

Trade-offs between speed and endurance in the frog *Xenopus laevis*: a multi-level approach

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Summary

One of the most interesting trade-offs within the vertebrate locomotor system is that between speed and endurance capacity. However, few studies have demonstrated a conflict between whole-animal speed and endurance within a vertebrate species. We investigated the existence of trade-offs between speed and endurance capacity at both the whole-muscle and whole-animal levels in post-metamorphs of the frog *Xenopus laevis*. The burst-swimming performance of 55 frogs was assessed using a high-speed digital camera, and their endurance capacity was measured in a constant-velocity swimming flume.

The work-loop technique was used to assess maximum power production of whole peroneus muscles at a cycle frequency of 6 Hz, while fatigue-resistance was determined by recording the decrease in force and net power production during a set of continuous cycles at 2 Hz. We found no significant correlations between measures of burst swimming performance and endurance capacity,

suggesting that there is no trade-off between these two measures of whole-animal performance. In contrast, there was a significant negative correlation between peak instantaneous power output of the muscles at 6 Hz and the fatigue-resistance of force production at 2 Hz (other correlations between power and fatigue were negative but non-significant). Thus, our data support the suggestion that a physiological conflict between maximum power output and fatigue resistance exists at the level of vertebrate muscles. The apparent incongruence between whole-muscle and whole-animal performance warrants further detailed investigation and may be related to factors influencing both whole-muscle and whole-animal performance measures.

Key words: trade-off, constraint, locomotion, performance, muscle, power output, fatigue resistance, frog, *Xenopus laevis*.

Introduction

An important research objective within the rapidly expanding discipline of evolutionary physiology is a more detailed understanding of the role of physiological constraints in evolution (Garland and Carter, 1994; Feder et al., 2000). Physiological constraints can arise when an organism has to perform multiple tasks that place conflicting demands on its physiological design: an increase in the performance of one task may lead to a decrease in the performance of the other, and an evolutionary trade-off will produce a compromised physiology that is optimal for neither task. The vertebrate locomotor system is ideal for studying such performance trade-offs because the same muscles must often perform disparate tasks that require conflicting physiological functions.

One well-known and intuitive trade-off within the vertebrate locomotor system is that between speed and endurance capacity. The intuitive basis of this performance trade-off probably stems from the observation that elite specialist sprinting and marathon runners differ substantially in size and shape, and no athlete excels in both events. Individual

differences in the proportion of fast, powerful muscle fibres to slow more fatigue-resistant fibres are believed to be the primary physiological basis of this performance trade-off (Bennett et al., 1984; Garland, 1988). Consistent with this suggestion, short-distance human sprinters possess a greater proportion of fast-twitch muscle fibres than do endurance athletes, and *vice versa* for the slower more fatigue-resistant muscle fibres (Komi, 1984; Esbjörnsson et al., 1993). Similarly, individual differences in the proportions of different muscle fibre types correlate with differences in locomotor performance in the lizard *Dipsosaurus dorsalis* (Gleeson and Harrison, 1988). However, only a few studies have demonstrated a conflict between speed and endurance within a vertebrate species (Dohm et al., 1996; Reidy et al., 2000).

Most studies of whole-animal locomotor performance have found either no correlation or a positive relationship between speed and endurance (e.g. Ford and Shuttlesworth, 1986; Garland and Else, 1987; Garland, 1988; Shaffer et al., 1989; Tsuji et al., 1989; Huey et al., 1990; Jayne and Bennett, 1990;

Sorci et al., 1995). Why these intraspecific studies have failed to find a trade-off between speed and endurance, particularly when the physiological mechanism underlying the trade-off appears to have been identified, is not clear. One possibility is that the presumed physiological conflict at the level of the muscles is not as ubiquitous among vertebrates as commonly suggested. Alternatively, the effects of the physiological conflict at the whole-muscle level may be difficult to detect at the level of the whole animal. One way of testing these competing hypotheses is to investigate the existence of trade-offs at different levels of organisation. The actual site of the speed/endurance trade-off is believed to be predominantly at the level of the muscles, so investigations of individual variations in whole-muscle performance may indicate whether the speed/endurance trade-off should even be expected at the whole-animal level.

