Relative shortening velocity in locomotor muscles: turkey ankle extensors operate at low $V/V_{\text{max}}$

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Gabaldón AM, Nelson FE, Roberts TJ. Relative shortening velocity in locomotor muscles: turkey ankle extensors operate at low $V/V_{\text{max}}$. Am J Physiol Regul Integr Comp Physiol 294: R200–R210, 2008. First published October 31, 2007; doi:10.1152/ajpregu.00473.2007.—The force-velocity properties of skeletal muscle have an important influence on locomotor performance. All skeletal muscles produce less force the faster they shorten and typically develop maximal power at velocities of ~30% of maximum shortening velocity ($V_{\text{max}}$). We used direct measurements of muscle mechanical function in two ankle extensor muscles of wild turkeys to test the hypothesis that during level running muscles operate at velocities that favor force rather than power. Sonomicrometer measurements of muscle length, tendon strain-gauge measurements of muscle force, and bipolar electromyograph were taken as animals ran over a range of speeds and inclines. These measurements were integrated with previously measured values of muscle $V_{\text{max}}$ for these muscles to calculate relative shortening velocity ($V/V_{\text{max}}$). At all speeds for level running the $V/V_{\text{max}}$ values of the lateral gastrocnemius and the peroneus longus were low (<0.05), corresponding to the region of the force-velocity relationship where the muscles were capable of producing 90% of peak isometric force but only 35% of peak isotonic power. $V/V_{\text{max}}$ increased in response to the demand for mechanical power with increases in running incline and decreased to negative values to absorb energy during downhill running. Measurements of integrated electromyograph activity indicated that the volume of muscle required to produce a given force increased from level to uphill running. This observation is consistent with the idea that $V/V_{\text{max}}$ is an important determinant of locomotor cost because it affects the volume of muscle that must be recruited to support body weight.

PROPERTIES OF MUSCLE AS A power-producing motor are well characterized, and it is possible to make predictions about how muscle motors might operate most effectively during movement. A. V. Hill did this, in a classic paper, when he suggested that during locomotion muscles should operate over only a subset of their range of possible shortening velocities (19). This prediction was based on the observation that both the mechanical power and the metabolic efficiency of contracting muscle fall off at speeds close to either the muscles’ upper limit ($V_{\text{max}}$), or at very slow speeds. Hill speculated that muscles operate at intermediate shortening velocities to produce power effectively, reducing metabolic cost and maximizing mechanical performance. This hypothesis has since been extended and supported by measurements of muscle shortening velocities in jumping frogs and swimming fish (25, 37). Evidence indicates that these muscles operate near 0.3 $V/V_{\text{max}}$, the $V/V_{\text{max}}$ that elicits maximal power output in isolated muscle. Many components of the musculoskeletal system influence the velocity of contracting muscle fibers, including the arrangement of skeletal lever systems, muscle architecture, and the stretch of elastic tendons. It has been proposed that the relatively narrow range of optimal performance for skeletal muscle has had a pervasive effect on the evolution of musculoskeletal form and function and may effectively serve as a design principle for the musculoskeletal system (38).

In this study, we investigate the $V/V_{\text{max}}$ of muscles that power walking and running. It is reasonable to expect that the shortening velocity of muscles that power walking and running will be an important determinant of metabolic and mechanical performance, just as for swimming and flying. However, the mechanical demands of running are different from those of swimming and flying. During steady-speed running on level ground, cyclical work is done in each step, as mechanical energy is absorbed in the first half of the step and produced in the second half of the step. The demand for net mechanical power is close to zero, however, because the center of mass of the body does not undergo any net change in potential or kinetic energy over the course of a stride, and air resistance is negligible. What muscles must do during running is provide the force required to support the weight of the body. According to Hill’s force-velocity relationship, power is maximal at 0.3 $V/V_{\text{max}}$ but the force developed at this velocity is only about one third of the force developed in an isometric contraction ($V/V_{\text{max}} = 0$). Thus it has been proposed that for running, muscles will operate most economically at the low relative shortening velocities that favor force production rather than at the velocities that are optimal for effective power production (36, 39).

