

CONSEQUENCES OF A GAIT CHANGE DURING LOCOMOTION IN TOADS (*BUFO WOODHOUSII FOWLERI*)

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Summary

Most animals cannot sustain speeds above that at which the rate of oxygen consumption reaches a maximum ($\dot{V}_{O_{2max}}$). Fowler's toad (*Bufo woodhousii fowleri*), by contrast, has a maximum aerobic speed (MAS, the speed at $\dot{V}_{O_{2max}}$) of 0.27 km h^{-1} but can sustain speeds as high as 0.45 km h^{-1} without increasing the \dot{V}_{O_2} above the $\dot{V}_{O_{2max}}$. The present study investigates the discrepancy between MAS and the maximum sustainable speed (MSS). Toads switched from walking to hopping as their speed increased. The cost of a hop ($4.1 \times 10^{-4} \text{ ml O}_2 \text{ g}^{-1} \text{ hop}^{-1}$) was greater than the cost of a walking stride ($2.5 \times 10^{-4} \text{ ml O}_2 \text{ g}^{-1} \text{ stride}^{-1}$) and was independent of speed for both hopping and walking. However, individual hops were much longer than walking strides, which more than offset the greater cost of a hop. The calculated cost to traverse a given distance was approximately 1.9 times as much for walking as for hopping. During natural locomotion animals used combined walking and hopping. Individual toads that favored walking had higher locomotor costs than those that favored hopping. The estimated cost of exclusive hopping was less than the cost of natural locomotion at all but the highest speeds. This discrepancy may reflect the fact that the natural gait is a combination of both the less economical walking gait and the more economical hopping gait. To achieve speeds above the MAS toads walked less and used the more economical hopping gait more, and thus did not increase energy cost above that of $\dot{V}_{O_{2max}}$. The speed at which the estimated cost of exclusive hopping exceeded the cost of a natural gait and approached the $\dot{V}_{O_{2max}}$ was close to the MSS. Creatine phosphate and lactate concentrations in the muscles of the thigh and calf did not change from resting levels at sustainable speeds greater than the MAS.

Introduction

In many animals, the rate of aerobic metabolism increases linearly as the speed

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of land locomotion increases. Oxygen consumption (\dot{V}_{O_2}) continues to increase with speed until a speed is reached at which oxygen consumption is maximal: the maximal aerobic speed or MAS (Bennett, 1982; Seeherman *et al.* 1981; Full *et al.* 1988; Full, 1986, 1987). Greater speeds are possible, but typically are not sustainable.

Toads are an exception to this general model. For example, *Bufo woodhousii fowleri* reach MAS at 0.27 km h^{-1} but can sustain speeds of 0.36 km h^{-1} for 2 h and 0.45 km h^{-1} for more than 10 min. Even more puzzling is that the rate of oxygen consumption does not increase between 0.27 and 0.45 km h^{-1} (Walton and Anderson, 1988). We tested two of the hypotheses that might explain these phenomena. (1) At speeds greater than the MAS, toads may undergo significant net anaerobiosis and accumulate lactate, thus increasing total metabolism without an increase in \dot{V}_{O_2} (Bennett and Licht, 1973, 1974; Carey, 1979). Such prolonged anaerobic metabolism, however, is generally considered to result in fatigue (Bennett, 1982). If accelerated glycolysis largely fuels the increase in speed, then toad muscle must resist fatigue or toads must avoid fatigue in other ways. (2) Toads may increase speed above the MAS with no increase in metabolic cost. This could occur if, in the manner of red kangaroos (Dawson and Taylor, 1973), they elastically store and recover a significant fraction of the kinetic energy derived from each hop and if elastic energy storage increases with speed above the MAS. Walton and Anderson (1988) rejected this hypothesis on the basis of metabolic and kinematic measurements at speeds above the MAS. Alternatively, toads may increase speed above the MAS without an increase in metabolic cost if, as in horses (Hoyt and Taylor, 1981), mink (Williams, 1983) and ground squirrels (Hoyt and Kenagy, 1988), they switch to a more economical gait (i.e. lower metabolic cost per distance travelled) as speed increases. Just as changing to a higher gear ratio may allow a motor vehicle to achieve a greater speed with no increase in fuel consumption, changing to a more economical gait could allow a toad to increase speed above the MAS without increasing \dot{V}_{O_2} . Indeed, toads do change gait, from a walk at slow speeds to a hop at faster speeds (Walton and Anderson, 1988). However, the exact relationship of gait to speed and its correspondence with \dot{V}_{O_2} is unknown. For this hypothesis to be supported, the net savings from hopping (i.e. the presumptive economical gait) should exactly counterbalance the increased energy requirement of locomotion at speeds greater than the MAS.

The present study evaluates these two hypotheses. To evaluate the gait change hypothesis we characterized the use of the two gaits, walking and hopping, in toads moving on a treadmill at a range of speeds. Then, we estimated the cost of the two gaits individually. To determine the cost of walking, we exercised toads on a shortened treadmill enclosed in a respirometer within which only walking was possible. To calculate the cost of hopping, we used 'instantaneous' measurements of \dot{V}_{O_2} (Bartholomew *et al.* 1981) during intervals of normal locomotion in which toads hopped exclusively. We also examined whether individual toads that rely disproportionately on walking have greater locomotor costs than toads that rely disproportionately on hopping. To evaluate the anaerobic metabolism hypothesis,

we determined net non-aerobic metabolism during walking and natural locomotion.

Materials and methods

Animals

Fowler's toads *Bufo woodhousii fowleri* (body mass = 20.6 ± 5.5 g, s.d.) were obtained at the Indiana Dunes National Lakeshore, Porter Co., IN, and from Charles D. Sullivan, a commercial supplier. Animals were maintained at 21°C on a 14 h:10 h L:D photoperiod centered on 13:00 h local time. Toads were fed crickets (*Acheta domestica*) twice weekly until 3 days before experiments. All experiments were conducted at 21°C.

