

# Locomotion without lungs: energetics and performance of a lungless salamander

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FULL, R. J. *Locomotion without lungs: energetics and performance of a lungless salamander*. Am. J. Physiol. 251 (Regulatory Integrative Comp. Physiol. 20): R775–R780, 1986.—Lungless salamanders (4.1 g) were exercised on a treadmill enclosed in a Plexiglas respirometer at a range of speeds (0.05–0.24 km/h). O<sub>2</sub> consumption ( $\dot{V}O_2$ ) was determined continuously by open-flow respirometry. At the onset of exercise  $\dot{V}O_2$  increased to a “steady state” in ~2–5 min.  $\dot{V}O_2$  (ml O<sub>2</sub>·g<sup>-1</sup>·h<sup>-1</sup>) increased linearly with speed (S),  $\dot{V}O_2 = 2.3(S) + 0.1$  ( $r^2 = 0.74$ ). The minimum cost of transport (2.3 ml O<sub>2</sub>·g<sup>-1</sup>·km<sup>-1</sup>) was three-fourths of that predicted for a vertebrate of the same mass. Maximum O<sub>2</sub> consumption ( $\dot{V}O_{2\max}$ ) was attained at 0.16 km/h (maximum aerobic speed), where this  $\dot{V}O_2$  was six to nine times above standard rates. The net rate of whole-body lactate production (WBL) was insignificant at slow speeds (<46%  $\dot{V}O_{2\max}$ ) but did increase at submaximal work loads (85%  $\dot{V}O_{2\max}$ ). The highest WBL (0.07 mg·g<sup>-1</sup>·min<sup>-1</sup>) was measured at a speed (0.20 km/h) that exceeded the maximum aerobic speed. Salamanders sustained exercise at slow speeds for over 2 h. The greatest decline in endurance (i.e., from 120 to 10 min) occurred at speeds below the maximum aerobic speed, where being lungless may have little consequence. At fast rates of locomotion a limited  $\dot{V}O_{2\max}$  was associated with a low maximum aerobic speed and a modest capacity for sustained activity.

amphibian; oxygen consumption; lactate production; endurance; treadmill locomotion

IF THE ABSENCE OF LUNGS significantly constrains gas exchange, then it might be expected to restrict activities such as locomotion that require increased and sometimes maximal rates of O<sub>2</sub> consumption ( $\dot{V}O_2$ ). Possible consequences of lunglessness, such as an inability to increase  $\dot{V}O_2$  rapidly at the onset of exercise, a low maximum rate of O<sub>2</sub> consumption ( $\dot{V}O_{2\max}$ ), and an increased reliance on anaerobic metabolism, could be associated with a limited capacity to sustain locomotion.

Previous studies of the consequences of lunglessness for activity have examined the highest rates of  $\dot{V}O_2$  attained during forced activity (6, 23, 42, 43), recovery from activity with respect to acid-base balance (3), or the size of the O<sub>2</sub> debt (10), and the ability of respiratory surface area to support high rates of  $\dot{V}O_2$  (9). These measurements, however, do not directly address the consequences of lunglessness for an animal's ability to locomote. A major problem has been the lack of an adequate method to control and quantify locomotion. Instead, amphibians have been shocked, overturned repeatedly, or prodded to yield activity that is presumably

maximal but not quantifiable in terms of speed or intensity (23, 35). As a result the relationship between energetics and endurance at known activity levels (e.g., speeds) remains obscure.

The present study differs from earlier work on amphibians in applying the treadmill techniques that have successfully characterized the relationship between the  $\dot{V}O_2$  and speed in many mammals (37), lizards (1), and invertebrates (12, 20, 21). It examines the potential consequences of lunglessness on salamander locomotion by exercising exclusively skin-breathing salamanders, *Plethodon jordani*, on a treadmill at controlled speeds for specified durations while measuring  $\dot{V}O_2$ , whole-body lactate, or endurance. The results demonstrate that this species has a considerable capacity for sustained locomotion at slow speeds; however, endurance at fast speeds is modest and is correlated with a low  $\dot{V}O_{2\max}$  and an increased dependence on anaerobic metabolism.

## MATERIALS AND METHODS

*Animals.* *P. jordani* [body mass =  $4.1 \pm 0.01$  (SE) g; snout-vent length =  $62.3 \pm 1.1$  (SE) mm] were obtained from Highlands, NC. Animals were maintained at 12–14°C on a 14:10 light-dark photoperiod centered at 1300 h local time. Animals were fed fly larvae regularly until 5 days before metabolic or endurance measurements. Individuals were exposed to the experimental temperature [ $27.2 \pm 0.25^\circ\text{C}$  (SE)] for at least 1 h before measurements.

*O<sub>2</sub> consumption.* Animals were exercised in an airtight Lucite respirometer that contained a variable-speed treadmill (19). After resting on the treadmill under dim light for 1 h, salamanders were exercised for 20 min at a single speed. The speeds selected were 0.05, 0.09, 0.13, 0.16, and 0.19 km/h. An attempt was made to exercise each salamander at every speed. A recovery period of 60 min followed the run. Animals were run only once a day and were given a minimum of 1 day between trials. Experiments in which animals did not run consistently were discarded.