Here we combine measurements of isotonic muscle force-velocity properties with in vivo measurements of muscle strain during running to test three hypotheses. First, we hypothesized that the ankle extensor muscles in wild turkeys (Meleagris gallopavo) operate at a $V/V_{\text{max}}$ close to zero during steady-speed level running. In recent years in vivo shortening strain and velocity have been measured for a handful of running and hopping animals. Some of these studies have measured small strains that suggest a low $V/V_{\text{max}}$, while other studies have measured higher strains and shortening velocities. However, in most cases, a direct test of our hypothesis has not been possible because measurements of muscle $V/V_{\text{max}}$ have not been available. We used turkeys in the present study because the force-velocity properties of turkey ankle extensors are known (31), and turkeys’ calcified tendons allow for direct measurements of muscle force (15, 36). We combined previous measurements of...
force-velocity properties with in vivo measurements of muscle length (from sonomicrometry), force (from strain gauges), and electrical activity [from electromyographs (EMG)] to calculate muscle $V/V_{\text{max}}$ during periods of active force production in the peroneus longus (PL) and lateral gastrocnemius (LG) muscles. Because previous studies indicated low strains in these muscles (15), we predicted that the operating $V/V_{\text{max}}$ would be low as well.

Second, we manipulated the demand for muscle mechanical power by adjusting treadmill incline, to test the hypothesis that muscle $V/V_{\text{max}}$ changes in parallel with the demand for locomotor power. Animals can run on level ground with almost no net mechanical power output, but uphill running requires muscle power to increase the potential energy of the body incrementally with each step. Decline running requires net mechanical energy absorption by muscles to lower and decelerate the body. We predicted that LG and PL muscles operate at higher positive $V/V_{\text{max}}$ values during incline vs. level running, i.e., at higher relative shortening velocities, and at negative $V/V_{\text{max}}$ (lengthening velocities) during decline running. Our prediction that the LG and PL muscles operate over a range of $V/V_{\text{max}}$ for different mechanical tasks is based on our previous studies showing that muscle work output parallels the demand for work on the body during running on different surface slopes (15, 36).

Third, we hypothesized that changes in muscle $V/V_{\text{max}}$ would be associated with an increase in the volume of muscle required to produce force. This hypothesis is based on the idea that the force-velocity relationship will influence the force output of active muscle fibers. As $V/V_{\text{max}}$ increases, the magnitude of force produced by a single muscle fiber decreases (36). A greater number of muscle fibers must therefore be recruited to produce a given whole muscle force. To estimate relative changes in recruited muscle volume, we used rectified integrated EMGs (iEMGs). The idea that $V/V_{\text{max}}$ affects recruited muscle volume is central to our understanding of running energetics, because the volume of muscle that must be active to produce force is likely an important determinant of the metabolic cost of running (4, 23, 35, 39).

**METHODS**

**Animals and treadmill training.** Our animal use protocol was approved by the Oregon State University Institutional Animal Care and Use Committee and in accordance with federal and institutional guidelines. Adult female Eastern wild turkeys (*Meleagris gallopavo*) were obtained from a local breeder and housed in an outdoor enclosure. Food and water were provided ad libitum. Mean body weight was $3.51 \pm 0.39 \text{ kg (SD)}, n = 6$. Treadmill training consisted of running on a level, inclined ($+6$ and $+12$ degrees) and declined ($-6$ and $-12$ degrees) motor-driven treadmill (Keys Pro 2000 Series; Keys Fitness Products, Dallas, TX) for 10–20 min/day, 4 to 5 days a week, for 4–6 wk. Birds ran on each slope on alternate days at speeds of 1–3 m/s. A wooden box with a Plexiglas window for video imaging and an opening at the back for access to the bird was placed around the edges of the treadmill track.

**Surgery.** Animals were induced and maintained on inhaled isoflurane anesthesia, and a sterile environment was maintained for all surgical procedures. A pair of sonomicrometry crystals (Sonometrics, London, ON, Canada) 1 mm or 2 mm diameter in size (we began with the 1-mm size and found that the 2 mm worked better for eliminating level shifts) were implanted into small pockets made with a 16-gauge hypodermic needle within each muscle to a depth of $\pm 3$ mm along the axis of a proximal fascicle. Both the PL and LG are pennate muscles, but proximal fascicles are easily visualized and accessed. Thus, crystals could be accurately aligned along the axis of fascicles. The crystals were positioned 8–12 mm apart and secured in place with a small drop of 3M Vet-bond glue, and the wire leads were sutured to the thin fascia associated with the fascicles (i.e., perimysium) using 6-0 silk suture. Two bipolar, hooked EMG electrodes constructed of silver wire (1 mm offset, with 1 mm of insulation removed from the tips) were implanted within each muscle near the sonomicrometry crystals using a 25-gauge hypodermic needle. The leads were sutured to the muscle’s fascia using 6-0 silk suture. Two small strain gauges (model FLK-1-11; Tokyo Sokki Kenkyujo) were glued to the superficial and deep aspects of the bony tendon of each muscle. The calcified tendons were prepared for gluing by gently scraping and then defatting the surface with chloroform. A thin layer of cyanoacrylate adhesive (Duro superglue, model SUP-5; Loctite, Avon, Ohio) was applied to each strain gauge, and it was pressed onto the tendon for 1 min for bonding. All transducer wires were routed subcutaneously from the muscle to a small skin incision near the middle of the synsacrum. The incision was closed, and small electrical connectors (Microtech, Boothwyn, PA) were secured to the skin with 3-0 silk suture. Animals were allowed to recover from surgery for 24–48 h before treadmill running experiments.

