

RESEARCH ARTICLE

Structured variability of steady-speed locomotion in rats

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ABSTRACT

By examining key locomotor parameters during terrestrial locomotion on a substrate without irregularities, we show that rats frequently accelerate and decelerate between two consecutive steps while maintaining an overall steady speed and that the touchdown order of contralateral limbs significantly influences those speed adjustments. The latter highly correlates with significant adjustments in relative forelimb protraction at touchdown and hindlimb extension at lift-off. We conclude that this remarkable level of variability in limb coordination would clearly be advantageous for the functional flexibility needed during terrestrial locomotion on much more irregular (rough) natural terrain. In addition, its occurrence on a substrate lacking irregularities suggests that much of stable, terrestrial steady-speed locomotion in rats is mechanically controlled.

KEY WORDS: Rat; Locomotion, Stability, Inter-step variation

INTRODUCTION

Terrestrial gaits are rhythmic patterns of footfalls. Because these cyclical movements commonly occur at high frequencies (i.e. during running, it is widely accepted that they cannot be actively controlled only by the nervous system (e.g. Biewener and Daley, 2007; Blickhan et al., 2007; Hooper, 2012). During perturbed locomotion, animals instead rely on passive dynamic mechanisms that include spring-mass mechanics and intrinsic mechanisms (see also Biewener and Daley, 2007). In humans and birds, simple spring-mass mechanics mitigate sudden changes in terrain height (Daley and Biewener, 2006; Geyer et al., 2005; Grimmer et al., 2008; Seyfarth et al., 2002). Limbs act as springs and help the system to return to the locomotor trajectory between steps. Interestingly, bipeds maintain passive spring-mass dynamics when limb contact angle, effective limb length and limb stiffness exhibit particularly limited ranges (Seyfarth et al., 2002; Geyer et al., 2005; Grimmer et al., 2008). In addition, intrinsic mechanisms, including force–length, force–velocity and history-dependent properties as well as postural effects on joint dynamics at the musculoskeletal level help to reduce the control effort and consequently the complexity of the nervous system (see also Biewener and Daley, 2007).

However, this does not suggest that neural control is not required during perturbed locomotion. Indeed, active neural control and passive mechanisms are linked. An animal that predicts a perturbation might change limb posture (e.g. contact angle) via muscle activity (Grimmer et al., 2008). Even with a short delay, reflex feedback may also contribute to further stabilization within a single stance phase (e.g. Hiebert and Pearson, 1999). Importantly, control mechanisms greatly depend on locomotor speed and body size (e.g. Biewener and Daley, 2007; Hooper, 2012). During fast

locomotion such as running, mechanical control mechanisms likely play a predominant role in locomotor stability due to neural transmission delays that could be destabilizing (e.g. Full and Koditschek, 1999; Full et al., 2002; Biewener and Daley, 2007). Walking gaits, which occur at slower speeds, are likely to be more actively than passively controlled. In addition, body size plays a crucial role in terms of neural feedback (Hooper, 2012). Mice, for example, have much shorter times for corrective neural computations than horses (Hooper, 2012).

However, gait mechanics are not perfectly reproduced between steps, leading to some level of variation even under restricted conditions (Wainwright et al., 2008). In particular, small quadrupedal mammals would be expected to match their high level of structural flexibility [flexed three-segmented limbs with low limb stiffness when running (Farley et al., 1993; Fischer et al., 2002)] with high functional variation during locomotion. This is inferred by the fact that quadrupeds have to coordinate their forelimbs and hindlimbs, leading to higher control efforts than in bipeds. We therefore tested the prediction that even during overall steady locomotor speed, small mammals show frequent inter-step speed variations that follow a regular and stereotyped pattern.