We investigated the existence of trade-offs between speed and endurance capacity at both the whole-muscle and whole-animal levels in post-metamorphs of the frog *Xenopus laevis*. Individual measures of burst swimming performance and endurance capacity were determined for juvenile *X. laevis*, and the work-loop technique was used to determine whether a conflict exists between maximum power production and fatigue-resistance of whole muscles. *X. laevis* are fully aquatic frogs, utilising both burst swimming for rapid escape from predators and extended bouts of swimming/jumping during migrations between connected water bodies (or overland) (Wilson et al., 2000; Passmore and Carruthers, 1995), so both measures of whole-animal performance appear to be relevant to their ecology.

Materials and methods

Juvenile *Xenopus laevis* Daudin at least 6 weeks post-metamorphosis (size range 0.54–1.98 g) were purchased from a local animal importer in Antwerp, Belgium. Frogs were returned to the laboratory at the University of Antwerp, where they were maintained in 201 aquaria at 20 °C until experiments began. Animals were exposed to a 12 h:12 h light:dark photoperiod and were fed frozen worms every second day. After several weeks, the burst swimming performance and endurance capacity of 55 individuals were assessed. After the locomotor trials, 18 of the post-metamorphic *X. laevis* were transported to the School of Science and the Environment at Coventry University, England, for analysis of isolated muscle performance using the work-loop technique.

Locomotor performance

Burst swimming performance was assessed by filming at least five swimming sequences for each individual with a high-speed digital camera. Swimming responses were elicited by placing a frog into the centre of a Perspex swimming arena (0.5 m × 0.5 m × 0.1 m deep) and tapping its urostyle with a fine wire probe, usually resulting in 2–3 swimming strokes. Only responses that produced swimming along the bottom of the aquarium from a stationary position were analysed. Swimming

sequences were filmed by recording the image at an angle of 90 ° above the arena with a high-speed digital camera (Redlake Imaging Corporation, USA) recording at 200 Hz. The accompanying Redlake software package was used to analyse the startle responses by playing back the sequences and digitising the tip of the frog's snout.

Two measures of burst swimming performance were calculated from each startle response; velocity during leg extension (U_{\max}) and velocity over an entire stride (U_{str}). Velocity over the leg extension phase was calculated by recording both the time and the distance moved between the beginning of leg extension (the frame before movement was first detected) and complete extension of the legs. Stride velocity was calculated over the entire swimming stroke from the beginning of leg extension until the end of leg flexion in preparation for the next stroke. The fastest of the swimming sequences analysed for each individual was used as a measure of its maximum burst swimming performance. Because there was considerable variation in the initial position of the hindlegs during the first swimming stroke of a startle response, only the second swimming stroke was used to assess maximal performance to ensure that all analysed swimming strokes started from maximum leg flexion. To assess the repeatability of performance measures, burst swimming was retested for 26 randomly selected individuals on the day after initial assessment. Endurance capacity did not differ between frogs subjected to a single burst swimming trial and those retested the following day.

Four days after assessment of burst swimming performance, the endurance capacity of each individual was measured in a Brett-type swimming flume (Brett, 1964). Endurance capacity tests were conducted after burst swimming trials to ensure that any long-term reduction in performance resulting from these more intensive trials did not influence the burst swimming trials. The flume consisted of a 30 cm long swimming section of 7 cm diameter through which a continuous water current was delivered by an adjustable Little Giant water pump (Little Giant, USA). Immediately prior to assessment of an individual's endurance capacity, it was placed into the swimming section of the flume and allowed an adjustment period of 10 min. The water velocity was then increased to 15 cm s⁻¹, and the frog responded to the continuous water flow by swimming against the current. The total time taken to reach exhaustion was recorded for each individual. A manually operated 10 V stimulator grid (Grass stimulator) was attached to the rear of the swimming section to prevent the frog from resting during the swimming trial. A 10 ms stimulus was delivered to the grid when a frog attempted to rest at the rear of the flume, usually resulting in an immediate return to swimming. We considered an individual had reached exhaustion when three successive stimuli separated by 1 s failed to induce a swimming response. Repeatability of endurance was assessed by retesting 24 randomly selected individuals 2 days after the initial assessment of endurance.

After locomotor performance trials, morphological measurements were recorded for each individual; these

included mass (with an electronic balance ± 0.01 g) and snout-to-vent, femur, tibia and foot length (Mitutoyo calipers ± 0.1 mm).