**Running experiments.** Measurements were taken as the birds ran on a level treadmill, followed by runs on an incline ($+6$ and $+12$ degrees) and decline ($-6$ and $-12$ degrees) at speeds of 1–3 m/s. Trials were generally started at 1 m/s and worked up to 3 m/s in 0.5-m/s speed increments. For each run, 10 s of data were collected. Birds remained on the treadmill at slow walking speeds between speed and slope changes, and they were rested when needed. Fascicle lengths were recorded with sonomicrometry at a frequency of 992 Hz using the data acquisition software SonoLAB. Muscle EMG signals were amplified 1,000 times using a DAM50 differential preamplifier (World Precision Instruments, Sarasota, FL) with high- and low-band pass filters of 3 Hz and 10 kHz, respectively. The EMG signals were subsequently filtered in software with a custom-designed FIR filter (band pass, 150–1,000 Hz). Tendon strain signals were amplified with a strain-gauge conditioner (model 2120; Vishay Measurements Group, Raleigh, NC). Data were collected at a frequency of 4,000 Hz to a Macintosh computer with a 12-bit A/D converter (model PCI-MIO-16-1; National Instruments Austin, TX) using the software program IGOR Pro (WaveMetrics, Lake Oswego, OR). High-speed video was recorded at 250 frames/s with an Imaging MotionScope (model 1000S; Redlake, Morgan Hill, CA).

**In situ calibration of muscle force.** Tendon strains were calibrated to muscle force in situ at the end of running experiments. The procedure involved electrically stimulating the muscle via the sciatic nerve while simultaneously measuring whole muscle force and tendon strain. The birds were kept under deep anesthesia with isoflurane gas during the experiments and body temperature was maintained at 37–39°C. The sciatic nerve was isolated and severed at the proximal end and then placed across two silver wires in a nerve cuff. Mineral oil was poured around the nerve, and the skin incision was sutured closed. The muscle origin was fixed in place by means of two bone screws inserted into the femur and attached to an aluminum frame. To calibrate muscle force, the tendon was cut free at its insertion and attached to an aluminum clamp connected to a servomotor (Aurora model 310B-LR; Aurora, ON, Canada) to measure muscle force. The sciatic nerve was stimulated with a Grass S48 stimulator (6–7 V supra-maximal stimulation voltage, 100-Hz frequency, and 250-ms train duration). Signals were averaged for gauges on the superficial and deep aspects of the tendon to account for bending. The slope of the line relating muscle force to average tendon strain was determined by linear regression analysis and was used to calculate muscle force from strain in vivo. More details of this method has been presented previously (15).
Data analysis. All wave analyses were performed using the software program IGOR Pro (WaveMetrics, Lake Oswego, OR). Sonomicroscopy signals were smoothed using the interpolation function (smoothing spline, with a smoothing factor of 1.0 and an SD of 0.01 to 0.025). Fascicle segment length (L), the distance between the two crystals, was differentiated to calculate instantaneous velocity. Fascicle segment length was expressed relative to the resting segment length (L0), which was calculated by averaging the maximum and minimum lengths in swing. To determine muscle power, we first calculated total muscle fascicle velocity by multiplying the fascicle segment velocity by the ratio of total fascicle length to measured segment length. Power was calculated as the product of muscle force and fascicle velocity and is expressed relative to muscle weight (W/kg).

Muscle V/Vmax was calculated from the average fascicle shortening velocity during stance and published Vmax values (31). Vmax measurements were taken from animals obtained from the same source and of the same age and condition as the animals used for the running measurements. Muscle fascicle pennation will tend to increase muscle shortening velocity in relation to muscle fascicle velocity (5); therefore our measurements of whole muscle Vmax are slight overestimates of Vmax at the level of fascicles. iEMG activity was calculated from the area under the rectified EMG signal for each stance period.