$\dot{V}O_2$  was measured continuously during rest, exercise, and recovery using open-flow respirometry. Humidified air was drawn through the chamber by a pump at a known flow rate ( $\dot{V}$ ) determined by a flowmeter. The air exiting the chamber was pulled through a column containing CaSO<sub>4</sub> (Drierite) and Ascarite to remove water

and  $\text{CO}_2$ , respectively. The incurrent fractional  $\text{O}_2$  concentration ( $\text{FI}_{\text{O}_2}$ ) was compared with the excurrent fraction ( $\text{FE}_{\text{O}_2}$ ) by a dual-channel  $\text{O}_2$  analyzer (Ametek Applied Electrochemistry). The analyzer was interfaced with an analog-to-digital converter (Isaac 41A system, Cyborg) and a computer (Apple II<sup>+</sup>). The  $\text{O}_2$  concentrations were sampled at 10-s intervals and averaged to produce a single value every 20 s. N-point smoothing ( $n = 4$ ) was conducted on all data to remove the noise found in control tests where  $\text{O}_2$  concentration was held constant.

Instantaneous  $\dot{V}\text{O}_2$  was calculated from the  $\text{FI}_{\text{O}_2}$  and  $\text{FE}_{\text{O}_2}$  (12, 13, 19). Calculations of instantaneous  $\dot{V}\text{O}_2$  provide accurate estimates of rapid changes in metabolic rate to allow identification of the steady-state  $\dot{V}\text{O}_2$ . A single rapid change in  $\dot{V}\text{O}_2$  will result in only gradual changes in  $\text{FE}_{\text{O}_2}$  within the metabolic chamber toward an equilibrium value ( $F_{\text{eq}}$ ). Given any two measurements separated by a brief interval and the "washout" characteristics of the chamber,  $F_{\text{eq}}$  can be calculated and substituted for  $\text{FE}_{\text{O}_2}$  in a standard equation

$$\dot{V}\text{O}_2 = \dot{V} (\text{FI}_{\text{O}_2} - F_{\text{eq}}) / (1 - \text{FI}_{\text{O}_2})$$

to estimate instantaneous  $\dot{V}\text{O}_2$ . Gas flowrate was 37 ml/min. The response time of the 380-ml chamber was 2 min to 50% full-scale deflection (0–0.100%  $\text{O}_2$ ). This was determined by addition of  $\text{N}_2$  gas to the chamber. All values were corrected to STP.

**Whole-body lactate.** Whole-body lactate (WBL) content was determined by a modification of the procedure of Full and Herreid (13). Animals were weighed the day before measurement and kept undisturbed. After a 1-h rest period animals were exercised at one of three speeds (0.05, 0.13, or 0.20 km/h). After the rest period or at 5, 10, or 20 min during the exercise bout, the animal was carefully removed from the treadmill and frozen in liquid  $\text{N}_2$ . One group of animals was exercised to exhaustion at 0.20 km/h. The time required for freezing was <2 s. After freezing the animal was homogenized in ~5 vol of 0.6 N perchloric acid. The L-lactate in the supernatant was measured with a spectrophotometric assay (Sigma diagnostic kit 826-UV).

**Endurance capacity.** Salamanders were exercised to fatigue on a motor-driven treadmill after a 10-min rest period. Running speeds ranged from 0.04 to 0.24 km/h. Animals maintained a consistent pace without prodding until just before fatiguing. Fatigue was defined as that time when an animal 1) did not maintain pace with the treadmill, 2) dragged its abdomen, and 3) did not respond to three successive prodding attempts. Animals were run only once a day and were given a minimum of 4 days between trials. Trials in which animals did not walk consistently were discarded.

## RESULTS

**$\text{O}_2$  consumption.** The kinetics of mass specific  $\dot{V}\text{O}_2$  are shown in Fig. 1. *P. jordani* exhibited only a small amount of exploratory behavior during the rest period. Standard rates for *P. jordani* measured previously equaled  $0.054 \text{ ml O}_2 \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  (42), whereas estimates based on allometric equations for lungless salamanders are somewhat

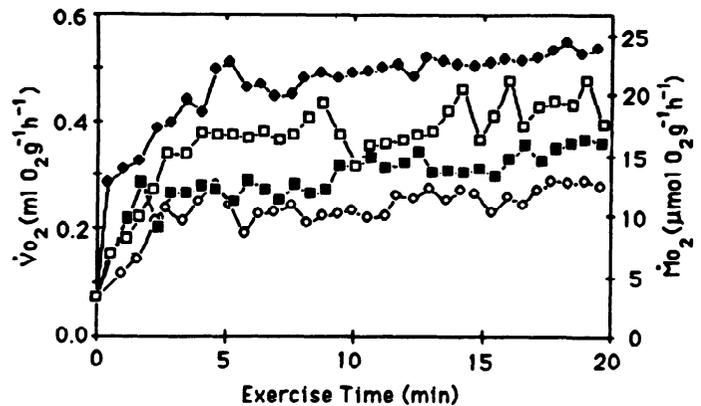


FIG. 1.  $\text{O}_2$  consumption ( $\dot{V}\text{O}_2$ ) of a salamander exercising on a treadmill at 0.16 (closed diamonds), 0.13 (open squares), and 0.09 (closed squares) km/h.  $\text{MO}_2$  represents  $\text{O}_2$  consumption in  $\mu\text{mol} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$ .

higher ( $0.072 \text{ ml O}_2 \cdot \text{g}^{-1} \cdot \text{h}^{-1}$ ) (5). The  $\dot{V}\text{O}_2$  during the final 15 min of the preexercise period averaged  $0.14 \pm 0.01$  (SE)  $\text{ml O}_2 \cdot \text{g}^{-1} \cdot \text{h}^{-1}$ .