Isolated muscle performance

The frogs were killed by pithing and transection of the spinal cord, and the peroneus muscle was removed from the right hindleg and used for the muscle performance experiments. As we were testing for trade-offs between maximum power and fatigue resistance at the level of whole muscle, it was imperative that an appropriate whole muscle was selected. We chose the peroneus muscle because it was an ideal size for our muscle rig and it was convenient to dissect. Although not a major muscle involved in locomotion, the peroneus acts as a knee extensor and is thought to be a power-producing muscle during frog locomotion (Marsh, 1994). The peroneus, gastrocnemius and sartorius muscles of *Xenopus laevis* are all composed primarily of similar proportions of fast glycolytic fibres (Putnam and Bennett, 1983).

Dissection was performed at room temperature (20–25 °C) in Ringer's solution with the following composition (in mmol l^{-1}): NaCl, 115; KCl, 2.5; Na_2HPO_4 , 2.15; NaH_2PO_4 , 0.85; sodium pyruvate, 5.0; CaCl_2 , 1.8; pH 7.2 at 20 °C. Aluminium foil clips were attached to the tendons at the ends of the fibre bundles and mounted on stainless-steel hooks, with one end attached to a force transducer (AE 801, SensoNor; force calibration of 150 mN V^{-1}) and the other to a servomotor (V201, Ling Dynamic Systems). The muscle preparation was mounted in a temperature-controlled Perspex bath with circulating Ringer's solution saturated with 100 % oxygen and maintained at a constant 20 ± 0.5 °C.

Stimuli of 2 ms duration were delivered *via* two parallel platinum wire electrodes placed on either side of the muscle preparation. A series of twitches was used to determine the stimulation amplitude and muscle length (L_0) that generated the greatest isometric twitch force. A train of stimuli was delivered to the muscle to elicit a tetanic contraction. The frequency of stimulation was adjusted to maximise tetanus height (95–120 Hz) for each muscle and then used in the subsequent work-loop experiments. The experimental apparatus was controlled and data were collected using the Testpoint software package (CEC, MA, USA). The data were then exported and analysed in Microsoft Excel.

Each muscle was then subjected to cycles of oscillatory work, symmetrical about L_0 , using the work-loop technique (Josephson, 1985). A sinusoidal strain waveform was used during the work-loop experiments because the *in vivo* waveform for the peroneus muscle is not known for any anuran during swimming. The total strain was arbitrarily set at 15 % of L_0 , a value that approximates the strain observed for other anuran hindlimb muscles involved in powering locomotion (Olson and Marsh, 1998; Gillis and Biewener, 2000). Preliminary experiments revealed that the power output of the peroneus muscle changed with cycle frequency and approached a maximum at around 6 Hz. The maximum power output of each muscle was therefore measured at a cycle

frequency of 6 Hz. The number of stimuli imposed on the muscle and the timing of stimulation were adjusted to give maximum power output. A 5 min recovery period was allowed between each work loop. Control work loops using identical stimulation parameters were repeated every fourth run to monitor the muscle's ability to produce power. These control runs showed that muscle power production did not change by more than 15 % during the measurement of maximum power output before fatigue runs.

The fatigue-resistance of each muscle was determined by recording the changes in power production and force during continuous work cycles at 2 Hz. Observations of whole locomotor performance for the juvenile *X. laevis*, whilst assessing endurance capacity, revealed that a realistic stride frequency approximated 2 Hz. Before a fatigue run, the number of stimuli and the timing of stimulation were altered to maximise power output at 2 Hz. The fatigue run consisted of 120 cycles of oscillatory work, which ensured that the power output of all muscles had reached zero. Only one run was carried out per individual, and the total strain was 15 % of L_0 . Preliminary experiments revealed that the net power production of the muscles during a fatigue run decreased rapidly to zero after approximately 20–30 cycles. Thus, the fatigue-resistance of power production for each muscle was estimated by recording the number of cycles required to reduce power output to 5 % of the initial value. Although maximum force production for each cycle also decreased rapidly, a performance plateau was usually reached after approximately 30 cycles. The fatigue-resistance of force production was therefore estimated by recording the maximum force after 30 cycles as a proportion of the initial value.

At the end of the fatigue runs, the tendons were removed from the muscle, and excess Ringer's solution was removed by blotting the muscles with absorbent paper. The wet mass of each muscle was determined with an electronic balance (A&D Instruments, Oxford, UK) (± 0.01 mg) and used to calculate isometric stress and normalised muscle power output. Morphological measurements for each frog were recorded using Mitutoyo calipers (Japan) (± 0.01 mm). Muscle cross-sectional area was calculated from muscle mass and muscle length, with muscle density assumed to be 1060 kg m^{-3} (Méndez and Keys, 1960).