Oxygen consumption during exclusive walking

Measurements of \dot{V}_{O_2} during rest and sustained walking were made in a gas-tight, clear acrylic respirometer containing a variable-speed treadmill (Herreid, 1981; Full, 1986; Walton and Anderson, 1988). The chamber within which the animals walked consisted of a half-cylinder 8.0 cm long, 8.7 cm wide and 5.2 cm tall at its highest point. Because of its relatively short length, this chamber permitted animals to walk comfortably but did not allow them to hop. The interior of the treadmill was moistened with 10 ml of distilled water before a toad was placed inside. A toad rested within the treadmill in dim light for 45 min to 1 h before exercise. \dot{V}_{O_2} was measured during the last 10–15 min of the rest period ($\dot{V}_{O_{2,rest}}$). Toads were forced to walk at four speeds beginning with the lowest. After steady-state \dot{V}_{O_2} had been maintained for 5–10 min, the treadmill speed was increased to the next higher speed. This procedure was repeated until animals had been tested at all speeds. At speeds of less than 0.04 km h^{-1} , animals explored the chamber rather than walking forward steadily. Toads could not walk steadily at speeds above 0.14 km h^{-1} . When possible, each toad was exercised at four speeds. If animals would not walk at all speeds during a single exercise bout, they were rested for at least 2 days and retested at the missing speeds. Multiple speed trials resulted in \dot{V}_{O_2} values that were comparable to those from single speed trials. Trials in which toads did not walk consistently were discarded.

Oxygen consumption was measured continuously during rest and walking with open-flow respirometry. An air pump drew humidified air through the system at 500 ml min^{-1} . Incurrent fractional oxygen content ($F_{I_{O_2}}$) was compared to the excurrent fractional oxygen content ($F_{E_{O_2}}$) with a dual-channel oxygen analyser (Ametek Applied Electrochemistry, model S-3A). Before analysis, gas was drawn through columns of Drierite and Ascarite to remove water and CO_2 , respectively. Output from the oxygen analyser was processed with an analog-to-digital converter (Isaac 41A system, Cyborg) and a microcomputer (Apple II⁺). Data were collected at 10 s intervals.

Instantaneous \dot{V}_{O_2} was calculated from $F_{I_{O_2}}$ and $F_{E_{O_2}}$ (Full and Herreid, 1983, 1984; Herreid *et al.* 1981; Herreid, 1981; Walton and Anderson, 1988). Given any

two measurements separated by a brief interval, the flow rate (\dot{V}) and the 'washout' characteristics of the chamber, F_{eq} (the equilibrium of FE_{O_2} inside the metabolic chamber) can be calculated and substituted for FE_{O_2} in a standard equation:

$$\dot{V}_{O_2} = \dot{V}(F_{I_{O_2}} - F_{eq}) / (1 - F_{I_{O_2}}) \quad (1)$$

to estimate instantaneous \dot{V}_{O_2} . Rates of \dot{V}_{O_2} are reported as ml O_2 at STPD. The 'effective volume' used in the calculation of F_{eq} was calculated from the washout of nitrogen gas. The effective volume was 6034 ml.

The \dot{V}_{O_2} measured during rest was subtracted from the observed \dot{V}_{O_2} during walking. The walking stride rate at each speed was measured by counting the number of walking strides in 10s intervals. The incremental aerobic cost of walking was divided by the walking stride rate to yield the cost of a single walking stride.

Oxygen consumption during exclusive hopping and natural locomotion

Measurements of $\dot{V}_{O_{2,rest}}$ and \dot{V}_{O_2} during sustained natural, mixed-gait locomotion (i.e. walking and hopping) were made in a gas-tight acrylic respirometer containing a variable-speed treadmill. The methods were similar to those described above except that the chamber was larger, a half-cylinder 25.4 cm long, 12.7 cm wide and 6.4 cm tall at its highest point. This chamber was large enough for toads to hop unencumbered (Walton and Anderson, 1988). The toads were exercised at a single speed in each exercise bout for 30 min, and given at least 2 days between bouts. Five speeds were tested. In the open-flow respirometry, air was drawn through the system at 300 or 500 ml min^{-1} depending on the size of the toad. The effective volumes of the chamber at these two flow rates were 4480 and 3620 ml, respectively. These trials were videotaped with a video camera (NEC, model TI-22P CCD) fitted with a video lens (Schneider-Kreuznach, Xenoplan 1.7/17) and connected to a videocassette recorder (Panasonic model AG 6300) and video monitor (NEC AutoColor) to measure the hopping and walking strides in each 10s interval.

The cost of a hop was estimated from intervals during which animals hopped predominantly or exclusively for 50s or longer. The aerobic cost of a single hop was calculated as the difference between the cumulative oxygen consumed and the resting consumption during each interval divided by the total number of hops.

To examine the generality and robustness of these estimates, we calculated the aerobic costs of toads using natural mixed gaits. The aerobic cost was calculated from the number and estimated cost of walking strides and hops for the entire period during which steady-state \dot{V}_{O_2} was available:

$$\text{Estimated aerobic cost } (\dot{V}_{O_2}) = (\dot{V}_{O_{2,rest}}) + (n_{hop} V_{O_2}) + (n_{walk} V_{O_2}), \quad (2)$$

where V_{O_2} is aerobic cost per hopping or walking strides, and n_{hop} and n_{walk} are the number of hops and walking strides, respectively, per unit time during an exercise bout.

Lactate and creatine phosphate concentrations in the hindlimb musculature

Animals were rested and exercised in the treadmills described above. Chambers were ventilated with room air at 500 ml min^{-1} . Animals rested in the chamber for 10 min before each experiment. Five different exercise regimes were used: (a) rest for 10 min; (b) exclusive walking to fatigue at 0.14 km h^{-1} ; (c) natural locomotion at 0.36 km h^{-1} for 10 min; (d) natural locomotion at 0.36 km h^{-1} for 30 min and (e) natural locomotion to fatigue at 0.63 km h^{-1} .

After the exercise period, the animals were immediately frozen in liquid nitrogen. They were submerged in the nitrogen within 2 s of the end of exercise and were frozen within 2 s of being submerged. After freezing the animals were stored at -80°C . Within 3 days the calf and thigh muscles were dissected while the animals were still frozen. Standard spectrophotometric assays (Bergmeyer, 1974) were conducted for lactate and creatine phosphate. Separate assays were done in duplicate on the thigh and calf muscles of each toad.