During the first 5 min after the start of exercise,  $\dot{V}\text{O}_2$  increased rapidly to an approximate "steady state" ( $\dot{V}\text{O}_{2\text{ss}}$ ).  $\dot{V}\text{O}_2$  increased slowly thereafter. The slow component (i.e.,  $\dot{V}\text{O}_2$  drift) was most prominent during exercise at the faster velocities (Fig. 1). The time it takes to attain 50% of the  $\dot{V}\text{O}_{2\text{ss}}$  ( $t_{1/2\text{on}}$ ) was used to evaluate the initial transient. Analysis of variance revealed no significant effect of speed on  $t_{1/2\text{on}}$  ( $P > 0.05$ ). The pooled mean  $t_{1/2\text{on}}$  was  $100 \pm 21$  (SE) s ( $n = 20$ ).

$\dot{V}\text{O}_{2\text{ss}}$  during the exercise period was determined by averaging all  $\dot{V}\text{O}_2$  values from 5 to 20 min.  $\dot{V}\text{O}_{2\text{ss}}$  showed a significant increase with speed ( $F(2,29) = 148$ ;  $P < 0.001$ ; Fig. 2). A test for linearity failed to reject the hypothesis of a linear relationship for  $\dot{V}\text{O}_{2\text{ss}}$  vs. speed [ $F(2,26) = 0.51$ ;  $P > 0.05$ ]. The least-squares regression equation based on 30 trials was  $\dot{V}\text{O}_{2\text{ss}} = 2.25 (\text{S}) + 0.10$  ( $r^2 = 0.74$ ), where S represents speed in kilometers per hour and  $\dot{V}\text{O}_2$  is in milliliters  $\text{O}_2$  per gram per hour. The 95% confidence interval for the slope was  $1.9\text{--}2.6 \text{ ml O}_2 \cdot \text{g}^{-1} \cdot \text{km}^{-1}$ . Stepwise multiple regression analysis showed that including body mass and temperature as independent variables along with speed did not significantly improve the explanation of  $\dot{V}\text{O}_{2\text{ss}}$  variance ( $P > 0.05$ ).

$\dot{V}\text{O}_{2\text{ss}}$  did not increase with an increase in speed at speeds  $>0.16$  km/h. At these fast speeds (0.19 km/h), some animals fatigued in <10 min and a  $\dot{V}\text{O}_{2\text{ss}}$  was not attained. Therefore the best estimate of  $\dot{V}\text{O}_{2\text{max}}$  was taken to be the  $\dot{V}\text{O}_{2\text{ss}}$  at 0.16 km/h (Fig. 2).

The volume of  $\text{O}_2$  used during 60 min of recovery was significantly greater at faster speeds;  $0.16 \text{ ml O}_2 \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  at 0.05 km/h,  $0.27 \text{ ml O}_2 \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  at 0.09 km/h, and  $0.31 \text{ ml O}_2 \cdot \text{g}^{-1} \cdot \text{h}^{-1}$  at 0.13 km/h (ANOVA,  $F(2,12) = 247$ ;  $P < 0.001$ ).

**Whole-body lactate.** The mean WBL content determined after the preexercise period for five animals was  $0.157 \pm 0.007$  (SE) mg/g animal. WBL was not significantly elevated above resting levels after either 5 or 20 min of exercise at 0.05 km/h (ANOVA;  $P > 0.05$ ; Fig. 3). Exercise for 5 min at 0.13 and 0.20 km/h did result in a significant increase in WBL ( $P < 0.05$ ). The highest WBL contents were found for salamanders exercised to

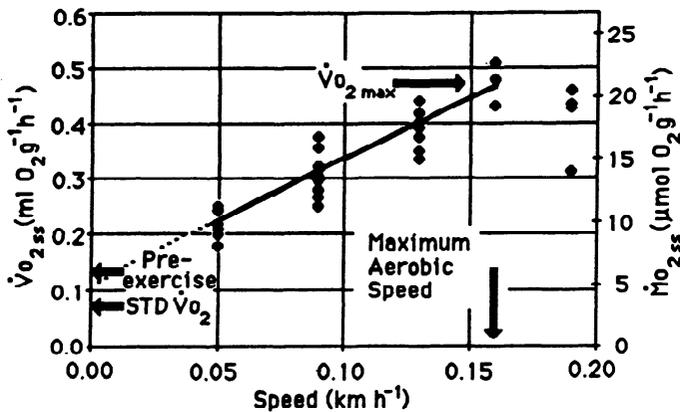


FIG. 2. Steady-state  $\dot{V}O_{2ss}$  as a function of speed. Maximal  $\dot{V}O_{2max}$  is shown at right, along with speed at which  $\dot{V}O_{2max}$  is attained (maximal aerobic speed). Preexercise rates are from present study. Standard (STD) rates are from Feder (5).  $\dot{M}O_{2ss}$  represents steady-state  $O_2$  consumption in  $\mu\text{mol}\cdot\text{g}^{-1}\cdot\text{h}^{-1}$ .