Statistical analyses

The relationship between body mass and performance parameters was expressed in the form $\log y = \log a + b \log M$, where M is body mass, a is the proportionality coefficient and b is the mass exponent. Equations were calculated using log-transformed data and least-squares regression techniques. Pearson's product moment correlations (r_p) were used to examine the repeatability of whole-animal performance measures between different days. These correlation analyses were based on the mass residuals of performance measures when a significant relationship between body mass and performance was detected. Correlations between measures of whole-animal performance and whole-muscle performance

were carried out on mass residuals (when appropriate; otherwise, log-transformed raw data were used) using Pearson's product moment correlation. Negative correlations between measures of speed and endurance indicate a trade-off in the performance traits. The Bonferroni method was used for multiple correlative tests by adjusting the significance values accordingly. Student's *t*-tests were used to compare measures of whole-animal endurance capacity between different testing days. All results are presented as means \pm S.E.M. Significance was taken at the level of $P < 0.05$.

Results

Whole-animal performance

Maximum burst swimming responses typically consisted of two or three swimming strokes involving rapid extension of the legs from a stationary crouched position followed by a gliding phase and active leg flexion, retracting the limbs in preparation for another stroke. For all individuals combined, U_{\max} was $59.4 \pm 1.4 \text{ cm s}^{-1}$ and U_{str} was $48.7 \pm 1.4 \text{ cm s}^{-1}$ ($N=55$). U_{\max} was positively correlated with body mass and was described by the equation $\log U_{\max} = 1.78 + 0.20 \log M$ ($N=55$; $r=0.30$; $P < 0.05$) (Fig. 1A). In contrast, U_{str} was not significantly influenced by body mass ($N=55$; $r=0.0001$; $P > 0.05$). For both measures of burst swimming, performance was significantly repeatable across different days ($N=26$; U_{\max} , $r_p=0.57$; U_{str} , $r_p=0.64$, $P < 0.01$).

The mean time taken to reach exhaustion whilst swimming in a constant-velocity flume was $475 \pm 73 \text{ s}$ ($N=55$) on the first

day of testing. E_{\max} was positively correlated with body mass and was described by the equation $\log E_{\max} = 2.57 + 1.4 \log M$ ($N=55$; $r=0.46$; $P < 0.01$) (Fig. 1B). E_{\max} differed significantly between testing days, being more than 40% lower on the second day of testing ($N=24$; day 1, $514.9 \pm 119.4 \text{ s}$; day 2, $301.1 \pm 44.0 \text{ s}$; $t=-3.35$; d.f.=22; $P < 0.01$). Nevertheless, the E_{\max} of each individual was positively correlated between testing days ($N=24$; $r_p=0.79$; $P < 0.01$), indicating that performance differences among individuals were significantly repeatable.

Burst swimming speed (either U_{\max} or U_{str}) was not significantly correlated with E_{\max} ($N=55$; $r_p=-0.01$; $P=0.94$ for U_{\max} ; $r_p=-0.028$; $P=0.84$ for U_{str}), indicating that there was no trade-off between whole-animal measures of speed and endurance (Fig. 2). Correlations between burst swimming speed (either U_{\max} or U_{str}) and E_{\max} based on total limb length residuals were also non-significant.

Isolated whole-muscle performance

The mean isometric stress for the peroneus muscle from juvenile *Xenopus laevis* was $268 \pm 12.7 \text{ kN m}^{-2}$ ($N=18$). Cycle frequency affected the peak power output of the peroneus muscle, with values of $38.7 \pm 2.3 \text{ W kg}^{-1}$ at 2 Hz and $71.1 \pm 4.0 \text{ W kg}^{-1}$ ($N=18$) at 6 Hz. Peak instantaneous power output was $165.7 \pm 10.0 \text{ W kg}^{-1}$ at 2 Hz and $257.4 \pm 19.4 \text{ W kg}^{-1}$ ($N=18$) at 6 Hz. During a typical optimised work loop at 6 Hz, force began to rise near the end of muscle lengthening and reached a peak just after maximum muscle length had been reached. Muscle force then gradually decreased throughout muscle shortening and reached a minimum close to its shortest length.