Results*Stride frequency and oxygen consumption in animals using a mixed gait*

Animals moving naturally used a mixed gait in which hops were interspersed with walking strides. Our measured rates of \dot{V}_{O_2} for natural mixed-gait locomotion (Fig. 1A) agree with those of Walton and Anderson (1988). The number of strides per unit time that were hops (n_{hop} , Hz) increased with increasing speed ($n_{\text{hop}}=0.51v+0.29$; $r=0.44$, $P=0.004$; Fig. 1B), whereas the frequency of walking strides (n_{walk}) decreased with increasing speed ($n_{\text{walk}}=-0.34v+0.23$; $r=0.32$, $P=0.045$; Fig. 1C; where v is speed in km h^{-1}). Total stride frequency (i.e. hopping+walking strides) was not correlated with the speed of locomotion ($r=0.13$, $P=0.44$; Fig. 1D). Most importantly, at the lowest speeds ($0.18\text{--}0.27 \text{ km h}^{-1}$) walking accounted for more than 30% of total strides in at least half of the toads, whereas at higher speeds walking never accounted for more than 30% of total strides (Fig. 1E). Thus, toads changed gait as speed increased.

Cost of a walking stride

When toads were forced to walk exclusively in a shortened treadmill, \dot{V}_{O_2} was not related to locomotor speed ($r=0.09$, $P=0.64$; Fig. 2A). The \dot{V}_{O_2} during exclusive walking ($0.83\pm 0.03 \text{ ml O}_2 \text{ g}^{-1} \text{ h}^{-1}$, s.e.) was greater than the average \dot{V}_{O_2} during natural locomotion at speeds of less than 0.14 km h^{-1} (Walton and Anderson, 1988). During exclusive walking, stride frequency (f_{walk} , Hz) increased with speed (km h^{-1}) ($f_{\text{walk}}=4.15v+0.29$; $r=0.90$, $P<0.001$; Fig. 2B).

Because stride frequency increased with walking speed while \dot{V}_{O_2} remained constant, the aerobic cost of a single walking stride decreased with speed. To facilitate subsequent comparisons with the cost of a single hop, the cost of a single walking stride was estimated for animals walking at 0.1 km h^{-1} , a speed below that at which significant anaerobic metabolism occurs (see below). The difference

between the average $\dot{V}_{O_2,rest}$ ($0.18 \text{ ml O}_2 \text{ g}^{-1} \text{ h}^{-1}$) and the \dot{V}_{O_2} during walking at 0.1 km h^{-1} ($0.8 \text{ ml O}_2 \text{ g}^{-1} \text{ h}^{-1}$) divided by the stride frequency at this speed (0.70 Hz) yielded a cost of $2.45 \times 10^{-4} \text{ ml O}_2 \text{ g}^{-1} \text{ stride}^{-1}$. In natural locomotion the length of a stride does not change with speed and is between 2 and 4 cm. Thus, this calculated cost of a walking stride is presumably equally applicable to a wide range of speeds.

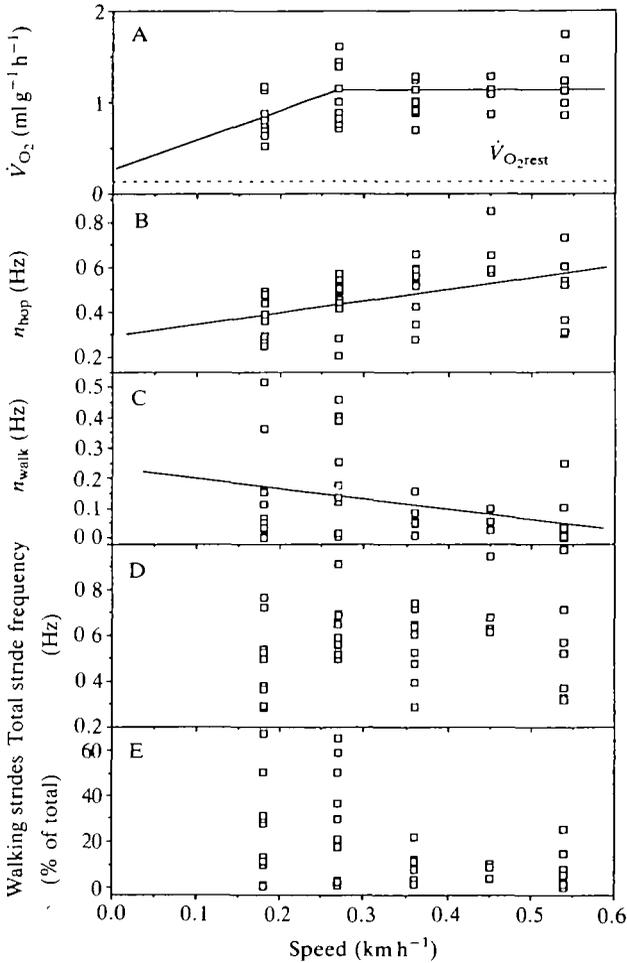


Fig. 1. Effect of speed during natural locomotion on aerobic energetics, proportion of walking and hopping strides and total stride frequency. Each point represents data for a single animal at a given speed. (A) Oxygen consumption. Lines represent the effect of speed on \dot{V}_{O_2} as previously reported by Walton and Anderson (1988). The dashed line represents $\dot{V}_{O_2,rest}$. Maximum aerobic speed (MAS) was attained at 0.27 km h^{-1} . (B) Number of strides that were hops per unit time (n_{hop}). (C) Number of strides that were walks per unit time (n_{walk}). (D) Total stride frequency (walking strides and hops). (E) The percentage of total strides that were walking strides. Note that walking is rare at high speeds.