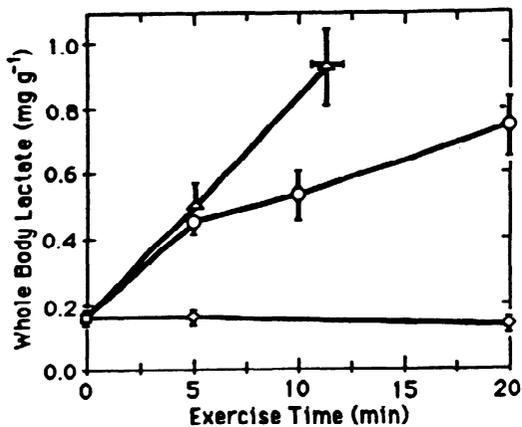


FIG. 3. Time course of whole-body lactate content during exercise at 0.20 (triangles), 0.13 (circles), and 0.05 (squares) km/h. Animals fatigued at 0.20 km/h for durations  $>5$  min. Bars represent  $\pm$ SE.

fatigue at 0.20 km/h (mean endurance time = 11.4 min) and for those exercised for 20 min at 0.13 km/h.

The net rate of WBL production increased as a function of speed (analysis of covariance,  $F(2,47) = 49$ ;  $P < 0.001$ ). Net WBL production at 0.20 km/h ( $0.069 \text{ mg}\cdot\text{g}^{-1}\cdot\text{min}^{-1}$ ) was greater than levels at 0.13 km/h ( $0.027 \text{ mg}\cdot\text{g}^{-1}\cdot\text{min}^{-1}$ ; Newman-Keuls multiple range test,  $q = 6.6$ ;  $P < 0.05$ ). Body mass was not significantly correlated with WBL content when speed and exercise time were held constant (partial correlation coefficient,  $r = 0.065$ ;  $P = 0.335$ ).

**Endurance capacity.** *P. jordani* walked steadily on the treadmill, especially at the lower speeds (i.e.,  $<0.14$  km/h). Exercise at faster speeds resulted in somewhat more intermittent locomotion in which salamanders tended to accelerate to the front of the chamber after contacting the back wall and then ride backwards. Large-amplitude lateral undulations of the trunk occurred as the animal accelerated forward at these speeds.

*P. jordani* could sustain locomotion for  $>1$  h at speeds  $<0.10$  km/h (Fig. 4). Six individuals walked at these low speeds for 2 h at which time trials were terminated. At speeds ranging from 0.10 to 0.14 km/h locomotor capacity decreased by  $\sim 60\%$  to near 20 min. Endurance time continued to decline at faster velocities (0.14–0.24 km/

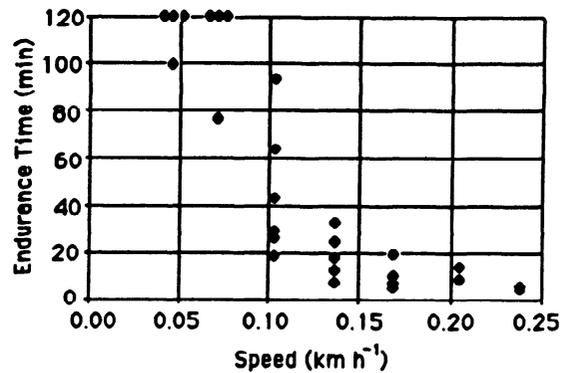


FIG. 4. Treadmill endurance (time to fatigue) as a function of speed. Each data point represents a separate trial. Experiments were terminated after 120 min of exercise if fatigue did not occur.

h), but did so at a slower rate. Animals exercising at 0.24 km/h could sustain locomotion for only 5 min or less.

## DISCUSSION

Lungless salamanders must rely on cutaneous gas exchange for  $\dot{V}O_2$  during activity. Cutaneous respiration has been reported to be "diffusion limited," resulting in low arterial  $PO_2$  and offering little opportunity for physiological control (31). If cutaneous respiration imposes a limitation on  $O_2$  transport, then it could result in slow  $\dot{V}O_2$  kinetics early in exercise, a low  $\dot{V}O_{2max}$ , and heavy dependence on anaerobic metabolism. Slow  $\dot{V}O_2$  kinetics would result in a larger  $O_2$  deficit at the onset of exercise and possibly a greater reliance on anaerobiosis. A low  $\dot{V}O_{2max}$  could set an upper limit on endurance at high speeds, since the additional energy, at speeds faster than those at which  $\dot{V}O_{2max}$  is attained, must be supplied by anaerobic sources. The limitation of a low  $\dot{V}O_{2max}$  would be particularly pronounced if the cost of locomotion were high (i.e., a steep slope of the  $\dot{V}O_{2ss}$  vs. speed function); a high cost of locomotion would result in  $\dot{V}O_{2max}$  being attained at relatively low speeds.