During a set of continuous work-loop cycles at 2 Hz, the maximum force produced during each cycle decreased rapidly over the first 20 cycles and reached a plateau after approximately 20–30 cycles (Fig. 3A). The plateau of force production varied among individuals and was approximately 5–20% of the force produced at the start of a fatigue run. Changes in work-loop shape during a fatigue run reflected both

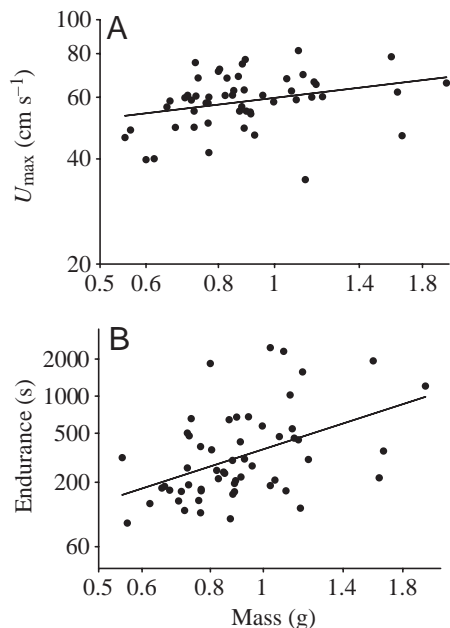


Fig. 1. Relationship between body mass (M) and (A) maximum burst swimming velocity (U_{\max}) ($\log U_{\max} = 1.78 + 0.20 \log M$, $N=55$; $r=0.30$; $P < 0.05$) and (B) the time taken to reach exhaustion whilst swimming at a constant velocity (endurance, E_{\max}) ($\log E_{\max} = 2.57 + 1.4 \log M$, $N=55$; $r=0.46$; $P < 0.01$) for 55 juvenile *Xenopus laevis*.

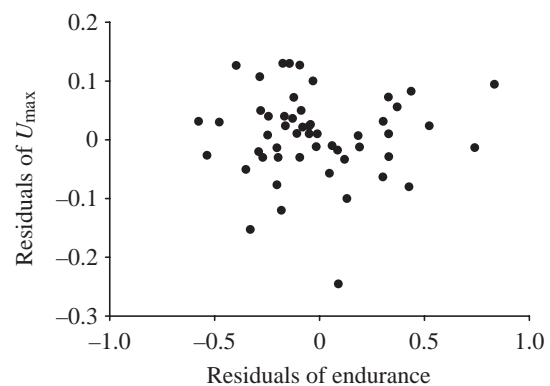


Fig. 2. Relationship between mass residuals of maximum burst swimming velocity (U_{\max}) and the time taken to reach exhaustion whilst swimming at a constant velocity (endurance) for 55 juvenile *Xenopus laevis*.

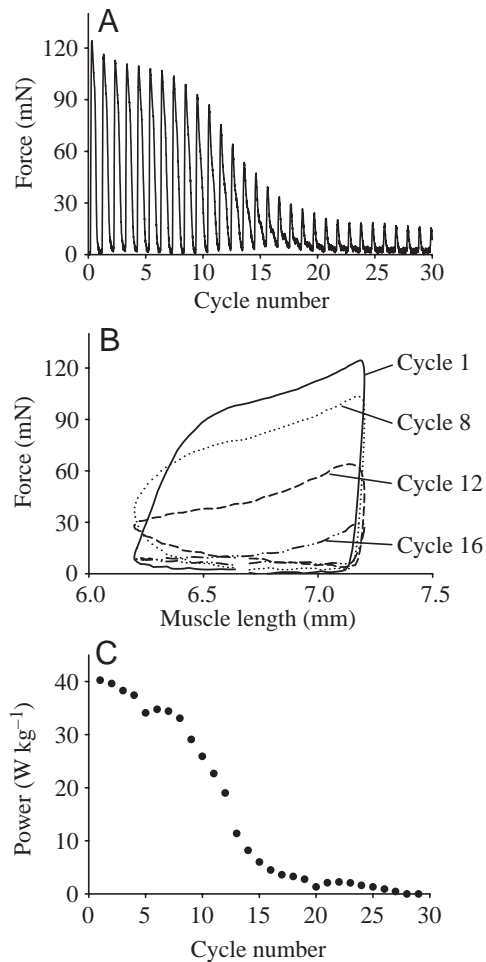


Fig. 3. Changes in (A) force, (B) work-loop shape and (C) net power output per cycle for a representative *Xenopus laevis* peroneus muscle during a continuous set of cycles at 2 Hz using the work-loop technique.