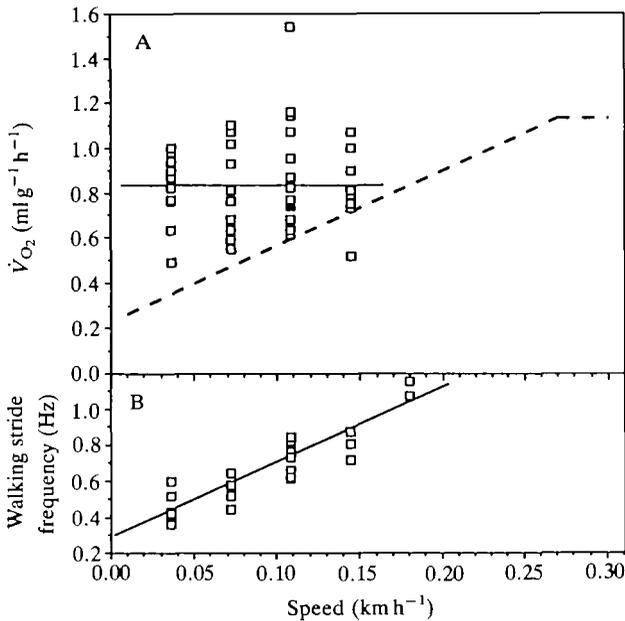


Fig. 2. Effect of speed on oxygen consumption and stride frequency in toads exclusively walking in a shortened treadmill. Each point represents data for a single animal at a given speed. (A) Oxygen consumption (\dot{V}_{O_2}) during walking was not correlated with speed. The solid line indicates the mean \dot{V}_{O_2} . The dashed line is the \dot{V}_{O_2} of toads using a natural mixed gait (Walton and Anderson, 1988). (B) Walking stride frequency (f_{walk}) increased linearly with speed.

Cost of a hop

From simultaneous measurements of locomotion and \dot{V}_{O_2} , we identified 103 intervals in which walking strides occurred less than five times per minute. These intervals ranged from 50 to 840 s (mean=155 s). We averaged all intervals for a given animal at a given speed, omitting data for the fastest speed (0.63 km h⁻¹), at which significant anaerobiosis occurs (see below).

The cost of a hop was calculated by dividing the net hopping \dot{V}_{O_2} by hop frequency. Resultant values varied from 2.3×10^{-4} to 7.1×10^{-4} ml O₂ g⁻¹ hop⁻¹ (mean = $4.15 \times 10^{-4} \pm 0.20 \times 10^{-4}$ ml O₂ g⁻¹ hop⁻¹, s.e.). These estimated hop costs were not correlated with speed of locomotion ($r=0.1$, $P=0.58$; Fig. 3A). Hop frequency (f_{hop} , Hz) during periods of exclusive hopping was positively correlated with speed (km h⁻¹) ($f_{\text{hop}}=0.77v+0.33$; $r=0.61$; $P<0.001$; Fig. 3B). The estimated hop length (l_{hop} , cm; calculated from hop frequency and adjusted for any walking) was also positively correlated with speed (km h⁻¹) ($l_{\text{hop}}=20.9v+7.7$; $r=0.75$; $P<0.001$; Fig. 3C). Hop cost and hopping length showed a positive relationship [cost of a hop (ml O₂ g⁻¹ hop⁻¹) = $1.3 \times 10^{-5} l_{\text{hop}}$ (cm) + 2.1×10^{-4} ; $r=0.62$; $P<0.001$; Fig. 4A]. Hop cost and hopping frequency showed a negative relationship [cost of a hop (ml O₂ g⁻¹ hop⁻¹) = $-3.1 \times 10^{-4} f_{\text{hop}}$ (Hz) + 5.8×10^{-4} ; $r=0.47$;

$P < 0.001$; Fig. 4B]. As speed increased, the greater cost of longer hops was offset by the lower cost of more frequent hops, resulting in the independence of cost per hop and speed (Fig. 3A).

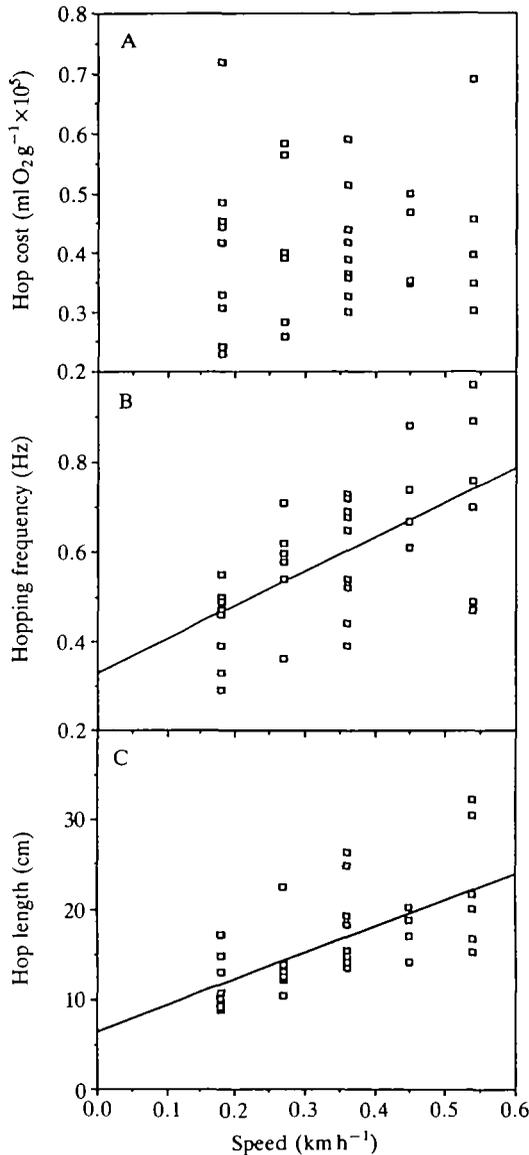


Fig. 3. Effect of speed on the aerobic cost per hop, hopping frequency and the length of individual hops. Each symbol represents the average of 1–10 intervals of near-exclusive hopping for a single animal at a given speed. (A) Aerobic cost per hop was not correlated with speed. (B) Hopping frequency (f_{hop}) increased linearly with speed. (C) Hop length (l_{hop}) also increased linearly with speed.

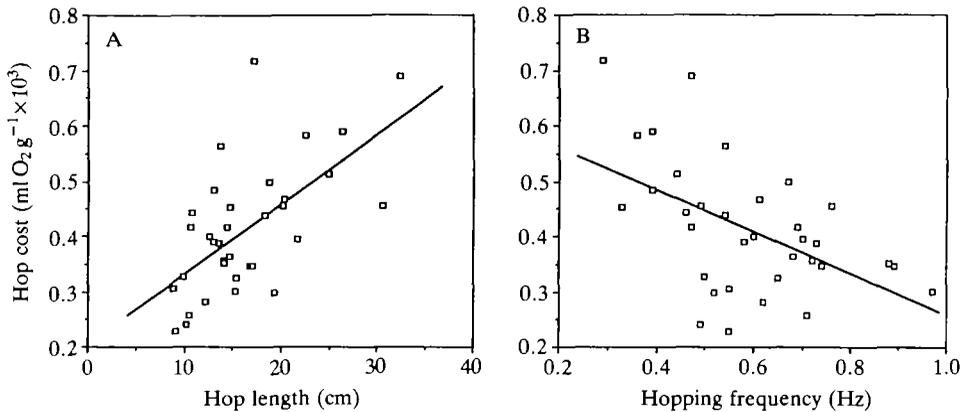


Fig. 4. Relationship between aerobic cost of individual hops and hopping frequency and length. Data were derived as in Fig. 3. (A) The cost per hop increased as hop length (l_{hop}) increased. (B) The cost per hop decreased as hopping frequency (f_{hop}) increased.