**$\dot{V}O_2$  kinetics.** The kinetics of  $\dot{V}O_2$  are significant in that they indicate the time required to initiate respiratory and circulatory responses that increase  $\dot{V}O_2$ . If cutaneous gas exchange is diffusion limited, then  $\dot{V}O_2$  during exercise could have relatively slow kinetics, since an increased  $O_2$  flux across the skin must be accomplished by an eventual decrease in arterial  $PO_2$  (i.e., an increase in the  $PO_2$  gradient across the skin resulting from a decrease in the  $PO_2$  of the arterial blood that travels to the skin). If the rate of  $O_2$  delivery during this adjustment does not meet the  $O_2$  demand by the muscles, then a greater reliance on anaerobic energy sources, such as high-energy phosphates and anaerobic glycolysis, should result.

The  $\dot{V}O_2$  kinetics of *P. jordani* at the onset of exercise ( $t_{1/2on} = 100$  s) are similar to those of mammals, which ventilate lungs ( $t_{1/2on} = 25$ –90 s), and some invertebrates, which rely on trachea ( $t_{1/2on} = 30$ –180 s) or gills ( $t_{1/2on} = 50$ –150 s) (4, 12, 20, 21, 29). The  $\dot{V}O_2$  increased from resting rates to a "steady state" in  $\sim 2$ –5 min (Fig. 1). Previous studies of uncontrolled, but intense, activity in lungless salamanders found that the  $\dot{V}O_2$  attained its highest rate in 3 min (23, 42). No increase in net WBL

production was found after 20 min for *P. jordani* during slow speeds of locomotion (Fig. 3). WBL increased during the first 5 min at faster speeds (0.13 km/h), but the net rate of production decreased as  $\dot{V}O_{2\text{ss}}$  was attained. Thus, with respect to  $\dot{V}O_2$  kinetics, lunglessness does not appear to handicap salamanders. Feder and Burggren (9) have recently contended that cutaneous gas exchange can be regulated by initiating blood flow through previously nonperfused capillaries. Plausibly, a rapid increase in "functional surface area" due to capillary recruitment could account for a more rapid increase in  $\dot{V}O_2$ .

**$\dot{V}O_{2\text{ss}}$  vs. speed.** The aerobic energy requirement of exercise increased in *P. jordani* as it moved faster.  $\dot{V}O_{2\text{ss}}$  increased linearly with speed (Fig. 2). This pattern is common in terrestrial locomotion for diverse animals (1, 12, 21, 37). Curvilinear relationships may occur within a single gait, but gait transitions may mask this relationship. It was not clear whether *P. jordani* changed gait (walk to trot). However, the increased contribution of lateral bending in the trunk was obvious as speed was increased. The data are insufficient to quantify any possible energetic consequences of this behavior.

The linear regression of  $\dot{V}O_{2\text{ss}}$  against speed did not extrapolate to resting levels at zero speed (Fig. 2); the Y-intercept was elevated 1.4–1.8 times above predicted standard  $\dot{V}O_2$  rates (5, 42). However, it was not significantly different from preexercise  $\dot{V}O_2$ . This elevation has been observed in many species (1, 37), and multiple explanations have been proposed (19). Whatever the cause, one consequence is an increased energetic cost at low speeds.

**Economy of locomotion.** The economy of salamander locomotion can be calculated from the  $\dot{V}O_{2\text{ss}}$  vs. speed relationship if  $\dot{V}O_{2\text{ss}}$  is divided by speed. The total cost of locomotion (amount of energy required by 1 g of animal to move 1 km) is a function of speed (Fig. 5). Locomotion at low speed is half as economical as moving at the fastest speeds. This hyperbolic function results from the relatively greater contribution of the Y-intercept at low speeds. At higher speeds the cost of locomotion approaches a minimum (slope of the  $\dot{V}O_{2\text{ss}}$  vs. speed function). The minimum cost of transport has been widely used for interspecific comparisons of economy (1, 19, 37). Body mass significantly affects the minimum

cost of transport, with small animals less economical than large animals on a mass-specific basis. *P. jordani* is actually somewhat more economical than would be predicted for a bird or mammal (37), or lizard (1), of its mass. These findings are in contrast to those for smaller lungless salamanders (2 g) walking in a rotating-wheel respirometer, which have much greater costs than those predicted for their size (8). Since these are the first measurements of locomotor economy in amphibians and for vertebrates of very small size, generalizations are not yet appropriate.

**Aerobic capacity.** The absolute  $\dot{V}O_{2\text{max}}$  determined herein by treadmill locomotion is nearly three times that reported previously for lungless salamanders of the same mass (6, 42). However, the  $\dot{V}O_{2\text{max}}$  of *P. jordani* is less than one-third the rate predicted for an anuran amphibian of the same mass (43). This low rate is consistent with diffusion limitations of cutaneous gas exchange operating at high speeds. However, because salamanders have some of the lowest metabolic rates found in vertebrates at rest (5, 7), the aerobic factorial scope ( $\dot{V}O_{2\text{max}}$ /standard  $\dot{V}O_2$ ) is not unusually small. The  $\dot{V}O_{2\text{max}}$  was six- to ninefold above standard rates (Fig. 2). This increase is nevertheless at the low end of the range of values (5- to 15-fold) reported for mammals (38), reptiles (1, 16, 27), amphibians being overturned repeatedly or prodded (22, 24, 34, 43), and invertebrates (12, 21). It is greater than the value reported previously for this species (4-fold) by repeated overturning for 3 min (42). An aerobic factorial scope of six- to ninefold does suggest that physiological control of cutaneous gas exchange is possible (i.e., controlling  $O_2$  flux through changes in blood flow). If this were not the case, then a six- to ninefold elevation in  $\dot{V}O_2$  must be accompanied by a six- to ninefold decrease in arterial  $PO_2$ , an unlikely occurrence, since the  $PO_2$  gradient can already be near 100 Torr at rest (31).

