst-assisted swimming occurred (unsteady swimming) (see Fig. 1). Each datum represents a 12 min period. Eqn 1 was fitted to the data without burst-assisted swimming (black lines) (see Table 1). The slope of each black line represents the rate of increase in \dot{M}_{O_2} as a function of U and is considered an index of steady swimming cost in individual fish.

Table 1. Model parameters and statistical analysis using an exponential equation (Eqn 1) fitted to oxygen consumption rate (\dot{M}_{O_2}) data as a function of swimming speed (U) in *Poecilia reticulata* (see Fig. 2)

	Value	s.e.m.	d.f.	t	P
μ_a	5.7622	0.0401	449	143.662	
μ_b	0.0262	0.0018	449	14.807	<0.001
σ_a	0.152				
σ_b	0.0068				
ρ	-0.425				
σ_ϵ	0.119				
ϕ	0.393				
Log-likelihood	321.3				

The significant P -value (<0.001) indicates that the mean rate of increase in \dot{M}_{O_2} as a function of U is different from zero.

Model components are described in the text and in the list of symbols and abbreviations.

(both $P>0.25$), indicating that $\dot{M}_{O_{2max}}$ did not influence the average pectoral fin use or U_{crit} .

The average pectoral fin use by individual fish was calculated using three different methods: (1) the average pectoral fin use throughout the complete swimming trial (i.e. from acclimation speed to U_{crit}); (2) the average pectoral fin use up to the initiation of burst-assisted swimming; and (3) the average pectoral fin use up to 9.5 BL s^{-1} (equivalent to 28.8 $cm s^{-1}$). This swimming speed represented the highest swimming speed that all fish managed to maintain for a complete respirometric loop (Figs 1, 2). The average pectoral fin use data presented in Figs 3 and 4 were based on methods 2 and 1, respectively. The relationships shown in Figs 3 and 4 were present and significant (all $P<0.02$, $R^2>0.31$) regardless of the method employed to calculate the average pectoral fin use for the individual fish. These findings indicated that the relationships between average pectoral fin use and steady swimming cost (Fig. 3) and U_{crit} (Fig. 4) were not artefacts caused by the variable swimming performance of the fish.

Fish condition index was calculated following a previous study (Marras et al., 2011) and correlated with average pectoral fin use. Employing methods 1 and 2 to calculate average pectoral fin use, there was no significant correlation between fish condition index and average pectoral fin use (both $P>0.11$). When method 3 was employed, fish condition index correlated negatively with the average pectoral fin use ($P=0.01$, $R^2=0.34$). Because of the inconsistent relationships, a possible effect of condition index on pectoral fin use was not considered any further.

Finally, \hat{b}_j correlated negatively with $U_{crit,j}$ ($P=0.002$, $R^2=0.46$; Fig. 5). Hence, *P. reticulata* with a low U_{crit} had increased steady swimming costs in comparison with fish with a high U_{crit} (Fig. 5).

Table 2. Reproductive traits of 18 female *P. reticulata* used for measurements of oxygen consumption rate and critical swimming speed

	Mean \pm s.d.	Range
Wet reproductive allocation (%)	16.38 \pm 4.52	4.93–24.67
Dry reproductive allocation (%)	16.03 \pm 5.70	1.23–26.45
Pregnancy stage	24.72 \pm 17.53	0–50

Reproductive allocation (wet and dry) was defined as the mass of the reproductive tissues divided by the total body mass. Pregnancy stage was determined morphologically following standard procedures (Haynes, 1995). Pregnancy stage was inferred from embryonic development stages ranging from stage 0 (an egg with yolkling ova) to stage 50 (fully developed embryos, ready to be born).

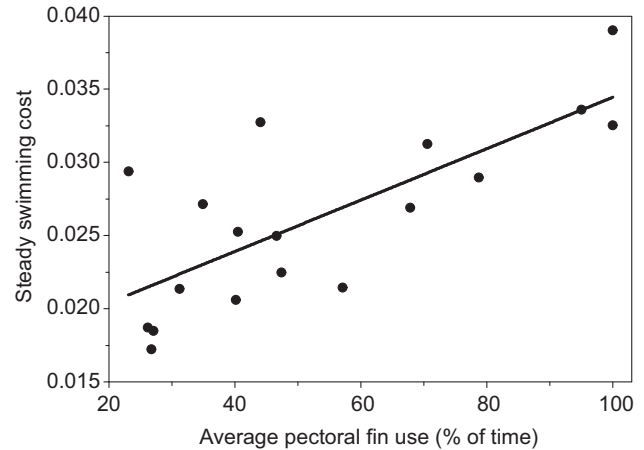


Fig. 3. Steady swimming cost as a function of the average pectoral fin use (% of time) in 18 individual *P. reticulata*. Steady swimming cost was measured as the rate of increase in the oxygen consumption rate as a function of swimming speed (see Fig. 2, Eqn 1, Table 1). Both steady swimming cost and average pectoral fin use were based on measurements up to the swimming speed where burst-assisted swimming occurred (see Fig. 1).

Collectively, the data shown in Figs 3–5 indicated that elevated pectoral fin use increased steady swimming costs, which translated into a low U_{crit} . It appeared that increased steady swimming costs meant that *P. reticulata* with elevated pectoral fin use reached the maximum metabolic rate at a relatively low speed and therefore had a low U_{crit} . The findings suggest that inefficient fin use at increasing swimming speeds is coupled with a low U_{crit} .

Metabolic rate data collected when unsteady swimming occurred were insufficient to estimate the actual rate of increase in the \dot{M}_{O_2} as a function of U , specific for this swimming gait (Fig. 2). It was clear, however, that the vast majority of the \dot{M}_{O_2} data points during unsteady swimming were higher than what would be expected based on extrapolation of the values representing steady swimming (Fig. 2). To examine these observations statistically, a sign test was used to investigate whether observations involving unsteady swimming (Fig. 2) were distributed around the prediction of the exponential Eqn 1 with an equal probability against the two-sided

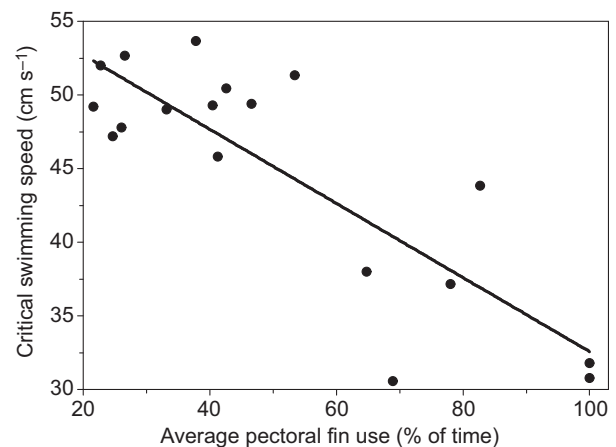


Fig. 4. Critical swimming speed (U_{crit} ; $cm s^{-1}$) as a function of average pectoral fin use (% of time) in 18 individual *P. reticulata*. Average pectoral fin use was based on measurements up to U_{crit} (i.e. including burst-assisted swimming) (see Fig. 1).