the rapid decrease in force production and the increase in muscle relaxation time (Fig. 3B). Thus, force generated at peak length and the maintenance of force during shortening decreased rapidly as the muscle fatigued. When fatigued, the muscle also failed to relax completely during shortening, resulting in an increase in force production during muscle lengthening and an increase in the work required for muscle lengthening. Total power production decreased rapidly during a set of continuous cycles and usually reached zero after 20–30 cycles (Fig. 3C). One of the characteristics of muscle fatigue is its reversibility following a period of recovery. The maximum power output of peroneus muscle from *X. laevis* returned to $78.1 \pm 2.3\%$ ($N=4$) of the pre-fatigue value after a recovery period of 20 min.

Peak instantaneous power output at 6 Hz was negatively correlated with the fatigue-resistance of force production at 2 Hz ($N=18$, $r_p=-0.58$; $P<0.01$) (Fig. 4A). Although all other correlations between measures of power production and fatigue-resistance also exhibited negative relationships, none was statistically significant (Fig. 4B–D).

There were no significant correlations between measures of whole-animal performance and *in vitro* muscle performance for individual *X. laevis*. For example, peak power output and peak instantaneous power output of the peroneus muscle at 6 Hz were not significantly correlated with U_{\max} ($N=18$, $r_p=0.13$; $P>0.05$ for peak power output; $r_p=0.08$; $P>0.05$ for peak instantaneous power output). In addition, the fatigue-resistance of force production at 2 Hz was not significantly correlated with E_{\max} ($N=18$, $r_p=-0.13$; $P>0.05$).

Discussion

The biophysical and biochemical properties of vertebrate skeletal muscle differ between fast and slow fibre types (Lannergren and Hoh, 1984; Gleeson and Harrison, 1986; Lannergren, 1987; Esbjörnsson et al., 1993; Rivero et al., 1993). A trade-off between speed and endurance capacity should be expected in vertebrate muscles as a result of this variation in muscle contractile properties (Bennett et al., 1984; Garland, 1988). We found evidence of a trade-off between maximum *in vitro* power output and fatigue-resistance of whole muscles for post-metamorphic *Xenopus laevis*. Peroneus muscles that produced high levels of *in vitro* mechanical power generally possessed lower fatigue-resistance than muscles producing low levels of mechanical power. Thus, our data support the suggestion that a physiological conflict between maximum power output and fatigue-resistance exists at the level of the muscles. Although most correlations between measures of power output and fatigue-resistance of whole muscles were non-significant, all demonstrated negative correlations.

Comparisons of mechanical properties of the peroneus with those reported previously are difficult because of its limited use for studies of muscle mechanics. However, its general structure appears to be similar to that of the gastrocnemius and sartorius muscles (Putnam and Bennett, 1983), which have been well studied for numerous anurans (for a review, see Marsh, 1994). In our study, mean maximum isometric stress for peroneus muscles was comparable both with that of *X. laevis* sartorius at 20 °C (mean maximum stress of 215 kN m^{-2}) (Altringham et al., 1996) and with the range of previous values for anuran muscles at 20 °C (Marsh, 1994). Maximum power outputs for *X. laevis* peroneus muscle at 6 Hz are also similar to those recorded for *X. laevis* sartorius from similar-sized individuals (Altringham et al., 1996). Fatigue-induced changes in the mechanical properties of *X. laevis* peroneus muscle were almost completely reversible, with *in vitro* performance returning to approximately 80% of pre-fatigue values within 20 min. Decreases in power output during a fatigue run were due to a reduction in force production and to an increase in relaxation time. Although few studies have investigated the fatigue-induced changes in contractile properties during sinusoidal work, all report that force production and relaxation rate changed simultaneously during fatigue (Syme and Stevens, 1989; Stevens and Syme, 1993; Askew and Marsh, 1997).