**Anaerobic energetics.** Lactate production is a function of the intensity as well as the time course of exercise. Net WBL production was insignificant during low-intensity exercise (<47%  $\dot{V}O_{2\text{max}}$ ). Exercise at levels that exceed 85% of the  $\dot{V}O_{2\text{max}}$  resulted in net WBL production in *P. jordani* (Fig. 3). The highest net rate of production occurred during the first 5 min, i.e., before  $\dot{V}O_{2\text{ss}}$  was attained. Accumulation of lactate at submaximal levels of work (50–80%  $\dot{V}O_{2\text{max}}$ ) has been reported for mammals (30, 32), reptiles (17), and anuran amphibians (33). The implications of this "anaerobic threshold" (40, 41) are controversial (18, 26). Such results do not necessarily indicate that an animal's  $O_2$  delivery system is limited, for evidence of tissue hypoxia is lacking. Furthermore, it is often difficult to separate the anaerobic energy mobilization necessary during the initial period of sustained exercise from that required during prolonged steady-state locomotion (36).

In *P. jordani*, most of the anaerobic glycolysis occurs early in the  $O_2$  deficit period, after which levels tend to stabilize or decrease. The contribution of the early lactate production to the energy required can be estimated if  $\dot{V}O_2$  and WBL production are converted to ATP equivalents using standard relationships (e.g., 1 mol  $O_2$  = 6.33

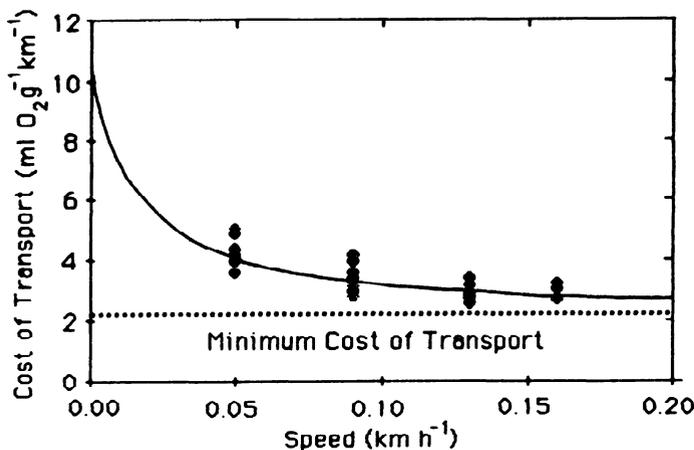


FIG. 5. Economy of locomotion (cost of transport) as a function of speed. Dashed line represents minimum cost of transport.

mol ATP and 1 mol lactate = 1.50 mol ATP). Anaerobic glycolysis supplied ~60% of the ATP during the first 5 min of exercise at 85%  $\dot{V}O_{2\max}$ , although providing only 15% during the rest of the exercise period. Net WBL production after 5 min of exercise at these speeds may simply reflect reliance on intermittent locomotion, during which fast glycolytic fibers may be recruited. At speeds above  $\dot{V}O_{2\max}$  anaerobic glycolysis contributed significantly, as net WBL content increased at a constant rate until fatigue (Fig. 3). Thirty to forty-five percent of the ATP was derived from accelerated glycolysis throughout the exercise bout.

**Endurance capacity.** *P. jordani* has a considerable capacity for sustained locomotion at slow speeds (<0.10 km/h). This lungless salamander can walk continuously for over 2 h, traveling distances exceeding 160 m (Fig. 4). The major decrease in stamina, and therefore distance traveled, occurs at speeds ranging from 0.10 to 0.14 km/h. These speeds are clearly below the speeds at which  $\dot{V}O_{2\max}$  is attained, so  $O_2$  transport may not play a major role in limiting endurance. Perhaps, as in mammals exercising at <60–80%  $\dot{V}O_{2\max}$ , fatigue is produced by the depletion of substrate in the muscle, such as glycogen (39).

At or above speeds where  $\dot{V}O_{2\max}$  is attained (>0.15 km/h), *P. jordani* fatigues rapidly (2–20 min) (Fig. 4).  $\dot{V}O_{2\max}$  has often been considered to be linked with the fastest sustainable speed. Few direct tests of this link between  $\dot{V}O_{2\max}$  and endurance have been attempted for terrestrial vertebrates. In lizards the speed at which  $\dot{V}O_{2\max}$  is attained (maximum aerobic speed, MAS) is positively correlated with the speed at which activity can be sustained for 15–30 min (2, 28). Garland (14, 15) has derived an allometric equation predicting the MAS as a function of body mass for lizards and mammals. MAS increases with body mass for both groups; however, the MAS for lizards is only 1/20 of that calculated for a mammal of a similar mass. This relationship predicts a MAS of 0.22 km/h for a 4-g lizard, somewhat faster than observed for *P. jordani* (0.16 km/h; Fig. 2). At the MAS, *P. jordani* could be active for only 15 min before fatiguing (Fig. 4). Lacertid lizards of the same mass have been shown to sustain speeds two to three times that of *P. jordani* for at least 15 min (25). Similarly, small invertebrates fatigue in 15 min at speeds ranging from 0.20 to 0.40 km/h (11–13). Recently it has been reported that another species of lungless salamanders has a MAS that is two orders of magnitude lower than predicted for lizards (8); however, these data were collected using a different apparatus, a rotating-wheel respirometer.