Locomotor costs of animals using mixed gaits

As Fig. 1 demonstrates, individual animals differ greatly in the relative number of hops and walking strides they employ during mixed-gait locomotion at any given speed. We examined whether individual toads that employed more than the average number of walking strides had greater than average locomotor costs and *vice versa*. We applied stepwise multiple regression to the 30 cases, to give the following equation: \dot{V}_{O_2} ($\text{ml O}_2 \text{ g}^{-1} \text{ h}^{-1}$) = $1.24 \times 10^{-3} v$ (km h^{-1}) - 2×10^{-5} mass (g) + 9.1×10^{-4} ; $r=0.76$. Locomotor speed accounted for 47% of the variance in \dot{V}_{O_2} and body mass an additional 8%. From this equation we calculated the standardized residual \dot{V}_{O_2} . The sign of the residual expresses whether the actual \dot{V}_{O_2} of an animal is above or below that expected for an animal of the same size moving at the same speed; the magnitude of the residual expresses the extent to which the observed \dot{V}_{O_2} departs from the expected value. The number of hopping strides per hour (n_{hop}) bore no relationship to the standardized residual \dot{V}_{O_2} ($r=0.19$; $P=0.30$; Fig. 5A). By contrast, the significant positive correlation between the standardized residual \dot{V}_{O_2} and the proportion of walking strides per hour (residual $\dot{V}_{\text{O}_2} = 3.0 \times 10^{-3} n_{\text{walk}} - 0.69$; $r=0.35$; $P<0.0001$) indicates that toads that walk more than average when using mixed gaits also have the greatest costs of locomotion (Fig. 5B).

To examine the generality and robustness of our estimated costs for single hops and walking strides, which are based on instances in which animals were uncharacteristically using a single gait, we compared total aerobic costs calculated from these estimates with actual aerobic costs for longer periods (up to 20 min) when toads used natural mixed gaits. The costs were calculated using the estimated aerobic cost equation (equation 2). A small discrepancy between actual \dot{V}_{O_2} and that calculated from the estimated costs of hopping and walking strides

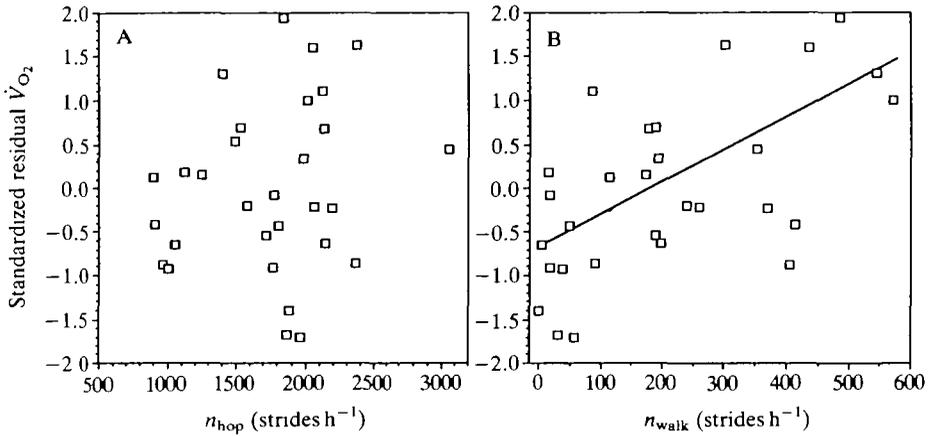


Fig. 5. Effect of the number of hopping and walking strides on the standardized residual \dot{V}_{O_2} of exercising toads. Standardized residuals were calculated from multiple regressions and represent the extent to which each individual measurement is greater (positive residual) or smaller (negative residual) than the value expected for a toad of equivalent mass at that speed. (A) No correlation between the number of hops per unit time (n_{hop}) and standardized residual \dot{V}_{O_2} . (B) Positive correlation between the number of walks per unit time (n_{walk}) and standardized residual \dot{V}_{O_2} .

would lend credence to the estimates. The average percentage discrepancy between the estimated and observed costs of locomotion was $2.4 \pm 3.0\%$, s.e.; $N=30$).

Lactate and phosphocreatine concentrations

The lactate concentrations of the thigh and calf muscles under the various experimental conditions were each compared with one-way analysis of variance (ANOVA) and Scheffe's *F*-test. The only experimental treatment to increase lactate significantly ($P < 0.05$) above resting levels was exercise to fatigue during natural locomotion at 0.63 km h^{-1} (Table 1). Lactate concentrations did not increase significantly between 10 and 30 min of exercise at 0.36 km h^{-1} , a speed 33% greater than the MAS but less than the MSS. Creatine phosphate levels did not vary significantly among different exercise regimes in either the calf or the thigh (Table 1).

Discussion

Gait changes

Our data support the contention of Walton and Anderson (1988) that toads change gait with speed. As toads increased speed, they increased the proportion of strides that were hops and decreased the proportion of walking strides (Fig. 1B–E). The gait change of toads differs from that of other quadrupeds. For example, horses change gait from walking to trotting and trotting to galloping

Table 1. *Lactate and creatine phosphate concentrations of toad leg muscle*

Exercise regime	N	Lactate		Creatine phosphate	
		Calf	Thigh	Calf	Thigh
Rest	6	2.20±0.52	1.61±0.27	10.45±1.43	10.32±1.53
Walk to fatigue (0.14 km h ⁻¹)	4	3.84±0.92	3.17±0.60	10.67±3.87	6.67±1.76
Natural locomotion (10 min; 0.36 km h ⁻¹)	6	5.12±0.54	3.97±0.62	7.02±0.81	8.98±1.37
Natural locomotion (30 min; 0.36 km h ⁻¹)	4	4.18±0.91	3.30±0.67	9.66±2.36	8.17±4.75
Natural locomotion to fatigue (0.63 km h ⁻¹)	5	6.35*±0.66	5.27*±0.75	7.23±0.46	8.13±1.19

Concentrations in $\mu\text{mol g}^{-1}$ muscle.
 Values are mean±S.E.
 *Significantly different from resting levels, $P<0.05$.

well-defined speeds (Hoyt and Taylor, 1981). Toads, by contrast, mix gaits over a broad range of speeds and gradually increase the relative proportion of hops as speed increases.