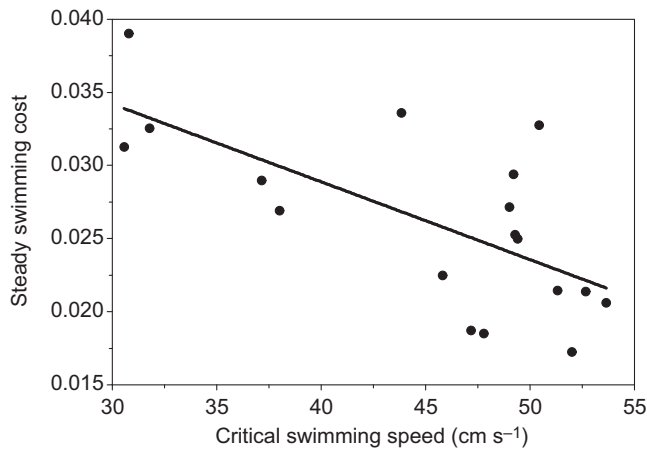


Fig. 5. Steady swimming cost as a function of critical swimming speed (U_{crit} ; cm s^{-1}) in 18 individual *P. reticulata*. Steady swimming cost was measured as the rate of increase in the oxygen consumption rate as a function of swimming speed (see Fig. 2, Eqn 1, Table 1). Steady swimming cost was based on measurements up to the swimming speed where burst-assisted swimming occurred (see Fig. 1).

alternative. Differences between predicted values, using Eqn 1, and the actual observations involving unsteady swimming were aggregated for each fish and the mean difference was used as the end point value. These calculations showed that for all 15 fish performing unsteady swimming, the mean difference was positive (i.e. higher mean \dot{M}_{O_2} than expected). Testing the data using the sign test revealed a highly significant result ($P < 0.001$), showing that the metabolic rate increased after transition to burst-assisted swimming.

The $\dot{M}_{\text{O}_2\text{max}}$ was $1270.69 \pm 40.50 \text{ mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$. This value was recorded as the highest \dot{M}_{O_2} measured during the swimming protocol (McKenzie et al., 2003). In four fish, the maximum metabolic rate was not associated with the highest swimming speed, but with the second highest swimming speed (Fig. 2). Thus, the mean \dot{M}_{O_2} recorded during the highest swimming speed ($1258.76 \pm 39.73 \text{ mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$) was slightly lower (1%) than $\dot{M}_{\text{O}_2\text{max}}$.

The metabolic scope (MS) was calculated as $\dot{M}_{\text{O}_2\text{max}} - \dot{M}_{\text{O}_2\text{std}}$ following Farrell and Richards (Farrell and Richards, 2009) and was on average $952.64 \text{ mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$. Depending on the fish, $\dot{M}_{\text{O}_2\text{max}}$ occurred during steady or unsteady swimming (Fig. 2). The highest \dot{M}_{O_2} recorded during steady swimming was on average $1015.61 \text{ mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$. This measure included $\dot{M}_{\text{O}_2\text{max}}$ of three individuals that did not perform any burst-assisted swimming (Figs 1, 2). The \dot{M}_{O_2} increased by $255.08 \text{ mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$ during the part of the swimming protocol that involved unsteady swimming (Fig. 2). In proportions of the MS, steady swimming accounted for 73.2%, whereas unsteady swimming accounted for 26.8%. These findings showed that unsteady swimming contributed significantly to MS.

Immediately after fatigue, the swimming speed was reduced to the acclimation speed (2 BL s^{-1}), following Lee and colleagues (Lee et al., 2003b). Starting at the \dot{M}_{O_2} recorded during the highest swimming speed ($1258.76 \pm 39.73 \text{ mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$), post-exercise \dot{M}_{O_2} declined rapidly and approached the pre-exercise \dot{M}_{O_2} (Fig. 6). The first measure of post-exercise \dot{M}_{O_2} was significantly higher than the pre-exercise \dot{M}_{O_2} ($P < 0.001$), providing evidence of EPOC and anaerobic power production in *P. reticulata* (Fig. 6). The rapidly declining \dot{M}_{O_2} , and the fact that we had no \dot{M}_{O_2} data between 0 and

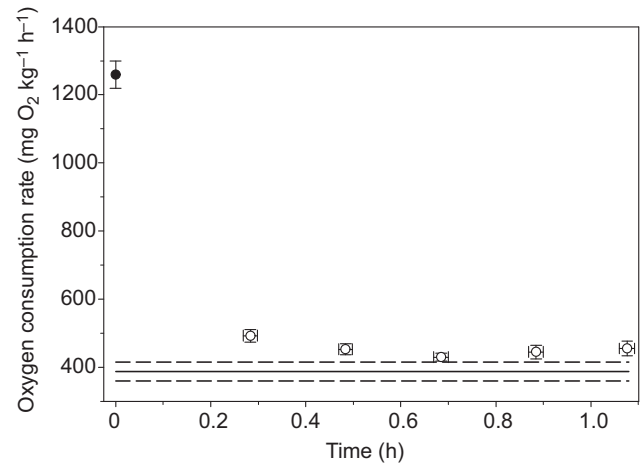


Fig. 6. Oxygen consumption rate (\dot{M}_{O_2} ; $\text{mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$) at the highest recorded swimming speed (closed circle), and post-exercise \dot{M}_{O_2} during the subsequent recovery phase (open circles). The swimming speed was adjusted to 2 BL s^{-1} (total body lengths per second) during the recovery phase. Pre-exercise \dot{M}_{O_2} at 2 BL s^{-1} is indicated using a solid line (dashed lines are 95% CL).

0.2 h (Fig. 6), precluded an accurate estimation of EPOC ($\text{mg O}_2 \text{ kg}^{-1}$). Post-exercise \dot{M}_{O_2} declined until 0.69 h and approached the pre-exercise \dot{M}_{O_2} . The last two measurements of post-exercise \dot{M}_{O_2} at 0.89 and 1.08 h remained slightly elevated relative to the pre-exercise \dot{M}_{O_2} (Fig. 6). The majority of the post-exercise decline in the \dot{M}_{O_2} occurred within 0.3 h after the swimming speed was returned to the acclimation speed (Fig. 6).