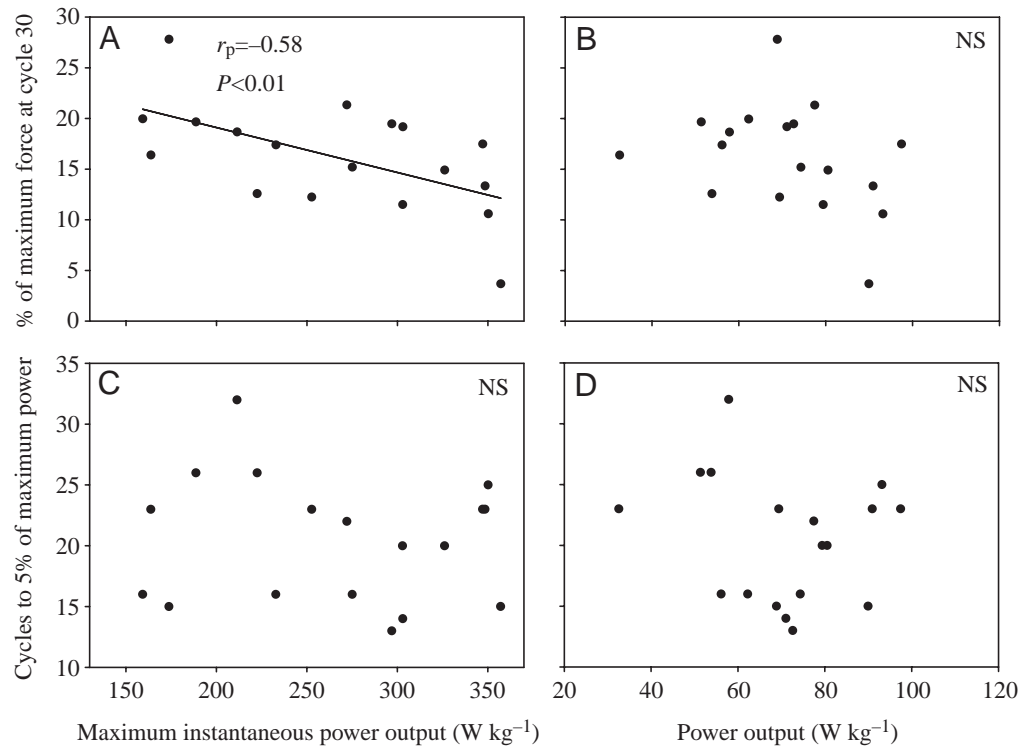


Fig. 4. Correlations between measures of maximum power output at 6 Hz and fatigue-resistance of the peroneus muscle at 2 Hz for 18 juvenile *Xenopus laevis*. There was a significant negative correlation between maximum instantaneous power output and fatigue-resistance of force (% of maximum force at cycle 30) (A). 'Cycles to 5% of maximum power' is the number of cycles taken to reduce net power to 5% of the initial value at cycle 1. NS, non-significant Pearson's product moment correlation.

We found no correlation between measures of maximum burst swimming speed and endurance capacity for juvenile *X. laevis*, suggesting that no trade-off exists between these two performance measures at the whole-animal level. Most other intraspecific studies of whole-animal performance also report no trade-off between whole-animal measures of speed and endurance capacity (e.g. Bennett, 1980; Ford and Shuttlesworth, 1986; Garland and Else, 1987; Garland, 1988, 1994; Tsuji et al., 1989; Huey et al., 1990; Jayne and Bennett, 1990; Secor et al., 1992; Sorci et al., 1995). Empirical support for the speed/endurance trade-off at the whole-animal level comes from only a few intra- and inter-specific studies. Reidy et al. (2000) found a negative correlation between a measure of burst swimming performance and endurance capacity in the Atlantic cod (*Gadus morhua*). In a recent interspecific phylogenetic study of lizard locomotion, Vanhooydonck et al. (2001) reported a trade-off between speed and endurance among several species of lacertid lizard. In a quantitative genetic analysis of sprinting and endurance capacity in the laboratory house mouse (*Mus domesticus*), Dohm et al. (1996) found a negative genetic correlation between sprint running and swimming endurance.

The absence of a speed/endurance trade-off for post-metamorphic *X. laevis* at the level of the whole animal, even though it was detected at the level of the whole muscles, suggests an apparent incongruence between whole-animal and whole-muscle performance. Assuming that the physiological conflict identified for the peroneus muscle also occurs in other larger muscles that have a greater direct influence on locomotor performance, then whole-animal performance should reflect the trade-offs imposed on the muscular system. Several alternative hypotheses relating to the robustness of the whole-

muscle and whole-animal analyses, and factors masking a possible trade-off at the whole-animal level, may help explain this apparent mismatch between whole-animal and whole-muscle performance.