For each animal, 10 strides/run were averaged for analysis. We analyzed all values (net work, average force, average velocity, and net fascicle strain) over the period of force production during stance for the LG and PL.

Statistics. To determine the influence of running slope on the measured variables, we used the program Systat (version 7.0 for the personal computer) to perform a two-way mixed-model ANOVA with slope, or slope and individual, as main effects. The F ratio for the main effect of speed or slope was calculated as the mean square divided by the mean square for speed/slope times individual interaction term (41). The criterion for statistical significance was P ≤ 0.05. Summary data are presented as means ± SE.

RESULTS

Effects of locomotor speed and surface slope on stride cycle parameters. Stride cycle parameters for turkeys walking and running at speeds of 1–3 m/s on a level, declined, and inclined treadmill are shown in Fig. 1. This speed range includes both walking and running gaits. A Froude number analysis predicts that turkeys should change from a walking to a running gait at speeds of 1–3 m/s (1, 16), and this range is confirmed by analyses of the mechanical energy changes of the body (9). Across the range of speeds measured here, stride frequency increased significantly with speed, while stride duration decreased (P < 0.05). Stance-phase and swing-phase durations both decreased with speed (P < 0.05), but it was mainly less time spent in stance that resulted in shorter stride when calculated as the product of muscle force and fascicle velocity and is expressed relative to muscle weight (W/kg).

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Fig. 1. Effects of running speed and slope on stride frequency (A), stride duration (B), stance-phase duration (C), swing-phase duration (D), and duty factor (E). Values for level (○), 12-degree incline (●), and 12-degree decline (△) are presented as means ± SE for n = 6 turkeys. There was a significant effect of speed on all measured variables (2-way repeated-measures ANOVA, P < 0.001). All variables were significantly affected by slope (P < 0.014) with the exception of stance duration (C).

Fig. 2. Both muscles generated force primarily during the stance phase. Differences between the two muscles in the timing of force production and in the pattern of muscle length changes have been reported previously (15). All subsequent analyses focus on the period of stance-phase force production, as indicated by the dark lines in Fig. 2.
There was a significant effect of locomotor speed on average power output per step for the LG and PL \((P < 0.05)\). The two muscles increased mechanical power production with increasing speed on the level and incline, and they increased mechanical power absorption with increasing speed on the decline (Fig. 3, A and B). Mechanical power is the product of force and velocity, thus changes in muscle mechanical power output might result from a change in either or both of these variables. For the LG, increases in both average muscle force \((P < 0.05)\) and average muscle velocity \((P < 0.05)\) contributed to the increase in mechanical power with speed. The small changes in PL power with speed could not be resolved in a significant trend for either force or velocity. Average muscle velocity during stance for the PL tended to increase with speed in parallel with mechanical power, but the small changes in PL power with speed could not be resolved in a significant trend for either force or velocity. Average muscle velocity during stance for the PL tended to increase with speed in parallel with mechanical power, but did not reach statistical significance \((P = 0.056)\). Relative EMG activity for the LG increased significantly with speed for all slopes \((P < 0.05)\); relative EMG activity for the PL did not vary significantly with speed \((P = 0.378)\).

There was a significant effect of surface slope on average power output for both the LG and PL \((P < 0.05; \text{Fig. 3})\). The muscles generated significantly higher power outputs during incline vs. level locomotion, and they absorbed mechanical energy (indicated by negative power) during decline locomotion (Fig. 3 A and B). To operate at higher power outputs during incline locomotion, the LG and PL produced force at higher positive (shortening) velocities (Fig. 3, E and F), and to shift mechanical function to power absorption during decline locomotion, both muscles operated at negative (lengthening) velocities. Average muscle force was unchanged at different surface slopes for LG and PL \((P > 0.05; \text{Fig. 3, C and D})\), and thus did not contribute to the slope-related changes in mechanical power output. There was an effect of surface slope on relative EMG activity (rectified, iEMG activity expressed relative to 2 m/s level running) for both the LG and PL \((P < 0.05)\). The muscles exhibited significantly higher relative EMG activities on the incline vs. level across all speeds (Fig. 3, G and H). However, relative EMG activities were not significantly different between level and decline.