DISCUSSION

Contrary to predictions, we found no evidence of correlations between reproductive traits and steady swimming cost, $\dot{M}_{\text{O}_2\text{std}}$ or prolonged swimming performance (U_{crit}). In contrast, pectoral fin use correlated positively with swimming cost and negatively with U_{crit} . We suggest that the use of pectoral fins indicated a mechanism to maintain swimming stability, rather than generate forward thrust. Further, we propose that elevated use of pectoral fins indicated an elevated need to support swimming stability resulting in increased swimming cost and therefore decreased U_{crit} . Finally, we found that the aerobic metabolic rate increased after transition to burst-assisted swimming suggesting that unsteady swimming constituted 26.8% of the MS.

Although pregnancy may influence metabolic rates and swimming performance in live-bearing fish, this study found no evidence of RA or pregnancy stage correlating with $\dot{M}_{\text{O}_2\text{std}}$, steady swimming cost or U_{crit} in wild *P. reticulata* from a low-predation river. A number of studies have reported elevated metabolic rate (Webb and Brett, 1972; Boehlert et al., 1991; Hopkins et al., 1995; Timmerman and Chapman, 2003; Skov et al., 2010) and diminished swimming performance (Plaut, 2002; Ghalambor et al., 2004; Belk and Tuckfield, 2010) in pregnant live-bearing fish. The studies differ from the present study in a number of ways. Firstly, two previous studies tested pregnancy effects on fast-start swimming performance (Ghalambor et al., 2004; Belk and Tuckfield, 2010) rather than U_{crit} . It is possible that fast-start swimming performance is more sensitive to pregnancy than U_{crit} . Secondly, previous studies followed individual fish over the course of the gestation period for repeated measurements (Webb and Brett, 1972; Plaut, 2002; Timmerman and Chapman, 2003) or made comparisons between gestating females and reproductively inactive females or males (Boehlert et al., 1991;

Hopkins et al., 1995; Skov et al., 2010). We were unable to make repeated measurements on individual fish because of the destructive nature of measuring RA, and our samples included no reproductively inactive fish (all wet RA $\geq 4.93\%$). Finally, our study examined low predation *P. reticulata* only. It is well known, however, that high-predation *P. reticulata* (e.g. from the Aripo River) have considerably higher RA than low-predation *P. reticulata* (Reznick and Endler, 1982). Further studies should test for pregnancy effects on metabolic rates and U_{crit} in both high- and low-predation fish, while also controlling for age and genetic background (Ghalambor et al., 2004; Belk and Tuckfield, 2010).

Previous studies have covered the energetics of gait transitions from (1) exclusive pectoral fin propulsion to combined pectoral and caudal fin propulsion (Korsmeyer et al., 2002; Cannas et al., 2006; Jones et al., 2007; Kendall et al., 2007; Svendsen et al., 2010); (2) steady swimming to unsteady swimming (Svendsen et al., 2010); (3) dorsal and anal fin propulsion to caudal fin propulsion (Korsmeyer et al., 2002); and (4) free-stream swimming to Karman gaiting (Liao, 2007; Taguchi and Liao, 2011). By contrast, little attention has been devoted to the energetic effects of fin use variation within a single gait. The present study found that within the steady BCF swimming gait, swimming cost correlated strongly with pectoral fin use. Fish that ceased using the pectoral fins at low speeds reduced swimming cost in comparison with fish that ceased using the pectoral fins at a higher speed or not at all. According to previous studies, BCF swimmers extending their paired fins should experience increased swimming costs (Webb, 1998; Webb, 2002) because of the additional drag (Videler and Wardle, 1991; Weihs, 2002; Green and Hale, 2012); however, this hypothesis has rarely been tested. Although furling of the pectoral fins at relatively low speeds is common (Drucker and Lauder, 2003), some BCF swimmers employ both caudal and pectoral fins at relatively high swimming speeds. For example, in the field, brook trout (*Salvelinus fontinalis*) combine the use of the caudal and pectoral fins at a wide range of swimming speeds (McLaughlin and Noakes, 1998). Notably, *S. fontinalis* using their pectoral fins swim with a higher caudal fin beat frequency at a given swimming speed than those not using their pectoral fins (McLaughlin and Noakes, 1998). Because there is a positive relationship between caudal fin beat frequency and \dot{M}_{O_2} (Ohlberger et al., 2007), these findings indicate that *S. fontinalis* using the pectoral fins experienced increased swimming cost. The observations on *S. fontinalis* are consistent with the present study, demonstrating a positive relationship between pectoral fin use and steady swimming cost in *P. reticulata*. Our data suggest that combining the caudal and pectoral fins over a wide speed range is an inefficient BCF swimming behaviour.

What proximate mechanism could underpin the observed intraspecific variation in pectoral fin use? A likely mechanism involves variable needs to support swimming stability and control. In BCF swimmers, pectoral fins are not used for forward thrust generation, but play an important role as trimming and/or powered correction systems to maintain swimming stability (Webb, 2002). The former involves positioning the fins to dampen or correct perturbations, whereas the latter involves active movements of the fins independent of the body to correct perturbations (Webb, 2002). Stability and control can be a major problem in swimming (Videler and Wardle, 1991; Webb, 1998; Webb, 2002). For example, there are six possible recoil motions for a rigid body resulting from propulsor movements, three of them translational and three rotational (Hove et al., 2001). BCF swimming generates large side forces that cause the anterior parts of the body to recoil (yaw and/or sideslip) (Hove et al., 2001; Weihs, 2002; Lauder, 2006), which may

represent a major stability problem in BCF swimming (Webb, 1988; Weihs, 2002). The yaw movements generated by the caudal fin are usually countered by movements of the pectoral or pelvic fins (Lauder, 2006). Such needs for stability control by balancing forces have led recent studies to emphasize the importance of multiple fins employed by swimming fish (Hove et al., 2001; Drucker et al., 2006; Lauder and Tytell, 2006; Tytell et al., 2008; Blake et al., 2009). The use of paired fins to maintain stability and control is most pronounced at lower speeds. At higher swimming speeds, various stability problems persist, but control is shifted more towards the body-caudal fin. In the present study, individual *P. reticulata* employed the pectoral fins at a variable speed range, and increased pectoral fin use was associated with increased steady swimming costs and a low U_{crit} . Although the exact function of extending the pectoral fins remains unknown, it is likely that the variation in pectoral fin use reflected, at least partly, different needs to balance forces and support swimming stability and control. According to this hypothesis, *P. reticulata* that continued using the pectoral fins at high swimming speeds did so to support swimming stability. As such, the extent of pectoral fin use at increasing speeds could be interpreted as an index of swimming stability in individual fish, with extensive pectoral fin use indicating a swimmer with stability problems.