RESULTS

Despite a relatively constant overall locomotor speed in each trial, almost two-thirds of all observed step transitions ($N=120$) were subjected to an increase ($N=46$) or decrease ($N=32$) in locomotor speed associated with slight changes of ground reaction forces (GRFs; Fig. 1). Interestingly, kinematic parameters that discriminate for preceding step (s_0) locomotor parameters were exclusively related to lift-off whereas those for the reference step (s_1) emphasized touchdown (Figs 2, 3, Table 1). The discriminant analysis of s_0 locomotor parameters showed that whole-body braking forces at s_0 tended to be higher when rats decelerated between s_0 and s_1 (DF2 in Fig. 2A; G1 versus G3 and G2 versus G4 in Fig. 3E,F). More noteworthy, however, is the finding that footfall order of a given step (s_1) is a consequence of characteristics of the preceding step (s_0), and is thus highly predictable. Rats that displayed small whole-body peak propulsive forces and a more extended hindlimb at s_0 lift-off touched down with the forelimb first in s_1 , regardless of acceleration or deceleration between s_0 and s_1 (DF1 in Fig. 2A; G1 versus G2 and G3 versus G4 in Fig. 3A–C; vice versa for hindlimb touchdown in s_1). Hindlimb extension at s_0 , however, was greater with an increase in speed between s_0 and s_1 (DF2 in Fig. 2A; G1 versus G2 and G3 versus G4 in Fig. 3D; vice versa for decelerating steps). In summary, the hindlimb at s_0 lift-off tended to be relatively more extended if the rats accelerated between s_0 and s_1 and landed on the forelimb first at s_1 (G4 in Fig. 2A; Fig. 3A,D), in contrast to s_0 steps where rats decelerated between s_0 and s_1 , and landed first on the hindlimb at s_1 (G1 in Fig. 2A; Fig. 3A,D; t -test $P=0.0015$).

The discriminant analysis of s_1 locomotor parameters revealed that s_1 has a higher whole-body peak braking force and a less protracted hindlimb when the forelimb of a diagonal couplet touched

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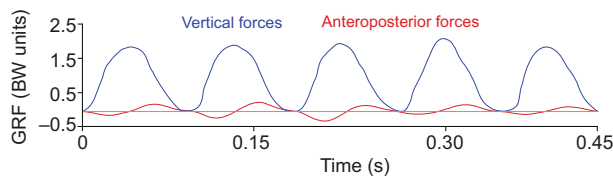


Fig. 1. Representative ground reaction force (GRF) profiles of five consecutive steps of a trotting rat at steady speed ($1.30 \pm 0.02 \text{ m s}^{-1}$) across a terrestrial substrate. Note slight fluctuations in force profile magnitude and shape. BW, body weight.

down first, compared with a hindlimb-initiated trotting step (DF1 in Fig. 2B; Fig. 3G–I, Table 1). Higher whole-body peak braking forces when landing first on the forelimb are likely due to the location of the center of mass (COM), which lies behind the center of pressure of the forelimb at touchdown (Fig. 4). This has a significant influence on inter-step speed adjustments: a rat that landed first on its forelimb was more likely to reduce locomotor speed between s_0 and s_1 (31% for the forelimb versus 19% for the hindlimb). Furthermore, we found a trend towards greater forelimb protraction at s_1 when the rats accelerated between s_0 and s_1 (DF2 in Fig. 2B; Fig. 3J). In order to maintain overall steady-speed locomotion, the rats switched frequently between footfall orders. For example, if s_1 was net propulsive and had a forelimb-initiated diagonal couplet step, then s_0 was likely to be initiated with a hindlimb touchdown regardless of whether s_0 was net braking or net propulsive (Fig. 5).

DISCUSSION

The use of frequent alterations in touchdown order between consecutive steps and the ability to adjust speed at each step shows that rats possess a remarkable level of structured variability in the coordination of their limbs during stable, terrestrial steady-speed locomotion. While such structured variability would clearly be advantageous for the flexibility needed during locomotion on rough terrain, its occurrence on a substrate lacking irregularities suggests that much of stable, terrestrial steady-speed locomotion in rats is mechanically controlled. Indeed, our statistical analyses revealed few parameters that discriminate between footfall orders. Those parameters include relative limb length as well as relative limb protraction and retraction – parameters that play a predominant role in self-stability during perturbed locomotion in bipeds (Geyer et al., 2005; Daley and Biewener, 2006; Seyfarth et al., 2002). We therefore suggest that these parameters play an overall important role during terrestrial locomotion.