**V/V\(_{\text{max}}\) values for LG and PL muscles during locomotion.**

Force and power output in an active muscle fiber are dependent upon \(V/V_{\text{max}}\) as described by the classic Hill force-velocity relationship. The force-velocity-power relationship for the muscles studied here has been determined in situ for isotonic contractions in maximally stimulated muscle (31). The \(V_{\text{max}}\) values calculated were 13.0 ± 1.6 L/s for the LG and 14.8 ± 1.0 L/s for the PL (31). Here we use this whole muscle \(V_{\text{max}}\) values to represent the muscle power output.

Fig. 2. In vivo recordings of muscle force, fascicle length, and electromyograph (EMG) activity for the lateral gastrocnemius (LG) and peroneus longus (PL) from one individual turkey during level running at 2 m/s. Shaded areas indicate the stance phase and unshaded areas indicate swing phase. Force, velocity, power, and EMG activity were analyzed for the period of stance-force production, as illustrated by the thick line segments. For clarity, thick line segments are shown only for force and velocity traces. L, fascicle segment length; L\(_o\), resting segment length; W/kg, muscle weight.
from in situ measurements, and the average shortening velocity measured in vivo, to determine muscle V/V_{max} in vivo.

During stance-phase force production, the PL and LG operated at a relatively small fraction of their maximal whole muscle shortening velocities (Fig. 4). Figure 4 shows the peak force produced by the muscle in vivo as a function of its average V/V_{max} during contraction along with the force-velocity and power-velocity relationships determined in situ. The muscles operated at the highest V/V_{max} during incline running. Decline running was associated with lengthening contractions or negative velocities. The total range of relative shortening velocities over all speeds and inclines, as indicated by the gray-shaded bar in Fig. 4, was a small fraction of the muscles’ total range of velocities.

For all running speeds and slopes, the maximum force produced was well below the force that could be produced at the measured V/V_{max}, according to the force-velocity relationship (Fig. 4). These submaximal forces likely reflect submaximal recruitment.

Due to variable fiber recruitment, the in vivo V/V_{max} is not predictive of the total muscle force developed in a contraction. Muscle V/V_{max} should, however, provide an indication of the capacity of the active fibers to develop force. For example, for a muscle contracting at 0.3 V/V_{max}, the active muscle fibers can produce only about 30% of the force they could develop if the contraction were isometric. To determine the influence of V/V_{max} on relative capacity for force and power production in vivo, we calculated the fraction of maximal force and power output corresponding to the average V/V_{max} determined in vivo for the LG and PL. The results express the potential for power and force output as a fraction of whole muscle maximal power and force output (Fig. 5). Both muscles operated at
V/V\text{max} values where a high percentage of peak isometric force can potentially be developed (Fig. 5). During level running, LG and PL muscles operate at relative shortening velocities where 85–100% of peak isometric force can be developed (Figs. 5, A and B). During 12-degree incline running, the percentages decrease as the muscles produce force at higher positive V/V\text{max} values, but they nevertheless remain high, as the muscles can potentially develop 70–85% of peak isometric force over the range of running speeds measured.

At all inclines and speeds studied, the LG and PL operated at relative shortening velocities that were below the V/V\text{max} where maximal power output occurs. From our previous in situ study, we determined that peak power outputs for the LG and PL muscles occur at V/V\text{max} = 0.31 (31). During level running, when muscle V/V\text{max} values are close to zero (V/V\text{max} < 0.04), the muscles can potentially develop <40% of peak power output. On the 12-degree incline, as the muscles produce force at higher positive V/V\text{max} values, the potential for peak power output approaches 60%. Thus, during incline running both muscles operate over regions of the force-velocity curve that are favorable for producing high force even while producing significant mechanical power.

\textit{Instantaneous force-velocity behavior in vivo.} Muscle velocity in vivo is not constant, but varies throughout the period of force production. To determine whether the force-velocity relationship predicted the behavior of the muscle as velocity varied dynamically throughout the contraction, we plotted in vivo force vs. velocity for the LG and PL (Fig. 6). The instantaneous force-velocity relationships for the PL and LG do not resemble the characteristic rectangular hyperbola of the in situ isotonic force-velocity relationship (Fig. 6). For the LG, the departure from isotonic force-velocity behavior is consistent with an activation/relaxation effect on force (see \textit{DISCUSSION}). The force-velocity relationship of the PL does not resemble the isotonic force-velocity curve over any region of the contraction; there is a stronger positive relationship between force and velocity in the PL than negative. The instantaneous force-velocity behavior of the PL may be affected by both activation/deactivation processes and graded recruitment (see \textit{EMG trace} in Fig. 2, \textit{bottom}).