What is the metabolic cost of stability control in swimming fishes? The metabolic cost of stability control is not known (Lauder, 2006), but it likely represents a significant part of the total swimming cost (Webb, 2002). In the present study, the pectoral fins were presumably not employed to generate forward thrust, but to support stability control. Using the pectoral fins as a trimming and/or powered correction system to stabilize BCF swimming should increase swimming costs (Webb, 1998; Weihs, 2002) because of the increased lateral surface and hence additional drag (Videler and Wardle, 1991; Weihs, 2002; Green and Hale, 2012). Correspondingly, we suggest that the positive relationship between pectoral fin use and steady swimming cost reflected, at least partially, the cost of stability control incurred when using the pectoral fins as a trimming and/or powered correction system. In the same vein, the negative relationship between pectoral fin use and U_{crit} could be explained by the fact that pectoral fin use correlated positively with swimming cost. Fish that made extensive use of the pectoral fins exhibited a significant increase in the swimming cost, which may have resulted in inferior swimming performance, because the fish reached the maximum metabolic rate at a relatively slow swimming speed.

Many fish species transition from steady to unsteady swimming at increasing swimming speeds. Few studies, however, have quantified the metabolic rate associated with burst-assisted swimming. Metabolic rate studies concerned with burst-assisted swimming at high speeds remain challenged by the facts that the gait can be maintained for only a short period of time (Farrell, 2007) and involves anaerobic metabolism (Burgetz et al., 1998; Lee et al., 2003b; Svendsen et al., 2010), which may complicate the measurements (Farrell, 2007; Ellerby, 2010). Anaerobic metabolism is evidenced by the activation of white muscles and the occurrence of glycolysis followed by EPOC (Burgetz et al., 1998; Lee et al., 2003b; Farrell, 2007; Svendsen et al., 2010). Farrell reviewed past studies and discussed the paradox that the relationship between swimming speed and aerobic \dot{M}_{O_2} often is exponential and not sigmoidal, as predicted by the anaerobic influence on the total metabolic cost (Farrell, 2007). The exponential relationship may be explained by a number of factors, including white muscles working in a partially aerobic fashion, and pooling fish that vary considerably

in their individual U_{crit} values, as this would tend to obscure any individual plateaus in \dot{M}_{O_2} (Farrell, 2007). The present study examined the relationship between swimming speed and \dot{M}_{O_2} up to U_{crit} in individual fish and found no evidence of a sigmoidal relationship. Instead, \dot{M}_{O_2} continued to increase during burst-assisted swimming, and the data points during unsteady swimming were higher than what would be expected based on extrapolation of the values representing steady swimming. Correspondingly, unsteady swimming constituted 26.8% of the MS. The absence of \dot{M}_{O_2} plateauing during unsteady swimming at increasing speeds suggests that anaerobic metabolism played a limited role in fuelling the swimming, even close to U_{crit} . This inference is consistent with past studies suggesting limited anaerobic capacity (Kieffer, 2000) and dependence on anaerobic power production during swimming (Goolish, 1991) in small fish (<10 cm in body length). In contrast, we did find evidence of EPOC, indicating that anaerobic power production occurred during the swimming trial. The majority of the post-exercise metabolic decline occurred within 0.3 h. Previous studies measuring metabolic recovery after U_{crit} tests have reported recovery times from ~0.2 to 4 h (Brett, 1964; Bushnell et al., 1994; Reidy et al., 1995; Lee et al., 2003a; Lee et al., 2003b). Scaling relationships between body size and the partitioning of aerobic and anaerobic power production in swimming fish remain an important future avenue of empirical research.

In many terrestrial animals, gait transitions reduce the metabolic cost of locomotion (Griffin et al., 2004; Rubenson et al., 2004; Nudds et al., 2011), but the proximate mechanism driving the transition may not be metabolic *per se*, but rather related to mechanical factors, such as musculoskeletal force (Farley and Taylor, 1991) and bone strain (Biewener and Taylor, 1986). Compared with terrestrial locomotion, gait transitions in aquatic locomotion remain poorly understood. The present study analysed metabolic consequences of the gait transition from steady to unsteady BCF swimming and found that the metabolic rate increased after the transition. These data raise the question of why an animal would perform a transition to a gait that is less energy efficient. In labriform swimmers, it has been suggested that the pectoral–caudal gait transition is driven by a need to supply additional mechanical power rather than to minimize metabolic swimming costs (Alexander, 1989; Korsmeyer et al., 2002; Cannas et al., 2006; Jones et al., 2007; Kendall et al., 2007). Only small amounts of muscle can be packed around the paired fins while larger amounts can be accommodated about the axial skeleton driving caudal propulsion (Webb, 1998). The transition from steady to unsteady BCF swimming could have a similar mechanistic basis, because additional mechanical power from white muscle fibres may be available after transition to burst-assisted swimming. However, this remains speculation, and additional studies of muscle recruitment patterns at increasing speeds combined with aerobic and anaerobic metabolic rates of disparate muscle types are required to evaluate the hypothesis.

The U_{crit} protocol provides a measure of physiological endurance capacity, initially used in fisheries sciences (Brett, 1964; Beamish, 1978). Recently, studies have started to examine U_{crit} in ecological and evolutionary contexts (Claireaux et al., 2007; Oufiero et al., 2011; Dalziel and Schulte, 2012; Dalziel et al., 2012a; Dalziel et al., 2012b). Using individual fish, the present study demonstrated a relationship between pectoral fin use, steady swimming cost and U_{crit} . Fish with a low U_{crit} spent more time with extended pectoral fins and exhibited increased steady swimming cost. We found no evidence that \dot{M}_{O_2max} correlated with U_{crit} . These findings indicate that U_{crit} reflects not only the physiological endurance capacity of individual fish, but indeed also the biomechanical performance.

This study shows that fish with a low U_{crit} may spend more energy on swimming, in comparison with fish with a high U_{crit} , because the former fish have higher swimming costs during steady swimming. This may be particularly relevant in species living in lotic habitats. For example, Nelson and colleagues reported a positive relationship between home-stream current velocity and U_{crit} in blacknose dace (*Rhinichthys atratulus*) (Nelson et al., 2003). Our finding that U_{crit} correlates negatively with steady swimming costs indicates that not only will a high U_{crit} allow fish to traverse fast-flowing riffles without fatigue, as suggested by Nelson et al. (Nelson et al., 2003), it also implies lower steady swimming cost at current velocities that represent sub-maximal swimming speeds. Thus, because of superior biomechanical performance, a high U_{crit} may allow fish to inhabit relatively high stream current velocities and yet have relatively low swimming costs. As such, the observed intraspecific variation in pectoral fin use, swimming cost and U_{crit} could have implications for habitat use in individual fish. For example, Ellerby and Gerry showed that habitat use varies with individual differences in energy economy, steady-state swimming and manoeuvrability in bluegill sunfish (*Lepomis macrochirus*) (Ellerby and Gerry, 2011). Although *P. reticulata* often occupy distinct pools in rivers, it remains to be tested whether intraspecific variation in pectoral fin use, swimming cost and U_{crit} influence habitat use within pools and to what degree such relationships (or lack of) affect daily energy expenditures (Careau and Garland, 2012).