Almost two-thirds of all observed net-propulsive steps (s_0) with the hindlimb touching the ground before the forelimb were followed by a net-braking step (s_1) with the forelimb touching the ground before the hindlimb (Fig. 5). This suggests that the frequent alterations in limb coordination and the frequent changes between net-braking and net-propulsive steps characterize the locomotion of small- to medium-sized mammals. Despite moving at an overall constant steady speed, however, landing first on the forelimb does not correlate per se with a net-braking step, and a net-braking step (s_0) per se is not followed by a net-propulsive step (s_1), or vice versa. Occasional deviations thus support the previous assumption that the sensory information transfer (e.g. limb posture and speed) occurs once every step to determine whether to adjust footfall order and/or locomotor parameters (Daley, 2008). The neural control system thus seems to play an additional and significant role in locomotor stability, even on a substrate without irregularities.

By pairing this functional variability with the structural flexibility of three-segmented limbs, small- to medium-sized quadrupedal mammals may have an advantage when navigating uneven substrates, which are frequent features of the landscape for mammals of this size. In addition, mechanical self-stabilizing mechanisms would clearly reduce the complexity of the (neural) control system. Further experiments are needed to investigate the interplay between limb coordination, locomotor stability and substrate characteristics. The high functional variability may also predispose these mammals to locomotor intermittency (relatively short traveled distances between frequent periods of rest) rather than long-distance, steady-speed travel (Eilam, 2004). Hence, morphology and eco-relevant factors may best explain frequent speed adjustments in rats.

MATERIALS AND METHODS

Animals and experimental setup

The Ohio University Institutional Animal Care and Use Committee approved animal care and experimental procedures. Data were obtained from five adult male Fischer 344 X Brown Norway rats [*Rattus norvegicus* (Berkenhout 1769); $421 \pm 29 \text{ g}$]. Rats were caged separately with food and water provided *ad libitum*. Animals were filmed as they moved at their voluntary speed across a force-plate-instrumented terrestrial trackway (length: 2200 mm; width: 150 mm) without irregularities. At least 20 trials per individual were recorded. From these we selected five trials that met the following three criteria. (1) Each trial consists of at least eight consecutive steps. The first and last two steps, respectively, are discarded to account for deceleration and acceleration steps during each trial. Trials for further data analysis consist, therefore, of at least four consecutive steps (mean $N=5$). (2) The gait used in each trial is a running trot (duty factor $< 50\%$; limb phase between 45 and 55%; see Gait determination, below). (3) Animals trot at a steady speed (speed range was

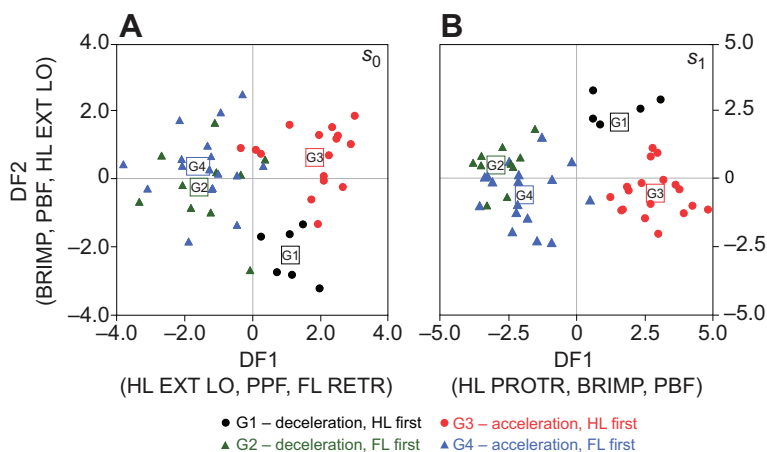


Fig. 2. Results of discriminant function analyses on locomotor parameters of the preceding step (s_0 ; A) and the reference step (s_1 ; B). Groups (G1–G4) used in each analysis are based on touchdown order and inter-step speed adjustments (see Materials and methods). For each discriminant function (DF), the best discriminating variables with structure matrix loadings > 0.3 are listed in parentheses. FL, forelimb; HL, hindlimb; PROTR, protraction; RETR, retraction; EXT, extension; TD, touchdown; LO, lift-off; BRIMP, braking impulse; PBF, peak braking force; PPF, peak propulsive force.