\textit{Muscle activity and V/V_{max}.} To test the influence of V/V_{max} on the volume of muscle required to produce force, we calculated the ratio of the rectified, iEMG activity to the force impulse in stance (force impulse is the product of average force and duration of force during stance). This iEMG-to-force ratio provides a measure of the volume of fibers active for a given whole muscle force developed.

The iEMG/force was significantly higher during incline running compared with level running for both the LG and PL, but was not significantly different between level and decline running (Fig. 7, A and B). To test the hypothesis that increases in V/V_{max} are associated with an increase in the volume of muscle required to produce force, we compared the iEMG/force with average V/V_{max} during force production (Fig. 7, C and D). These results indicated a significant relationship between iEMG/force and V/V_{max} for the PL. For the LG, this relationship was not significant. However, for both muscles there is a significant relationship between iEMG/force and V/V_{max} if data for only uphill and level running are considered. The lack of a significant relationship for the LG across all conditions results from the lack of a significant difference between iEMG/force and V/V_{max} in a comparison of level and downhill running. The relationship between iEMG/force and V/V_{max} is also not significant for the PL, if only level and downhill running data are considered. Thus, a comparison of level running and uphill running supports our hypothesis that recruited muscle volume increases with an increase in V/V_{max}, but a comparison of level and downhill running does not.

\textbf{DISCUSSION}

\textit{Muscle V/V_{max} during locomotion.} The two turkey ankle extensors studied here operate at low relative shortening velocities (V/V_{max}) during walking and running. During level
running, the V/V\text{max} values for the PL and LG corresponded to regions of the muscles’ force-velocity curve where active fibers develop high forces. The muscles’ capacity for mechanical power production at the V/V\text{max} observed during level running is relatively low. For example, at the average V/V\text{max} measured in the LG for level running at 2.5 m/s, the muscle is capable of producing nearly 90% of its peak isometric force, while at the same velocity the muscle is able to develop <35% of its peak isotonic power (Fig. 5). These observations support the idea that these muscles operate at velocities that favor effective force, not power production from active muscle fibers during level, steady-speed locomotion.

Muscle V/V\text{max} in the LG and PL varies with the demand for net mechanical power. Muscle V/V\text{max} increases with running incline as more power must be developed to increase the body’s potential energy, and it decreases to negative values during decline running, when energy must be absorbed. The increase in V/V\text{max} with incline is associated with a decrease in the force output of active fibers and an increase in their mechanical power output, according to the isotonic force-velocity curve. Our measured V/V\text{max} values suggest that for the running inclines and speeds measured here, the muscle does not reach V/V\text{max} levels that are high enough to elicit maximal power (i.e., 0.31 V/V\text{max}). The power potential of the muscle at these velocities, however, is still substantial. At the average V/V\text{max} values observed in vivo for uphill running, the LG and PL are capable of producing nearly 60% of their peak isotonic power outputs.

Our data for turkey ankle extensors support the idea that the V/V\text{max} of active muscles is an important determinant of locomotor performance. Available measurements of muscle V/V\text{max} for swimming and jumping generally indicate that for these high-power activities, muscles operate at intermediate shortening velocities where power is developed effectively (25, 37). Measurements of muscle V/V\text{max} have not been previously available for walking and running. Muscles that contract isometrically necessarily operate at a low (i.e., 0) V/V\text{max}, and previous observations of isometric or near-isometric force production in turkey gastrocnemius (36) and human triceps surae (14) have been interpreted as evidence of contractions favorable for force economy. Larger strains and velocities have been observed in studies using sonomicrometry to measure muscle fascicle lengths in extensor muscles of a variety of animals, including dogs (8), rats (7, 17), goats (18), horses (20), guinea fowl (10, 28), and ducks (3). Variation in measured strain may be related to architectural features of the various muscles studied. It has been proposed that distal muscles, in particular, may be specialized for near isometric force production by virtue of their short fascicles and long tendons (4, 17). In some cases, larger strains have been interpreted as evidence that muscles are operating at velocities that are favorable for efficient work production rather than eco-