In summary, this study found that elevated pectoral fin use is associated with increased swimming cost and decreased U_{crit} . It is unclear why some *P. reticulata* continued using the pectoral fins while others quickly ceased using them at increasing swimming speeds. We propose that use of the pectoral fins is related to stability and control rather than generation of forward thrust. Extending the pectoral fins may help maintain swimming stability, but it comes with increased swimming cost, which in turn is associated with reduced U_{crit} . The causes and consequences of variation in pectoral fin use remain, however, poorly understood and further study is warranted. Finally, we found that the aerobic metabolic rate increased after transition to burst-assisted swimming, and unsteady swimming constituted 26.8% of the MS.

LIST OF SYMBOLS AND ABBREVIATIONS

a	oxygen consumption rate at zero swimming speed
\hat{a}_j	estimated fish-specific oxygen consumption rate at zero swimming speed
a_j	fish-specific oxygen consumption rate at zero swimming speed
a'_j	fish-specific log oxygen consumption rate at zero swimming speed
AR1	autoregressive process of order 1
b	rate of increase in the oxygen consumption rate as a function of swimming speed (an index of swimming cost)
\hat{b}_j	estimated fish-specific rate of increase in the oxygen consumption rate as a function of swimming speed
b_j	fish-specific rate of increase in the oxygen consumption rate as a function of swimming speed
BCF	body-caudal fin
BL	total body length
e_{ij}	autocorrelated residuals
EPOC	excess post-exercise oxygen consumption
\dot{M}_{O_2}	metabolic rate
\dot{M}_{O_2ij}	metabolic rate for the i th observation on the j th fish
\dot{M}_{O_2max}	maximum metabolic rate
$\dot{M}_{O_2max,j}$	fish-specific maximum metabolic rate
\dot{M}_{O_2std}	standard metabolic rate
MS	metabolic scope
RA	reproductive allocation
RA_j	fish-specific reproductive allocation

U	swimming speed
U_{crit}	critical swimming speed
$U_{crit,j}$	fish-specific critical swimming speed
U_{STmax}	gait transition speed from steady to unsteady swimming
ϵ_{ij}	uncorrelated residuals assumed to be independently and identically normally distributed
μ_a	mean fish-specific log oxygen consumption rate at zero swimming speed
μ_b	mean fish-specific rate of increase in the oxygen consumption rate as a function of swimming speed
ρ	correlation between the fish-specific log oxygen consumption rate at zero swimming speed (a'_j) and the fish-specific rate of increase in the oxygen consumption rate as a function of swimming speed (b_j)
σ_a^2	variance of the fish-specific log oxygen consumption rate at zero swimming speed
σ_b^2	variance of the fish-specific rate of increase in the oxygen consumption rate as a function of swimming speed
σ_ϵ^2	variance of the uncorrelated residuals (ϵ_{ij})
ϕ	autoregressive (AR1) parameter

ACKNOWLEDGEMENTS

We thank C. Jørgensen for help with conversion factors, and J. Laustsen for recovering the data. We thank C. Hoover and D. Callaghan for help with R; and R. D. Bassar, C. E. Oufiero and A. T. Silva for helpful comments on an earlier version of the manuscript. We thank two anonymous reviewers for their helpful and constructive comments. We thank A. B. Dydensborg and K. Nilsson for help with the fieldwork.

AUTHOR CONTRIBUTIONS

All five authors contributed to the conception and design of the study. J.C.S. and A.I.B. collected the data. R.H.B.C. and J.C.S. analysed the data. All five authors contributed to the interpretation of the data and the drafting and revising of the article.

COMPETING INTERESTS

No competing interests declared.

FUNDING

This research was supported by a grant from the Danish Research Council to the research school SLIP and the Fishnet Research Network and allocated to J.C.S. and J.F.S. The Idella Foundation provided travel funding to J.C.S. The research was also funded by the National Science Foundation (DDIG DEB-0710185 to D. N. Reznick and A.I.B.).