Muscles *in vivo* are used in dynamic and complex ways, so it is possible that the *in vitro* measures of muscle performance do not adequately reflect the *in vivo* locomotor state. Compared with whole-animal measures of endurance, *in vitro* fatigue of force and power production for the peroneus muscles of juvenile *X. laevis* occurred rapidly. The mean number of swimming strides before exhaustion was more than 800, while for most whole peroneus muscles power output decreased to zero within 30 cycles. During an *in vitro* fatigue experiment, muscle stimulation parameters were set to elicit maximum power production for non-fatigued muscle and presumably recruited all fibres in each cycle. In contrast, not all the muscle fibres are simultaneously recruited *in vivo* during extended bouts of locomotion, with muscle fibres alternating between rest and work. Thus, it is possible that our *in vitro* measures of muscle performance do not relate to the *in vivo* locomotor conditions. Future studies of muscle fatigue may benefit from replicating the cycling of muscle fibre activation by alternating passive and active stimulation loops during a fatigue run, thereby more closely representing *in vivo* muscle fatigue.

The impact of motivation and other behavioural effects on whole-animal measures of locomotor performance is more difficult to quantify. It is possible that the measures of locomotor performance used in this study do not adequately determine the actual maximum locomotor capabilities of each individual frog. This is a potential problem facing all

researchers studying whole-animal performance. Although measures of *X. laevis* performance were repeatable for individuals across different days, it is still possible that much of this inter-individual variability is attributable to underlying motivational/behavioural rather than physiological/morphological variation. This seems especially likely for measures of endurance capacity for *X. laevis*, which displayed great inter-individual variation (100–2000 s to reach exhaustion). Coefficients of variation differed substantially between measures of endurance (approximately 115%) and burst swimming performance (approximately 20%). Such large inter-individual variation may be due to motivational or general behavioural differences rather than to underlying physiological determinants of performance.

Other physiological or behavioural factors besides muscular performance may also be masking the speed/endurance trade-off at the whole-animal level. Several recent studies examining the physiological basis of avian locomotor physiology suggest that factors other than muscular performance (e.g. the mass of central organs) may be more important determinants of whole-animal aerobic performance (Chappell et al., 1999; Hammond et al., 2000). Individual variation in endurance capacity caused by other physiological factors may require very large sample sizes to detect muscle-mediated trade-offs at the whole-animal level. Even more problematic are factors, such as general health and condition, prior injury or motivation, that may result in positive correlations between different whole-animal performance traits and effectively swamp more subtle muscle-mediated trade-offs within an individual. Several recent studies of locomotion have also emphasised that most organisms use a more intermittent type of locomotion during extended bouts of movement rather than the steady-state type most commonly examined (for a review, see Kramer and McLaughlin, 2001). Both the whole-muscle and the whole-animal measures of endurance capacity used in our study are based on a steady-state type of locomotion and fatigue, possibly questioning the ecological relevance of the performance measures studied.

For the juvenile *X. laevis*, we found no correlation between whole-animal measures of performance and whole-muscle performance for any variables. The absence of correlation between different levels of organisation may indicate that the traits measured are not important determinants of performance or that measures of whole-animal performance are not adequate estimates of locomotor capacities. However, given that the mass of the peroneus muscle of *X. laevis* is very small in comparison with the total muscle mass used during locomotion, a correlation between whole animal and whole muscle is unlikely. Studies investigating the physiological determinants of locomotor performance in vertebrates have experienced mixed results. Although several studies have found correlations between individual variation in locomotor or aerobic performance and a variety of physiological processes (Garland, 1984; Bennett et al., 1989; Kolok, 1992; Chappell et al., 1999; Garenc et al., 1999; Hammond et al.,

2000), just as many have found either no correlation or only for a particular sex or life history stage (Garland, 1984; Kolok and Farrell, 1994; Garland et al., 1995; Garenc et al., 1999; Hammond et al., 2000).

In the present study, we found evidence of a trade-off between maximum power output and the fatigue-resistance of whole muscles for *Xenopus laevis*. In contrast, we found no correlation between whole-animal measures of speed and endurance capacity. This apparent incongruence between different levels of analysis may be due to several factors affecting both whole-muscle and whole-animal performance measures and warrants further detailed investigation.

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