Animals may change gait for several reasons. (1) Gait changes in horses, goats and dogs appear to reduce bone strain (Biewener and Taylor, 1986; Farley and Taylor, 1989). Bone strain reduction is unlikely to figure in the gait change of toads. When toads land after the aerial phase of hopping, they probably exert more strain and have greater impact loads on their bones than during walking. In fact, by switching to hopping toads probably increase their chance of injury. (2) A gait change may circumvent mechanical constraints on walking at high speeds. Toads cannot maintain moderate and high speeds of locomotion by pure walking because their relatively short forelimbs and elongate hindlimbs prevent them from increasing stride length appreciably during walking. As walking speed increases, toads become statically unstable and significantly increase their degree of body pitching (Barclay, 1946). Thus, toads must switch to hopping to attain high speeds. (3) Reduction in energy cost may explain gait changes (Hoyt and Taylor, 1981). Changing gait can act to reduce muscle stress (i.e. force/cross-sectional area; Taylor, 1985). Reducing muscle stress may result in a saving of metabolic energy. By training horses to extend a gait to higher and lower speeds, Hoyt and Taylor (1981) showed that horses decrease the metabolic cost of locomotion by changing gait. Likewise, mink (Williams, 1983) and squirrels (Hoyt and Kenagy, 1988) decrease the energy cost to travel a given distance when changing gait from a walk to a bound. The present data on toads indicate that hopping a given distance is only about half as expensive as walking the same distance.

Costs of a walking stride, a hop and a mixed gait

We calculated the cost of walking strides and hops from intervals in which

animals were either forced to adopt a single gait or did so spontaneously but briefly. These circumstances are unlike the normal locomotion of a toad, which is a combination of hops and walking strides. Nonetheless, several lines of evidence suggest that our estimates are reasonable.

With respect to the gait kinematics, walking strides were always 2–4 cm long, during natural locomotion and forced walking, regardless of locomotor speed. Walking stride length and speed were not correlated in our shortened treadmill in the narrow speed range within which walking occurred. Thus, our estimated walking stride cost of $2.5 \times 10^{-4} \text{ ml O}_2 \text{ g}^{-1} \text{ stride}^{-1}$ is likely to be applicable to the range of speeds used during natural locomotion (Fig. 2). Surprisingly, the cost of a walking stride in a toad (i.e. $5.0 \text{ J kg}^{-1} \text{ stride}^{-1}$) is remarkably similar to that predicted from size-scaling relationships derived from mammalian locomotion (i.e. $5.6 \text{ J kg}^{-1} \text{ stride}^{-1}$; Heglund and Taylor, 1988). We calculated hop costs from periods during which toads were moving freely (Fig. 3). Thus, although we used only periods in which the animals hopped almost exclusively to estimate hop costs, the kinematics of hops in these periods did not differ from that in natural locomotion.

The cost of a hop ($4.1 \times 10^{-4} \text{ ml O}_2 \text{ g}^{-1} \text{ hop}^{-1}$) was greater than the cost of a walking stride. However, individual hops were much longer than walking strides, which more than offset the greater cost of a hop (Fig. 4). From stride frequency and stride length, we calculated the cost to traverse a given distance at 0.1 km h^{-1} . This calculated cost of transport was $8.2 \times 10^{-5} \text{ ml O}_2 \text{ g}^{-1} \text{ cm}^{-1}$ during forced walking and $4.2 \times 10^{-5} \text{ ml O}_2 \text{ g}^{-1} \text{ cm}^{-1}$ for bouts of exclusive hopping during natural locomotion. Thus, to traverse a given distance by walking was about 1.9 times more expensive than hopping the same distance at this speed. This is a conservative estimate of the energy savings of hopping for two reasons. First, our calculation uses a value for walking stride length of 3 cm, which is in the upper range of walking stride lengths observed. If a shorter distance, such as 2 or 2.5 cm, is used, walking a unit distance becomes even more expensive. Second, our calculations are for a slow speed at which hop length is relatively small. Even though hop cost increased with hop length, the regression coefficient of this relationship is less than one. Thus, the difference in the cost per distance between hopping and walking increases with speed. These differences in the cost per distance are consistent with the hypothesis that a gait change in toads conserves metabolic energy during locomotion.

The natural mixed gait reflects differences in the cost of walking and hopping (Fig. 5). Individual toads that tended to walk more than hop had a significantly higher cost of locomotion. Similarly, cost estimates of the natural mixed gaits that incorporate the numbers of component hops and walking strides closely resemble actual measurements of total cost. The average discrepancy between the actual and estimated cost was $2.4 \pm 3.0\%$ (s.e.). These discrepancies are small considering that they reflect relatively long intervals of locomotion. Any small error in the cost of a hop or a walking stride would inflate the discrepancy between actual and estimated costs.

Consequences of a gait change

Walton and Anderson (1988) proposed that a gait change in toads reduces the energetic cost of locomotion, but did not test this suggestion. The present study supports their hypothesis and is the first to report on the energetic consequence of a gait change in an amphibian (see review of Gatten *et al.* 1992).

The differential energetic costs of walking and hopping can be used to explain the exceptional endurance of toads at speeds that appear to be above that at which oxygen consumption ($\dot{V}_{O_{2max}}$) is maximal. $\dot{V}_{O_{2max}}$ is often linked with the MSS. In salamanders and lizards, the MAS is equal to the speed at which activity can be sustained (Bennett and John-Alder, 1984; John-Alder and Bennett, 1981; Full *et al.* 1988). All else being equal, animals with a greater $\dot{V}_{O_{2max}}$ can sustain higher speeds. Likewise, a reduced cost of locomotion will cause $\dot{V}_{O_{2max}}$ to be attained at higher speeds and, therefore, can increase endurance (Full *et al.* 1988).