REFERENCES

- Alexander, R. M. (1989). Optimization and gaits in the locomotion of vertebrates. *Physiol. Rev.* **69**, 1199-1227.
- Arnott, S. A., Chiba, S. and Conover, D. O. (2006). Evolution of intrinsic growth rate: metabolic costs drive trade-offs between growth and swimming performance in *Menidia menidia*. *Evolution* **60**, 1269-1278.
- Beamish, F. W. H. (1978). Swimming capacity. In *Fish Physiology*, Vol. VII (ed. W. S. Hoar and D. J. Randall), pp. 101-187. New York, NY: Academic Press.
- Belk, M. C. and Tuckfield, R. C. (2010). Changing costs of reproduction: age-based differences in reproductive allocation and escape performance in a livebearing fish. *Oikos* **119**, 163-169.
- Bell, W. M. and Terhune, L. D. B. (1970). *Water Tunnel Design for Fisheries Research*. Nanaimo, BC: Fisheries Research Board of Canada, Biological Station.
- Berger, J. (1991). Pregnancy incentives, predation constraints and habitat shifts: experimental and field evidence for wild bighorn sheep. *Anim. Behav.* **41**, 61-77.
- Biewener, A. A. and Taylor, C. R. (1986). Bone strain: a determinant of gait and speed? *J. Exp. Biol.* **123**, 383-400.
- Birchard, G. F., Black, C. P., Schuett, G. W. and Black, V. (1984). Influence of pregnancy on oxygen consumption, heart rate and hematology in the garter snake: implications for the 'cost of reproduction' in live bearing reptiles. *Comp. Biochem. Physiol.* **77A**, 519-523.
- Biro, P. A. and Stamps, J. A. (2010). Do consistent individual differences in metabolic rate promote consistent individual differences in behavior? *Trends Ecol. Evol.* **25**, 653-659.
- Blake, R. W., Li, J. and Chan, K. H. S. (2009). Swimming in four goldfish *Carassius auratus* morphotypes: understanding functional design and performance employing artificially selected forms. *J. Fish Biol.* **75**, 591-617.
- Boehlert, G. W., Kusakari, M. and Yamada, J. (1991). Oxygen consumption of gestating female *Sebastes schlegelii*: estimating the reproductive costs of livebearing. *Environ. Biol. Fishes* **30**, 81-89.
- Bolinick, D. I., Svanbäck, R., Fordyce, J. A., Yang, L. H., Davis, J. M., Hulseley, C. D. and Forister, M. L. (2003). The ecology of individuals: incidence and implications of individual specialization. *Am. Nat.* **161**, 1-28.
- Brett, J. R. (1964). The respiratory metabolism and swimming performance of young sockeye salmon. *J. Fish. Res. Board Can.* **21**, 1183-1226.
- Burgetz, I. J., Rojas-Vargas, A., Hinch, S. G. and Randall, D. J. (1998). Initial recruitment of anaerobic metabolism during sub-maximal swimming in rainbow trout (*Oncorhynchus mykiss*). *J. Exp. Biol.* **201**, 2711-2721.
- Burton, T., Killen, S. S., Armstrong, J. D. and Metcalfe, N. B. (2011). What causes intraspecific variation in resting metabolic rate and what are its ecological consequences? *Proc. Biol. Sci.* **278**, 3465-3473.
- Bushnell, P. G., Steffensen, J. F., Schurmann, H. and Jones, D. R. (1994). Exercise metabolism in two species of cod in arctic waters. *Polar Biol.* **14**, 43-48.
- Cannas, M., Schaefer, J., Domenici, P. and Steffensen, J. F. (2006). Gait transition and oxygen consumption in swimming striped surf perch (*Embiotoca lateralis*). *J. Fish Biol.* **69**, 1612-1625.
- Careau, V. and Garland, T., Jr (2012). Performance, personality, and energetics: correlation, causation, and mechanism. *Physiol. Biochem. Zool.* **85**, 543-571.
- Chen, W.-J., Bonillo, C. and Lecointre, G. (2003). Repeatability of clades as a criterion of reliability: a case study for molecular phylogeny of Acanthomorpha (Teleostei) with larger number of taxa. *Mol. Phylogenet. Evol.* **26**, 262-288.
- Claireaux, G., Couturier, C. and Groison, A.-L. (2006). Effect of temperature on maximum swimming speed and cost of transport in juvenile European sea bass (*Dicentrarchus labrax*). *J. Exp. Biol.* **209**, 3420-3428.
- Claireaux, G., Handelsman, C., Standen, E. and Nelson, J. A. (2007). Thermal and temporal stability of swimming performance in the European sea bass. *Physiol. Biochem. Zool.* **80**, 186-196.
- Dalziel, A. C. and Schulte, P. M. (2012). Correlates of prolonged swimming performance in F2 hybrids of migratory and non-migratory threespine stickleback. *J. Exp. Biol.* **215**, 3587-3596.
- Dalziel, A. C., Vines, T. H. and Schulte, P. M. (2012a). Reductions in prolonged swimming capacity following freshwater colonization in multiple threespine stickleback populations. *Evolution* **66**, 1226-1239.
- Dalziel, A. C., Ou, M. and Schulte, P. M. (2012b). Mechanisms underlying parallel reductions in aerobic capacity in non-migratory threespine stickleback (*Gasterosteus aculeatus*) populations. *J. Exp. Biol.* **215**, 746-759.
- DeMarco, V. (1993). Metabolic rates of female viviparous lizards (*Sceloporus jarrovi*) throughout the reproductive cycle: do pregnant lizards adhere to standard allometry? *Physiol. Zool.* **66**, 166-180.
- Drucker, E. G. and Lauder, G. V. (2003). Function of pectoral fins in rainbow trout: behavioral repertoire and hydrodynamic forces. *J. Exp. Biol.* **206**, 813-826.
- Drucker, E. G., Walker, J. A. and Westneat, M. W. (2006). Mechanics of pectoral fin swimming in fishes. In *Fish Biomechanics* (ed. R. E. Shadwick and G. V. Lauder), pp. 369-423. San Diego, CA: Academic Press.
- Ellerby, D. J. (2010). How efficient is a fish? *J. Exp. Biol.* **213**, 3765-3767.
- Ellerby, D. J. and Gerry, S. P. (2011). Sympatric divergence and performance trade-offs of bluegill ecomorphs. *Evol. Biol.* **38**, 422-433.
- Farley, C. T. and Taylor, C. R. (1991). A mechanical trigger for the trot-gallop transition in horses. *Science* **253**, 306-308.
- Farrell, A. P. (2007). Cardiorespiratory performance during prolonged swimming tests with salmonids: a perspective on temperature effects and potential analytical pitfalls. *Philos. Trans. R. Soc. B* **362**, 2017-2030.
- Farrell, A. P. and Richards, J. G. (2009). Defining hypoxia: an integrative synthesis of the responses of fish to hypoxia. In *Hypoxia*, Vol. 27 (ed. J. G. Richards, A. P. Farrell and C. J. Brauner), pp. 488-504. London: Academic Press.
- Ghalambor, C. K., Walker, J. A. and Reznick, D. N. (2003). Multi-trait selection, adaptation, and constraints on the evolution of burst swimming performance. *Integr. Comp. Biol.* **43**, 431-438.
- Ghalambor, C. K., Reznick, D. N. and Walker, J. A. (2004). Constraints on adaptive evolution: the functional trade-off between reproduction and fast-start swimming performance in the Trinidadian guppy (*Poecilia reticulata*). *Am. Nat.* **164**, 38-50.
- Goollish, E. M. (1991). Aerobic and anaerobic scaling in fish. *Biol. Rev. Camb. Philos. Soc.* **66**, 33-56.
- Green, M. H. and Hale, M. E. (2012). Activity of pectoral fin motoneurons during two swimming gaits in the larval zebrafish (*Danio rerio*) and localization of upstream circuit elements. *J. Neurophysiol.* **108**, 3393-3402.
- Griffin, T. M., Kram, R., Wickler, S. J. and Hoyt, D. F. (2004). Biomechanical and energetic determinants of the walk-trot transition in horses. *J. Exp. Biol.* **207**, 4215-4223.
- Hayes, J. P. and Jenkins, S. H. (1997). Individual variation in mammals. *J. Mammal.* **78**, 274-293.
- Haynes, J. (1995). Standardized classification of poeciliid development for life history studies. *Copeia* **1995**, 147-154.
- Hopkins, T. E., Eldridge, M. B. and Cech, J. J., Jr (1995). Metabolic costs of viviparity in yellowtail rockfish, *Sebastes flavidus*. *Environ. Biol. Fishes* **43**, 77-84.
- Hove, J. R., O'Bryan, L. M., Gordon, M. S., Webb, P. W. and Weihs, D. (2001). Boxfishes (Teleostei: Ostraciidae) as a model system for fishes swimming with many fins: kinematics. *J. Exp. Biol.* **204**, 1459-1471.
- James, R. S. and Johnston, I. A. (1998). Influence of spawning on swimming performance and muscle contractile properties in the short-horn sculpin. *J. Fish Biol.* **53**, 485-501.
- Jones, E. A., Lucey, K. S. and Ellerby, D. J. (2007). Efficiency of labriform swimming in the bluegill sunfish (*Lepomis macrochirus*). *J. Exp. Biol.* **210**, 3422-3429.
- Kendall, J. L., Lucey, K. S., Jones, E. A., Wang, J. and Ellerby, D. J. (2007). Mechanical and energetic factors underlying gait transitions in bluegill sunfish (*Lepomis macrochirus*). *J. Exp. Biol.* **210**, 4265-4271.
- Kieffer, J. D. (2000). Limits to exhaustive exercise in fish. *Comp. Biochem. Physiol.* **126A**, 161-179.
- Korsmeyer, K. E., Steffensen, J. F. and Herskin, J. (2002). Energetics of median and paired fin swimming, body and caudal fin swimming, and gait transition in parrotfish (*Scarus schlegelii*) and triggerfish (*Rhinecanthus aculeatus*). *J. Exp. Biol.* **205**, 1253-1263.
- Lauder, G. V. (2006). Locomotion. In *The Physiology of Fishes*, 3rd edn (ed. D. H. Evans and J. B. Claiborne), pp. 3-46. Boca Raton, FL: CRC Press.