Fig. 5. The potential for force (A and B) and power (C and D) production at each running speed, based on the in vivo V/V\text{max} (E and F) and the value of force and power from the isotonic force-power-velocity relationship. The fraction of force or power that could potentially be developed at a given running speed was calculated by dividing the value on the isotonic curve at the V/V\text{max} observed in vivo by the maximum isotonic force or power. Muscle velocities are from data in Fig. 3, but are expressed as V/V\text{max}, where V\text{max} = 13.0 L/s for the lateral gastrocnemius and V\text{max} = 14.8 L/s for the peroneus longus (see Ref. 31).
A Lateral gastrocnemius (LG)

![Diagram of Lateral gastrocnemius (LG)](image)

B Peroneus longus (PL)

![Diagram of Peroneus longus (PL)](image)

Fig. 6. Instantaneous force-velocity relationship for the LG and PL muscles for 1 individual turkey running at 2 m/s on the level (dotted line), 12-degree incline (solid), and 12-degree decline (dashed). The isotonic force-velocity relationships for each muscle are shown as solid lines. Time, as a fraction of the period of force production, is indicated for the LG during incline running, with I = 10%, 2 = 20%, etc. Force and velocity vary dynamically throughout the contraction for both muscles.

A low V/V_{max} for active muscles has important implications for many aspects of locomotor performance, including maximal running speed. Evidence suggests that top running speed is limited by the maximum force that can be developed during stance phase (40). If this is the case, then top speed would presumably be reduced if muscles operated at 0.3 V/V_{max}, rather than the low V/V_{max} values observed here.

In several muscles studied to date, there is a pronounced lengthen-shorten cycle during stance-phase muscle activity (8, 17, 18, 36). These muscles presumably operate at velocities very favorable for force production during the first half of the cycle (while lengthening) and then operate at less favorable velocities during shortening. It is unclear, given our current understanding of the energetics of dynamic muscle contractions, whether a lengthen-shorten cycle that operates on average at zero velocity is more or less economical than an isometric contraction.

*Interpreting whole muscle V/V_{max} values.* Our estimates of active muscle V/V_{max} and force and power potential in active muscles rely on a calculated V_{max} from maximally activated, whole muscle isotonic contractions. A criticism of this approach is that the contractile behavior of the entire population of muscle fibers, as measured by maximally activated whole muscle contractile measurements, may not be representative of the subpopulation of muscle fibers that are recruited during in vivo measurements. The V_{max} measured in maximally activated muscle represents the unloaded shortening velocity of the fastest fibers in the muscles. If slower fibers are recruited during running, our measurements would tend to underestimate the V/V_{max} of the fibers active in vivo.

These concerns should be considered in any interpretation of our data. However, there is good reason to expect that the trends in V/V_{max} that occur across incline in our data are good representations of the changes in V/V_{max} of active fibers. The LG is likely a relatively homogeneous muscle in fiber type. Fiber-type data from another running bird, the emu, indicate that the LG is composed of only fast (fast oxidative glycolytic and fast glycolytic) fibers (33). Our preliminary analyses of turkey LG (data not shown) confirm this fiber composition. Thus, the range of maximal shortening velocities in the LG is likely less than in more heterogeneous muscles. The observation that iEMG per unit force increases with an increase in the calculated V/V_{max} also supports the idea that our measurements of V/V_{max} reflect the V/V_{max} of active fibers.

The fact that in vivo forces are substantially lower than the force observed at the same velocity in situ is not surprising. The force produced in situ would match the force produced in vivo only if the entire muscle were fully active and maximally recruited in vivo. This discrepancy between the forces produced at a given V/V_{max} in vivo vs. in situ is likely explained by submaximal muscle recruitment in vivo. Length-tension effects on force output, or a limited time available to develop maximal forces, could also contribute to the lower forces in vivo compared with the in situ force-velocity curve.

It is clear from the measurements of instantaneous force-velocity behavior in the LG and PL muscles that the magnitude of force developed instantaneously is influenced by many factors other than the force-velocity relationship. The in vivo muscle force-velocity relationship rarely followed a trajectory that resembled the shape of the muscle’s isotonic force-velocity relationship (Fig. 6). These in vivo force-velocity relationships should resemble the in vitro force-velocity curves only when the following conditions are met: 1) the muscle is not in the

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process of activation or relaxation; 2) the fraction of fibers recruited during the course of the contraction does not vary (i.e., there is no graded recruitment); and 3) the muscle operates over a range of lengths where passive and active length-tension effects have negligible influence on force output. The important influence of activation and deactivation timing on muscle force-velocity behavior in vivo has been previously demonstrated in scallop adductor muscle (27). Scallop adductor muscle undergoing length changes and stimulation patterns observed in vivo operates for only a fraction of its cycle along the isotonic force-velocity curve, primarily due to activation/deactivation processes. The influence of the timing of activation and deactivation in turkey ankle extensors is apparent in the force-velocity curves presented in Fig. 6. The decline in force with time during a period of relatively constant velocity in the LG, for example, that occurs from about 40% of the force period to 100% is likely due to the gradual deactivation of the active motor units. These results highlight that force production in these muscles during running is a short duration event relative to the muscle’s rates of activation and deactivation.