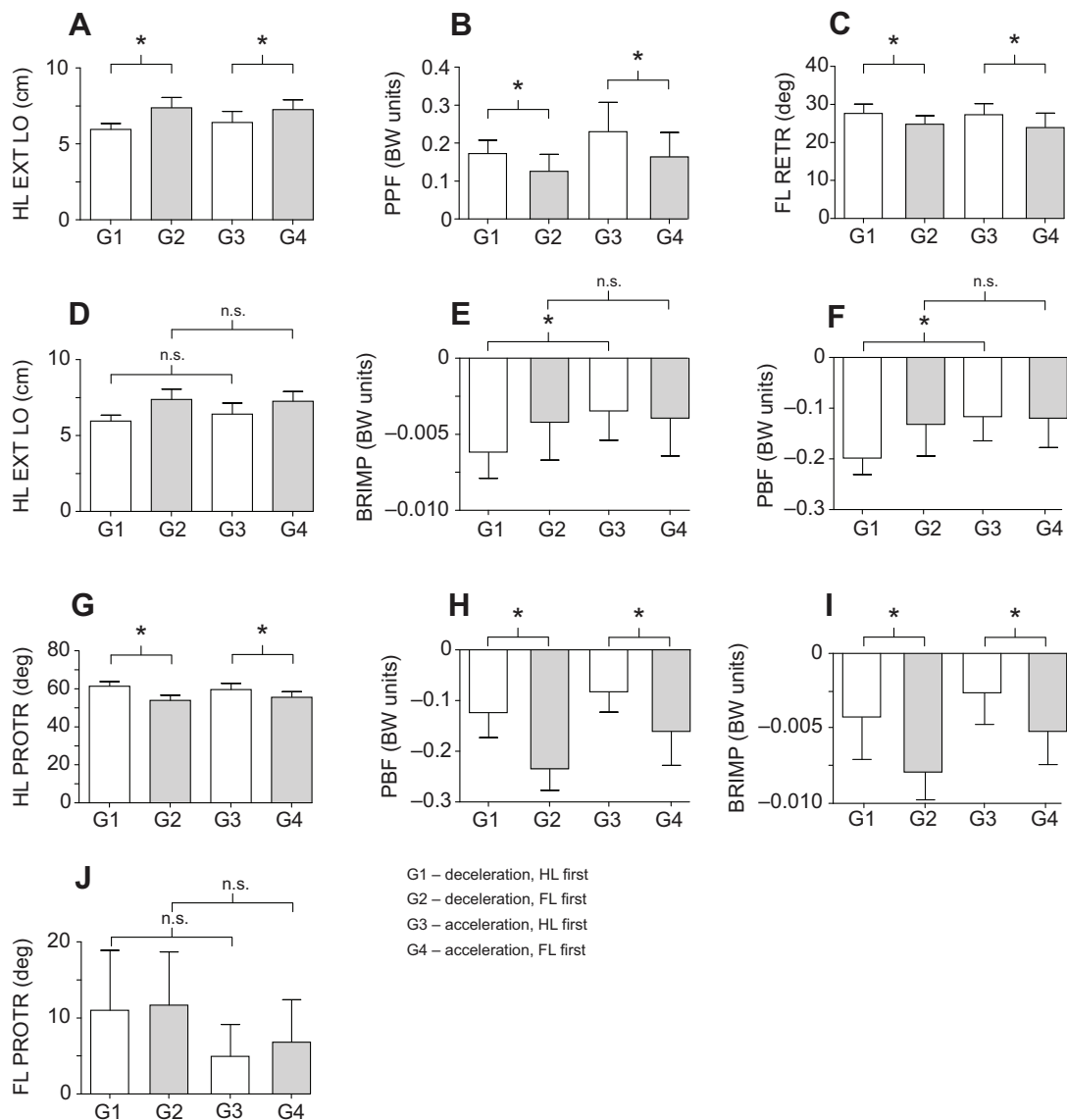


Fig. 3. Results of statistical analyses (t-test) performed on locomotor parameters with discriminant function analyses structure matrix loadings higher than 0.3 (see Fig. 2, Table 1). Asterisks indicate significant differences between groups (e.g. G1 vs G2; DF1 of s_0 ; * $P < 0.05$; n.s., not significant). (A–C) G1 vs G2; G3 vs G4; DF1 of s_0 ; (D–F) G1 vs G3; G2 vs G4; DF2 of s_0 ; (G–I) G1 vs G2; G3 vs G4; DF1 of s_1 ; (J) G1 vs G3; G2 vs G4; DF2 of s_1 . For definitions, see Fig. 2.