- Lauder, G. V. and Tytell, E. D. (2006). Hydrodynamics of undulatory propulsion. In *Fish Biomechanics* (ed. R. E. Shadwick and G. V. Lauder), pp. 425-468. San Diego, CA: Academic Press.
- Lee, C. G., Devlin, R. H. and Farrell, A. P. (2003a). Swimming performance, oxygen consumption and excess post-exercise oxygen consumption in adult transgenic and ocean-ranched coho salmon. *J. Fish Biol.* **62**, 753-766.
- Lee, C. G., Farrell, A. P., Lotto, A., Hinch, S. G. and Healey, M. C. (2003b). Excess post-exercise oxygen consumption in adult sockeye (*Oncorhynchus nerka*) and coho (*O. kisutch*) salmon following critical speed swimming. *J. Exp. Biol.* **206**, 3253-3260.
- Liao, J. C. (2007). A review of fish swimming mechanics and behavior in altered flows. *Philos. Trans. R. Soc. B* **362**, 1973-1993.
- Magurran, A. E. (2005). *Evolutionary Ecology: The Trinidadian Guppy*. Oxford: Oxford University Press.
- Marras, S., Claireaux, G., McKenzie, D. J. and Nelson, J. A. (2010). Individual variation and repeatability in aerobic and anaerobic swimming performance of European sea bass, *Dicentrarchus labrax*. *J. Exp. Biol.* **213**, 26-32.
- Marras, S., Killen, S. S., Claireaux, G., Domenici, P. and McKenzie, D. J. (2011). Behavioural and kinematic components of the fast-start escape response in fish: individual variation and temporal repeatability. *J. Exp. Biol.* **214**, 3102-3110.
- Marsh-Matthews, E., Brooks, M., Deaton, R. and Tan, H. (2005). Effects of maternal and embryo characteristics on post-fertilization provisioning in fishes of the genus *Gambusia*. *Oecologia* **144**, 12-24.
- Marsh-Matthews, E., Deaton, R. and Brooks, M. (2011). Survey of matrotrophy in lecithotrophic poeciliids. In *Viviparous Fishes II. The Proceedings of the III International Symposium on Viviparous Fishes* (ed. M. C. Uribe and H. J. Grier), pp. 1-4. Homestead, FL: New Life Publications.
- McKenzie, D. J., Martinez, R., Morales, A., Acosta, J., Morales, R., Taylor, E. W., Steffensen, J. F. and Estrada, M. P. (2003). Effects of growth hormone transgenesis on metabolic rate, exercise performance and hypoxia tolerance in tilapia hybrids. *J. Fish Biol.* **63**, 398-409.
- McLaughlin, R. L. and Noakes, D. L. G. (1998). Going against the flow: an examination of the propulsive movements made by young brook trout in streams. *Can. J. Fish. Aquat. Sci.* **55**, 853-860.
- Miles, D. B., Sinervo, B. and Anthony Frankino, W. (2000). Reproductive burden, locomotor performance, and the cost of reproduction in free ranging lizards. *Evolution* **54**, 1386-1395.
- Nelson, J. A., Gotwalt, P. S. and Snodgrass, J. W. (2003). Swimming performance of blacknose dace (*Rhinichthys atratulus*) mirrors home-stream current velocity. *Can. J. Fish. Aquat. Sci.* **60**, 301-308.
- Nudds, R. L., Folkow, L. P., Lees, J. J., Tickle, P. G., Stokkan, K. A. and Codd, J. R. (2011). Evidence for energy savings from aerial running in the Svalbard rock ptarmigan (*Lagopus muta hyperborea*). *Proc. Biol. Sci.* **278**, 2654-2661.
- Ohlberger, J., Staaks, G. and Hölker, F. (2007). Estimating the active metabolic rate (AMR) in fish based on tail beat frequency (TBF) and body mass. *J. Exp. Zool. A* **307**, 296-300.
- Oufiero, C. E. and Garland, T., Jr (2009). Repeatability and correlation of swimming performances and size over varying time-scales in the guppy (*Poecilia reticulata*). *Funct. Ecol.* **23**, 969-978.
- Oufiero, C. E., Walsh, M. R., Reznick, D. N. and Garland, T., Jr (2011). Swimming performance trade-offs across a gradient in community composition in Trinidadian killifish (*Rivulus hartii*). *Ecology* **92**, 170-179.
- Peake, S. J. (2008). Gait transition as an alternate measure of maximum aerobic capacity in fishes. *J. Fish Biol.* **72**, 645-655.
- Pinheiro, J. and Bates, D. DeRoy, S., Sarkar, D. and the R Development Core Team (2011). nlme: linear and nonlinear mixed effects models. R package version 3.1-101. Vienna, Austria: R Foundation for Statistical Computing.
- Plaut, I. (2002). Does pregnancy affect swimming performance of female mosquitofish, *Gambusia affinis*? *Funct. Ecol.* **16**, 290-295.
- Pohlmann, K., Grasso, F. W. and Breithaupt, T. (2001). Tracking wakes: the nocturnal predatory strategy of piscivorous catfish. *Proc. Natl. Acad. Sci. USA* **98**, 7371-7374.
- Poulsen, S. B., Jensen, L. F., Schulz, C., Deacon, M., Meyer, K. E., Jäger-Kleinicke, T., Schwarten, H. and Svendsen, J. C. (2012). Ontogenetic differentiation of swimming performance and behaviour in relation to habitat availability in the endangered North Sea houting (*Coregonus oxyrinchus*). *Aquat. Living Resour.* **25**, 241-249.
- R Development Core Team (2011). R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing.
- Reidy, S. P., Nelson, J. A., Tang, Y. and Kerr, S. R. (1995). Post-exercise metabolic rate in Atlantic cod and its dependence upon the method of exhaustion. *J. Fish Biol.* **47**, 377-386.
- Reznick, D. N. (1983). The structure of guppy life histories: the tradeoff between growth and reproduction. *Ecology* **64**, 862-873.