The low MAS of *P. jordani* could result from either a high minimum cost of transport (i.e., reaching a given  $\dot{V}O_{2\max}$  at a slower speed because the  $\dot{V}O_{2ss}$  vs. speed function has a steep slope) or from a low  $\dot{V}O_{2\max}$ . The cost of transport in *P. jordani* is not unusually high. Therefore the relatively low  $\dot{V}O_{2\max}$  is correlated with a low MAS and a modest capacity for sustained activity at fast speeds. Diffusion limitations at these speeds may very well impose a limitation on gas transport and therefore endurance. However, this conclusion must await further study of  $O_2$  transport during exercise.

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## REFERENCES

- BENNETT, A. F. The energetics of reptilian activity. In: *Biology of Reptila*, edited by C. Gans C. and F. H. Pough. New York: Academic, 1982, vol. 13, p. 155–199.
- BENNETT, A. F., AND H. B. JOHN-ALDER. The effect of body temperature on the locomotor energetics of lizards. *J. Comp. Physiol.* 155: 21–27, 1984.
- BOUTILIER, R. G., D. G. McDONALD, AND D. P. TOEWS. The effect of enforced activity on ventilation, circulation and blood acid-base balance in the aquatic gill-less urodele, *Cryptobranchus alleganiensis*, a comparison with the semi-terrestrial anuran, *Bufo marinus*. *J. Exp. Biol.* 84: 289–302, 1980.
- CERRETELLI, P., D. PENDERGAST, W. C. PAGANELLI, AND D. W. RENNIE. Effects of specific training on  $\dot{V}O_2$  on-response and early blood lactate. *J. Appl. Physiol.* 47: 761–769, 1979.
- FEDER, M. E. Lunglessness, body size, and metabolic rate in salamanders. *Physiol. Zool.* 49: 398–406, 1976.
- FEDER, M. E. Oxygen consumption and activity in salamanders: effects of body size and lunglessness. *J. Exp. Zool.* 202: 403–414, 1977.
- FEDER, M. E. Integrating the ecology and physiology of plethodontid salamanders. *Herp.* 39: 291–310, 1983.
- FEDER, M. E. Effect of thermal acclimation on locomotor performance in a lungless salamander, *Desmognathus ochrophaeus*. *J. Exp. Biol.* 121: 271–283, 1985.
- FEDER, M. E., AND W. W. BURGGREN. Cutaneous gas exchange in vertebrates: design, patterns, control, and implications. *Biol. Rev.* 60: 1–45, 1985.
- FEDER, M. E., AND L. E. OLSEN. Behavioral and physiological correlates of recovery from exhaustion in the lungless salamander, *Batrachoseps attenuatus* (Amphibia: Plethodontidae). *J. Comp. Physiol.* 128: 101–107, 1978.
- FULL, R. J., J. A. ASSAD, AND C. F. HERREID. Energetics of the exercising wharf crab, *Sesarma cinereum*. *Physiol. Zool.* 58: 605–615, 1985.
- FULL, R. J., AND C. F. HERREID. The aerobic response to exercise of the fastest land crab. *Am. J. Physiol.* 244 (Regulatory Integrative Comp. Physiol. 13): R530–R536, 1983.
- FULL, R. J., AND C. F. HERREID. Fiddler crab exercise: the energetic cost of running sideways. *J. Exp. Biol.* 109: 141–161, 1984.
- GARLAND, T. Scaling maximal running speed and maximal aerobic speed to body mass in mammals and lizards. *Physiologist* 25: 388, 1982.
- GARLAND, T. The relationship between maximal running speed and maximal aerobic speed in terrestrial mammals. *J. Zool. Lond.* 199: 157–170, 1983.
- GARLAND, T. Physiological correlates of locomotor performance in a lizard: an allometric approach. *Am. J. Physiol.* 247 (Regulatory Integrative Comp. Physiol. 16): R806–R815, 1984.
- GLEESON, T. T. Metabolic recovery from exhaustive exercise by a large lizard. *J. Appl. Physiol.* 48: 689–694, 1980.
- HAGBERG, J. M., J. P. MULLIN, AND F. J. NAGLE. Oxygen consumption during constant load exercise. *J. Appl. Physiol.* 45: 381–384, 1978.
- HERREID, C. F. Energetics of pedestrian arthropods. In: *Locomotion and Energetics in Arthropods*. New York: Plenum, 1981.
- HERREID, C. F., AND R. J. FULL. Cockroaches on a treadmill: aerobic running. *J. Insect Physiol.* 30: 395–403, 1983.
- HERREID, C. F., R. J. FULL, AND D. A. PRAWEL. Energetics of cockroach locomotion. *J. Exp. Biol.* 94: 189–202, 1981.
- HILLMAN, S. S. Cardiovascular correlates of maximal oxygen in anuran amphibians. *J. Comp. Physiol.* 109: 199–207, 1976.
- HILLMAN, S. S., V. H. SHOEMAKER, R. PUTNAM, AND P. C.