Toads have a high aerobic scope compared with those of other amphibians (Bennett and Licht, 1973; Carey, 1979; Hillman, 1976; Miller and Hutchison, 1980), and toad sartorius muscle has a significantly greater mitochondrial content than muscle from other amphibians (Renaud and Stevens, 1983). This greater aerobic capacity and high $\dot{V}_{O_{2max}}$ may underlie the relatively great endurance of toads (Putnam and Bennett, 1981). However, a high $\dot{V}_{O_{2max}}$ alone cannot explain the exceptional endurance capacity of toads above the MAS.

Metabolic cost can also affect the MAS (Full *et al.* 1988). In toads, changing gait decreased the cost of travelling without changing the $\dot{V}_{O_{2max}}$. Changing gait to a hop made travelling a given distance less energetically expensive. The effect of the decreased cost of locomotion due to the gait change can be seen in Fig. 6. We multiplied the average hop frequency for exclusive hopping at a given speed by the cost per hop. We then added these rates to $\dot{V}_{O_{2rest}}$ to give the cost of 'exclusive hopping'. The estimated cost of exclusive hopping is less than the cost of natural locomotion at speeds of 0.45 km h⁻¹ or below, but rises and approaches the cost of natural locomotion at higher speeds. This pattern reflects the more economical hopping gait replacing the less economical walking as speed increases. Accordingly, not until the cost of exclusive hopping exceeds the actual $\dot{V}_{O_{2max}}$ does fatigue ensue; the speed at which the two costs are equivalent is close to the MSS. Presumably the constant \dot{V}_{O_2} at speeds between the MAS and the MSS represents the savings realized from the change in gait.

Creatine phosphate depletion and lactate accumulation

During natural locomotion and forced walking below the MSS, phosphocreatine and lactate concentrations were not significantly different from resting levels (Table 1). These concentrations were similar to those measured in other amphibians, and toads in particular (Bennett and Licht, 1973; Hutchison *et al.* 1981; Miller and Sabol, 1989; Putnam, 1979; Withers *et al.* 1988). Our data differ from those of Taigen and Beuchat (1984), who found significant accumulation of lactate in *Bufo americanus* well below $\dot{V}_{O_{2max}}$ after only a few minutes of exercise. The differences

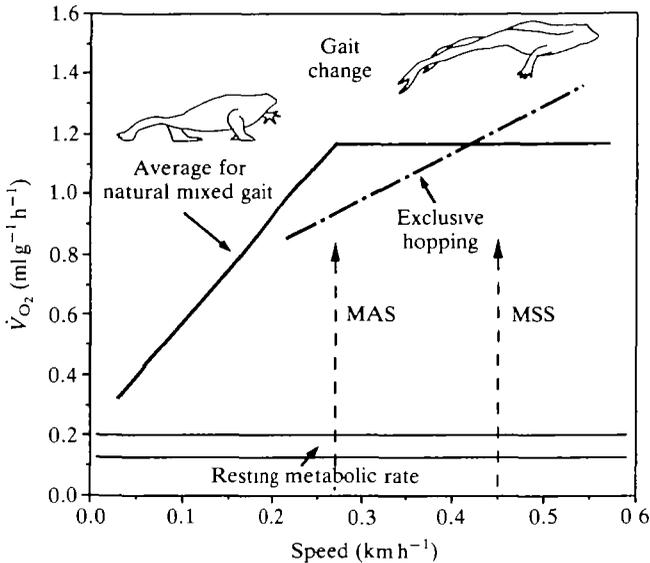


Fig. 6. Comparison of the aerobic costs for natural, mixed-gait locomotion and exclusive hopping as a function of speed. The measured costs of the natural mixed gait are from Walton and Anderson (1988). The cost of exclusive hopping was estimated from the stride frequency measured during intervals of exclusive hopping (Fig. 4B) and the aerobic cost per hop of the indicated length (Fig. 4A). The cost of exclusive hopping was less than that for natural locomotion at speeds below the maximum aerobic speed (MAS) and did not exceed maximal oxygen consumption ($\dot{V}_{O_2, \max}$) until the maximum sustainable speed (MSS). The cost of exclusive hopping will better represent \dot{V}_{O_2} at speeds above MAS, where toads change from more frequent walking to more frequent hopping.

in lactate concentration may be accounted for by the different methods used to control activity.

If activity above the MAS was powered by anaerobic glycolysis and toad muscle was fatigue resistant, then lactate should have accumulated, certainly after 30 min (Table 1). On the contrary, toad muscle had a lower tolerance to lactate accumulation than frog muscle in isolated muscle studies (Putnam and Bennett, 1983). Also, lactate only accumulated significantly during natural locomotion when toads exercised to fatigue at high speeds (0.63 km h^{-1}) above MSS. Thus, at speeds above the MAS and below the MSS, no net anaerobiosis was apparent. We cannot rule out the possibility that significant amounts of lactate were produced and then rapidly cleared. However, studies on *Bufo americanus* (Withers *et al.* 1988) suggest that lactate is cleared slowly after activity, with the largest fraction being resynthesized back to glycogen.