- Reznick, D. N. (1989). Life history evolution in guppies: 2. Repeatability of field observations and the effects of season on life histories. *Evolution* **43**, 1285-1297.
- Reznick, D. N. and Endler, J. A. (1982). The impact of predation on life history evolution in Trinidadian guppies (*Poecilia reticulata*). *Evolution* **36**, 160-177.
- Reznick, D. N. and Yang, A. P. (1993). The influence on fluctuating resources on life history: patterns of allocation and plasticity in female guppies. *Ecology* **74**, 2011-2019.
- Reznick, D. N., Bryga, H. and Endler, J. A. (1990). Experimentally induced life history evolution in a natural population. *Nature* **346**, 357-359.
- Reznick, D. N., Butler, M. J., Rodd, F. H. and Ross, P. (1996). Life-history evolution in guppies (*Poecilia reticulata*): 6. Differential mortality as a mechanism for natural selection. *Evolution* **50**, 1651-1660.
- Rubenson, J., Heliam, D. B., Lloyd, D. G. and Fournier, P. A. (2004). Gait selection in the ostrich: mechanical and metabolic characteristics of walking and running with and without an aerial phase. *Proc. R. Soc. B* **271**, 1091-1099.
- Sears, M. W., Hayes, J. P., Banta, M. R. and McCormick, D. (2009). Out in the cold: physiological capacity influences behaviour in deer mice. *Funct. Ecol.* **23**, 774-783.
- Skov, P. V., Steffensen, J. F., Sørensen, T. F. and Qvortrup, K. (2010). Embryonic suckling and maternal specializations in the live-bearing teleost *Zoarces viviparus*. *J. Exp. Mar. Biol. Ecol.* **395**, 120-127.
- Speakman, J. R. (2005). Body size, energy metabolism and lifespan. *J. Exp. Biol.* **208**, 1717-1730.
- Stearns, S. C. (1989). Trade-offs in life-history evolution. *Funct. Ecol.* **3**, 259-268.
- Steffensen, J. F. (1989). Some errors in respirometry of aquatic breathers: how to avoid and correct for them. *Fish Physiol. Biochem.* **6**, 49-59.
- Svendsen, J. C., Tudorache, C., Jordan, A. D., Steffensen, J. F., Aarestrup, K. and Domenici, P. (2010). Partition of aerobic and anaerobic swimming costs related to gait transitions in a labriform swimmer. *J. Exp. Biol.* **213**, 2177-2183.
- Svendsen, J. C., Steffensen, J. F., Aarestrup, K., Frisk, M., Etzerodt, A. and Jyde, M. (2012). Excess posthypoxic oxygen consumption in rainbow trout (*Oncorhynchus mykiss*): recovery in normoxia and hypoxia. *Can. J. Zool.* **90**, 1-11.
- Swanson, C., Young, P. and j. (1998). Swimming performance of delta smelt: maximum performance, and behavioral and kinematic limitations on swimming at submaximal velocities. *J. Exp. Biol.* **201**, 333-345.
- Taguchi, M. and Liao, J. C. (2011). Rainbow trout consume less oxygen in turbulence: the energetics of swimming behaviors at different speeds. *J. Exp. Biol.* **214**, 1428-1436.
- Timmerman, C. M. and Chapman, L. J. (2003). The effect of gestational state on oxygen consumption and response to hypoxia in the sailfin molly, *Poecilia latipinna*. *Environ. Biol. Fishes* **68**, 293-299.
- Tudorache, C., O'Keefe, R. A. and Benfey, T. J. (2011). Optimal swimming speeds reflect preferred swimming speeds of brook charr (*Salvelinus fontinalis* Mitchell, 1874). *Fish Physiol. Biochem.* **37**, 307-315.
- Tytell, E. D., Standen, E. M. and Lauder, G. V. (2008). Escaping Flatland: three-dimensional kinematics and hydrodynamics of median fins in fishes. *J. Exp. Biol.* **211**, 187-195.
- Videler, J. J. and Wardle, C. S. (1991). Fish swimming stride by stride: speed limits and endurance. *Rev. Fish Biol. Fish.* **1**, 23-40.
- Webb, P. W. (1988). Simple physical principles and vertebrate aquatic location. *Am. Zool.* **28**, 709-725.
- Webb, P. W. (1993). Swimming. In *The Physiology of Fishes* (ed. D. H. Evans), pp. 47-73. Boca Raton, FL: CRC Press.
- Webb, P. W. (1998). Swimming. In *The Physiology of Fishes* (ed. D. H. Evans), pp. 3-24. Boca Raton, FL: CRC Press.
- Webb, P. W. (2002). Control of posture, depth, and swimming trajectories of fishes. *Integr. Comp. Biol.* **42**, 94-101.
- Webb, J. K. (2004). Pregnancy decreases swimming performance of female northern death adders (*Acanthopis praelongus*). *Copeia* **2004**, 357-363.
- Webb, P. W. and Brett, J. R. (1972). Oxygen consumption of embryos and parents, and oxygen transfer characteristics within the ovary of two species of viviparous seaperch, *Rhacochilus vacca* and *Embiotoca lateralis*. *J. Fish. Res. Board Can.* **29**, 1543-1553.
- Webb, P. W. and Fairchild, A. G. (2001). Performance and maneuverability of three species of teleostean fishes. *Can. J. Zool.* **79**, 1866-1877.
- Weih, D. (2002). Stability versus maneuverability in aquatic locomotion. *Integr. Comp. Biol.* **42**, 127-134.
- Williams, T. D. (2008). Individual variation in endocrine systems: moving beyond the 'tyranny of the golden mean'. *Philos. Trans. R. Soc. B* **363**, 1687-1698.
- Williams, T. D. (2012). Hormones, life-history, and phenotypic variation: opportunities in evolutionary avian endocrinology. *Gen. Comp. Endocrinol.* **176**, 286-295.