limited to $\pm 10\%$ of mean velocity; see Analysis of spatio-temporal gait parameters, below). In total, 150 steps were analyzed (speed range: $0.7\text{--}1.3\text{ m s}^{-1}$). The high variability in touchdown order and speed changes between these 150 steps was the basis of the following categorization. We focused on steps where both contralateral limbs touched the ground in series (this step was designated the reference step, s_1). Steps with synchronous touchdown events were not considered to reduce the complexity of further analyses. The step preceding s_1 is s_0 . Finally, s_1 steps were categorized into four groups (G) based on the nature of the speed change between s_0 and s_1 and which limb touched the ground first in s_1 : G1, speed reduction, hindlimb touchdown first; G2, speed reduction, forelimb touchdown first; G3, speed increase, hindlimb touchdown first; and G4, speed increase, forelimb touchdown first. Our analysis focused on two major events that are likely to be most important for speed adjustments between s_0 and s_1 ; namely, s_0 lift-off and s_1 touchdown.

Analysis of spatio-temporal gait parameters

Metric and kinematic data were obtained with the Qualisys Motion Capture System (QTM, Gothenburg, Sweden). To film the rats across the length of the

trackway, we placed two normal light high-speed cameras (Oqus 310 series, QTM) laterally and in series at a distance of 1.00 m to the direction of movement. Capture frequency was set to 125 Hz. Videos were calibrated and analyzed using TRACKER software v.4.05 (www.opensourcephysics.org). For each trial, ipsilateral limbs facing the camera were analyzed at touchdown and lift-off. The distal tips of the third manual and pedal digits were digitized at touchdown and lift-off to obtain total stance duration (both contralateral limbs). The eye and the tail base were similarly digitized at touchdown and lift-off. Relative limb protraction (at touchdown) and retraction (at lift-off) were then calculated as the positions of the manual and pedal landmarks relative to the positions of the eye and the tail base, respectively (Nyakatura et al., 2008). Positive values in the forelimb indicate a more protracted limb whereas negative values in the hindlimb indicate a more retracted limb. The traveled distance of the eye (digitized at each second frame) was used to calculate mean locomotor speed for the complete trial as well as locomotor speed for each step. The latter was used to calculate changes in locomotor speed (Δv) between s_0 and s_1 . Step transitions were subjected to an increase or decrease in locomotor speed if $\Delta v \geq 0.02\text{ m s}^{-1}$. Raw data were filtered using a fourth-order low-pass Butterworth filter with a cut-off frequency of 25 Hz.

Table 1. Discriminant analysis structure matrices, eigenvalues and percentages of variance explained by each function of the preceding step (s_0) and the reference step (s_1)