The influence of the dynamic nature of muscle contraction on force output has been investigated previously in studies that combine modeled muscle behavior with empirical measurements of dynamic muscle contractions. In a comprehensive study, Brown et al. (6) demonstrated strong effects of stimulus frequency and length on the force-velocity relationship of contracting cat muscles. This work showed that the force-velocity relationship in vivo is likely more complex and variable than that described by classic isotonic curves. However, a decline in force output with increasing shortening velocity was a consistent feature of muscle behavior for the range of stimulation frequencies and lengths tested (6). Askew and Marsh (2) showed that interaction between muscle shortening/lengthening velocity and activation/deactivation processes, characterized by well-known phenomenon, such as shortening-dependent deactivation, can influence the relationship between force and velocity in contracting mouse muscles. They found that a model in which activation/deactivation, force-length, and force-velocity were considered to act additively to determine instantaneous values of muscle force could reliably predict empirical force measurements. These studies generally support the idea that for dynamic, in vivo contractions, muscle shortening velocity is one of many factors that will determine muscle force output.

Muscle shortening velocity and the energy cost of locomotion. The V/V_max of active fibers likely influences the metabolic energy requirements for running. All runners must develop force to support body weight. Active muscle fiber V/V_max may affect the cost of producing this force in two ways. First, muscles consume ATP at a higher rate when they produce force while shortening, a phenomenon first described by W. O. Fenn and commonly referred to as the “Fenn effect” (13). Second, muscle V/V_max will influence the volume of muscle that must be recruited to develop a given force. The force-velocity relationship dictates that force output will be reduced in active fibers as V/V_max increases. Thus, increases in V/V_max will be associated with an increase in the number of
fibers and volume of active muscle required to produce a given whole muscle force. It is this force-velocity effect on recruited muscle volume that is reflected in the increased iEMG from level to uphill running in the LG and PL. These results suggest that the higher cost of uphill running is explained in part by an increase in recruited muscle volume. The increase in active muscle volume results from the fact that fiber force output is reduced at higher V/V\text{\textsubscript{max}}. To produce the same muscle force, more of these fibers must be recruited.

The force-velocity relationship also indicates that the force developed in an actively lengthening muscle (i.e., at negative velocities) should exceed the force developed during shortening or isometry. Thus, we predicted that the negative V/V\text{\textsubscript{max}} values observed during downhill running would be associated with a decline in rectified iEMG activity relative to level or uphill running. This result would also be consistent with the reduced metabolic energy cost of downhill running. Our results, however, did not reveal a decrease in the rectified iEMG activity from level running to downhill running. It is not clear why this is the case. The mechanical behavior of active muscle during lengthening is more difficult to predict because there is no single fixed relationship between velocity and force (21, 24). The force produced at a given lengthening velocity can vary depending on length and contraction history (21). It must also be remembered when interpreting our EMG data that, as an indicator of active muscle volume, the rectified iEMG signal may be a blunt instrument. Variation in conduction velocity between fiber types, amplitude cancellation in discharges from multiple motor units, and other factors can all affect the EMG and potentially obscure a relationship between active muscle volume and EMG signal (12). It has been suggested that the motor unit activation strategies for eccentric contraction may be different for those of concentric contractions (11, 29), and experimental evidence suggests that eccentric contractions involve different discharge rates and the selective recruitment of fast fiber types (30). Thus, changes in activation strategies from concentric to eccentric contractions may also affect our ability to interpret iEMG signals for the eccentric contractions observed in the present study.

Perspectives and Significance

Most attempts to link the energy cost of locomotion to the energy cost of muscular contraction assume that active muscles generate mechanical power with maximal efficiency. If our results are representative of many of the muscles involved in walking and running, then the energy cost of force production in muscles that operate at low V/V\text{\textsubscript{max}} and low power output is an important component of the energy cost of terrestrial locomotion. The link between muscle mechanical function, metabolic function, and locomotor performance may be best developed from a model of isometric contraction for steady-speed, level locomotion.

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