- WITHERS. Reassessment of aerobic metabolism in amphibians during activity. *J. Comp. Physiol.* 129: 309-313, 1979.
24. HILLMAN, S. S., AND P. C. WITHERS. An analysis of the respiratory surface area as a limit to activity metabolism in anurans. *Can. J. Zool.* 57: 2100-2105, 1979.
  25. HUEY, R. B., A. F. BENNETT, H. JOHN-ALDER, AND K. A. NAGY. Locomotor capacity and foraging behavior of Kalahari lacertid lizards. *Anim. Behav.* 32: 41-50, 1984.
  26. HUGHES, E. F., S. C. TURNER, AND G. A. BROOKS. Effect of glycogen depletion and pedaling speed on "anaerobic threshold." *J. Appl. Physiol.* 52: 1588-1607, 1982.
  27. JOHN-ALDER, H. B. Seasonal variations in activity, aerobic energetic capacity, and plasma thyroid hormones (T3 and T4) in an iguanid lizard. *J. Comp. Physiol.* 154B: 409-419, 1984.
  28. JOHN-ALDER, H. B., AND A. F. BENNETT. Thermal dependence of endurance and locomotory energetics in a lizard. *Am. J. Physiol.* 241 (Regulatory Integrative Comp. Physiol. 10): R342-R349, 1981.
  29. MARCONI, C., D. PENDERGAST, P. SELYK, D. W. RENNIE, AND P. CERRETELLI. Dynamical and steady-state metabolic changes in running dogs. *Respir. Physiol.* 50: 93-110, 1982.
  30. NAGLE, F., D. ROBINHOLD, E. HOWLEY, J. DANIELS, G. BAPTISTA, AND K. STOEDEFALKE. Lactic acid accumulation during running at submaximal aerobic demands. *Med. Sci. Sports* 2: 182-186, 1970.
  31. PIIPER, J., R. N. GATZ, AND E. C. CRAWFORD. Gas transport characteristics in an exclusively skin-breathing salamander, *Desmognathus fuscus* (Plethodontidae). In: *Respiration of Amphibious Vertebrates*, edited by G. M. Hughes. New York: Academic, 1976, p. 339-356.
  32. SEEHERMAN, H. J., C. R. TAYLOR, G. M. O. MALOIY, AND R. B. ARMSTRONG. Design of the mammalian respiratory system. II. Measuring maximum aerobic capacity. *Respir. Physiol.* 44: 11-24, 1981.
  33. TAIGEN, T. L., AND C. A. BEUCHAT. Anaerobic threshold of anuran amphibians. *Physiol. Zool.* 57: 641-647, 1984.
  34. TAIGEN, T. L., S. B. EMERSON, AND F. H. POUGH. Ecological correlates of anuran exercise physiology. *Oecologia Berlin* 52: 49-56, 1982.
  35. TAIGEN, T. L., AND F. H. POUGH. Metabolic correlates of anuran behavior. *Am. Zool.* 25: 987-997, 1985.
  36. TAYLOR, C. R. Aerobic and anaerobic components of exercise metabolism in mammals (Abstract). *Proc. Int. Union Physiol. Sci.* 15: 277, 1983.
  37. TAYLOR, C. R., N. C. HEGLUND, AND G. M. O. MALOIY. Energetics and mechanics of terrestrial locomotion. I. Metabolic energy consumption as a function of speed and body size in birds and mammals. *J. Exp. Biol.* 97: 1-21, 1982.
  38. TAYLOR, C. R., G. M. O. MALOIY, E. R. WEIBEL, V. A. LANGMAN, J. M. Z. KAMAU, H. J. SEEHERMAN, AND N. C. HEGLUND. Design of the mammalian respiratory system. III. Scaling maximum aerobic capacity to body mass: wild and domestic mammals. *Respir. Physiol.* 44: 25-37, 1981.
  39. TERJUNG, R. L., G. A. DUDLEY, AND R. A. MEYER. Metabolic and circulatory limitations to muscular performance at the organ level. *J. Exp. Biol.* 115: 307-318, 1985.
  40. WASSERMAN, K., B. J. WHIPP, S. N. KOYAL, AND W. L. BEAVER. Anaerobic threshold and respiratory gas exchange during exercise. *J. Appl. Physiol.* 35: 236-243, 1973.
  41. WHIPP, B. J., AND K. WASSERMAN. Oxygen uptake kinetics for various intensities of constant load work. *J. Appl. Physiol.* 33: 351-356, 1972.
  42. WITHERS, P. C. Oxygen consumption of plethodontid salamanders during rest, activity, and recovery. *Copeia* 1980: 781-786, 1980.
  43. WITHERS, P. C., AND S. S. HILLMAN. Oxygen consumption of *Amphiuma means* during forced activity and recovery. *Comp. Biochem. Physiol. A Comp. Physiol.* 69: 141-144, 1981.