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References

- BARCLAY, O. R. (1946). The mechanics of amphibian locomotion. *J. exp. Biol.* **23**, 177-203.
- BARTHOLOMEW, G. A., VLECK, D. AND VLECK, C. M. (1981). Instantaneous measurement of oxygen consumption during pre-flight warm-up and post-flight cooling in sphingid moths. *J. exp. Biol.* **90**, 17-34.
- BENNETT, A. F. (1982). The energetics of reptilian activity. In *Biology of the Reptilia*, vol. **13** (ed. C. Gans and F. H. Pough), pp. 155-199. New York: Academic Press.
- BENNETT, A. F. AND JOHN-ALDER, H. B. (1984). The effect of body temperature on the locomotor energetics of lizards. *J. comp. Physiol.* **155**, 21-27.
- BENNETT, A. F. AND LICHT, P. (1972). Anaerobic metabolism during activity in lizards. *J. comp. Physiol.* **81**, 277-288.
- BENNETT, A. F. AND LICHT, P. (1973). Relative contributions of anaerobic and aerobic energy production during activity in Amphibia. *J. comp. Physiol.* **87**, 351-360.
- BENNETT, A. F. AND LICHT, P. (1974). Anaerobic metabolism during activity in amphibians. *Comp. Biochem. Physiol.* **48A**, 319-327.
- BERGMEYER, H. V. (1974). *Methods of Enzymatic Analysis*, vol. III. New York: Academic Press.
- BIEWENER, A. A. AND TAYLOR, C. R. (1986). Bone strain: a determinant of gait and speed? *J. exp. Biol.* **123**, 383-400.
- CAREY, C. (1979). Aerobic and anaerobic energy expenditure during rest and activity in montane *Bufo b. boreas* and *Rana pipiens*. *Oecologia* **39**, 213-228.
- DAWSON, T. J. AND TAYLOR, C. R. (1973). Energetic cost of locomotion in kangaroos. *Nature* **246**, 313-314.
- FARLEY, C. AND TAYLOR, C. R. (1989). Gait transitions: mechanical and energetic constraints. *Proc. int. Union physiol. Sci.* **17**, 430.
- FULL, R. J. (1986). Locomotion without lungs: Energetics and performance of a lungless salamander. *Am. J. Physiol.* **251**, R775-R780.
- FULL, R. J. (1987). Locomotion energetics of the ghost crab. I. Metabolic cost and endurance. *J. exp. Biol.* **130**, 137-153.
- FULL, R. J., ANDERSON, B. D., FINNERTY, C. M. AND FEDER, M. E. (1988). Exercising with and without lungs. I. The effects of metabolic cost, maximal oxygen transport and body size on terrestrial locomotion in salamander species. *J. exp. Biol.* **138**, 471-485.
- FULL, R. J. AND HERREID, C. F. (1983). The aerobic response to exercise of the fastest land crab. *Am. J. Physiol.* **244**, R530-R536.
- FULL, R. J. AND HERREID, C. F. (1984). Fiddler crab exercise: energetic cost of running sideways. *J. exp. Biol.* **109**, 141-161.
- GATTEN, R. E., JR, MILLER, K. AND FULL, R. J. (1992). Energetics of amphibians at rest and during locomotion. In *Environmental Physiology of Amphibians* (ed. M. E. Feder and W. W. Burggren). Chicago: University of Chicago Press (in press).
- HEGLUND, N. C. AND TAYLOR, C. R. (1988). Speed, stride frequency and energy cost per stride: how do they change with body size and gait? *J. exp. Biol.* **138**, 301-318.
- HERREID, C. F. (1981). Energetics of pedestrian arthropods. In *Locomotion and Energetics in Arthropods*. (ed. C. F. Herreid and C. R. Fournier), pp. 491-526. New York: Plenum Press.
- HERREID, C. F., FULL, R. J. AND PRAWEL, D. A. (1981). Energetics of cockroach locomotion. *J. exp. Biol.* **94**, 189-202.
- HILLMAN, S. S. (1976). Cardiovascular correlates of maximal oxygen consumption rates in anuran amphibians. *J. comp. Physiol.* **109**, 199-207.
- HOYT, D. F. AND KENAGY, G. J. (1988). Energy costs of walking and running gaits and their aerobic limits in golden-mantled ground squirrels. *Physiol. Zool.* **61**, 34-40.

- HOYT, D. F. AND TAYLOR, C. R. (1981). Gait and the energetics of locomotion in horses. *Nature* **292**, 239–240.
- HUTCHISON, V. H., MILLER, K. AND GRATZ, R. K. (1981). The anaerobic contributions to sustained activity in the giant toad *Bufo marinus*. *Comp. Biochem. Physiol.* **69A**, 693–696.
- JOHN-ALDER, H. B. AND BENNETT, A. F. (1981). Thermal dependence of endurance and locomotory energetics in a lizard. *Am. J. Physiol.* **241**, R342–R349.
- MILLER, K. AND HUTCHISON, V. H. (1980). Aerobic and anaerobic scope for activity in the giant toad, *Bufo marinus*. *Physiol. Zool.* **53**, 170–175.
- MILLER, K. AND SABOL, J. L. (1989). The role of phosphocreatine breakdown in the metabolic support of locomotion by clawed frogs, *Xenopus laevis*. *Comp. Physiol. Biochem.* **93B**, 251–254.
- PUTNAM, R. W. (1979). The basis for differences in lactic acid content after activity in different species of anuran amphibians. *Physiol. Zool.* **52**, 509–519.
- PUTNAM, R. W. AND BENNETT, A. F. (1983). Histochemical, enzymatic, and contractile properties of skeletal muscles of three anuran amphibians. *Am. J. Physiol.* **244**, R558–R567.
- RENAUD, J. M. AND STEVENS, E. D. (1983). A comparison between field habitats and contractile performance of frog and toad sartorius muscle. *J. comp. Physiol.* **151**, 127–131.
- SEEHERRMAN, H. J., TAYLOR, C. R., MALOY, G. M. O. AND ARMSTRONG, R. B. (1981). Design of the mammalian respiratory system: Measuring maximum aerobic capacity. *Respir. Physiol.* **44**, 11–24.
- TAIGEN, T. L. AND BEUCHAT, C. A. (1984). Anaerobic threshold of anuran amphibians. *Physiol. Zool.* **57**, 641–647.
- TAYLOR, C. R. (1978). Why change gaits? Recruitment of muscles and muscle fibers as a function of speed and gait. *Am. Zool.* **18**, 153–161.
- TAYLOR, C. R. (1985). Force development during sustained locomotion: a determinant of gait, speed and metabolic power. *J. exp. Biol.* **115**, 253–262.
- WALTON, M. AND ANDERSON, B. D. (1988). The aerobic cost of saltatory locomotion in the Fowler's toad (*Bufo woodhousei fowleri*). *J. exp. Biol.* **136**, 273–288.
- WILLIAMS, T. M. (1983). Locomotion of the North American mink, a semi-aquatic mammal. II. The effect of an elongate body on running energetics. *J. exp. Biol.* **48**, 153–161.
- WITHERS, P. C., LEA, M., SOLBERG, T. C., BAUSTIAN, M. AND HEDRICK, M. (1988). Metabolic fates of lactate during recovery from activity in an anuran amphibian, *Bufo americanus*. *J. exp. Zool.* **246**, 236–243.