	s_0			s_1		
	DF1	DF2	DF3	DF1	DF2	DF3
Eigenvalue	2.666	0.865	0.230	6.483	0.846	0.617
% Variance explained	70.9	23.0	6.1	81.6	10.6	7.8
Cumulative %		93.9	100.0		92.2	100.0
Canonical correlation coefficient	0.853	0.681	0.433	0.931	0.677	0.618
<i>P</i>	0.010	0.588	0.953	<0.0001	0.175	0.309
HL _{PROTR}	0.019	0.222	0.071	0.326	0.259	0.285
HL _{EXT-TD}	0.039	-0.135	0.378	-0.110	0.021	0.110
HL _{RETR}	0.092	-0.114	0.173	-0.152	-0.092	0.083
HL _{EXT-LO}	-0.447	0.365	0.253	0.185	0.047	0.029
FL _{PROTR}	0.065	0.098	0.220	-0.108	0.414	-0.164
FL _{EXT-TD}	-0.201	-0.096	-0.048	0.134	0.239	-0.048
FL _{RETR}	0.309	-0.151	0.146	-0.148	-0.090	0.184
FL _{EXT-LO}	0.069	-0.173	0.282	-0.174	0.227	0.122
BRIMP	0.022	0.409	0.001	0.326	-0.269	0.363
PBF	-0.029	0.541	-0.050	0.408	-0.271	0.411
PRIMP	0.066	0.162	-0.433	-0.030	-0.229	0.190
PPF	0.347	0.219	-0.441	-0.097	-0.185	0.254
VIMP	-0.112	0.055	-0.046	-0.035	0.211	0.046
PVF	0.084	-0.187	-0.008	-0.125	-0.207	0.083
% Recovery	-0.256	0.167	-0.122	0.285	-0.173	-0.297
PS	-0.015	0.210	-0.263	0.198	-0.152	-0.234
SD	-0.037	-0.055	0.008	-0.034	0.267	0.048
DFI	-0.127	-0.203	0.096	0.088	0.150	-0.196

HL, hindlimb; FL, forelimb; PROTR, protraction; EXT, extension; TD, touchdown; RETR, retraction; LO, lift-off; BRIMP, braking impulse; PBF, peak braking force; PRIMP, propulsive impulse; PPF, peak propulsive force; VIMP, vertical impulse; PVF, peak vertical force; PS, phase shift; SD, stance duration; DFI, duty factor index; DF1–DF3: discriminant functions 1–3.

Values in bold indicate structure matrix coefficient loadings greater than 0.30.

Analysis of kinetic locomotor parameters

Three-dimensional whole-body GRFs, comprising anteroposterior, mediolateral and vertical components, were recorded at 1000 Hz using two Bertec force plates (part of a quad-belt-instrumented treadmill system); the 2 mm gap between force plates did not influence locomotor behavior in the rats. Analog signals from the force plates were transferred directly to the Qualisys system via an analog digital board (Qualisys 64 channel A/D board, S/N 8124) and an amplifier (Bertec, AM-6800 signal converter). All data were exported into Excel and filtered using a fourth-order low-pass Butterworth filter with cut-off frequencies between 20 and 50 Hz depending on the force component. Finally, the GRFs were analyzed and normalized to each animal's body weight. Kinetic data included whole-body peak vertical, braking and propulsive force and associated impulses (mediolateral peak forces and impulses were not considered). To estimate the effect of vaulting and bouncing mechanics, we calculated fluctuations in the external mechanical energies and percentage energy recoveries over a step (Cavagna et al., 1977) (see below).

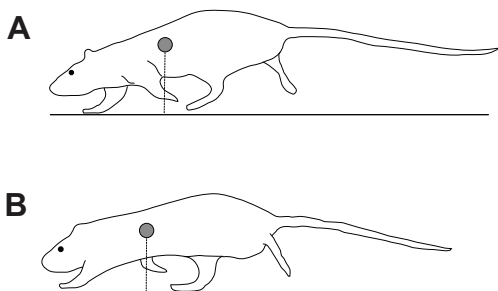


Fig. 4. Drawings of a trotting rat from still images at touchdown. (A) Forelimb first; (B) hindlimb first. Note that the center of mass (COM; gray circle) is located behind and in front of the limbs that touch the ground first and second, respectively (A.S., personal observation).

Analysis of whole-body mechanics

GRFs were exported into a custom-made LabView program (National Instruments, Austin, TX, USA) (Parchman et al., 2003). Accelerations of the COM in all three directions were calculated by dividing out body mass (body weight was first subtracted from the vertical force). Velocities of the COM for each direction were then estimated by taking the first integration of acceleration (integration constant estimated as the mean value for each force record). These velocities were used to calculate kinetic energies ($E_K = \frac{1}{2}mv^2$, where m is body mass in kg and v is velocity in $m s^{-1}$) in the vertical (E_{K-V}), cranio-caudal (E_{K-CC}) and medio-lateral (E_{K-ML}) directions. Summing the three kinetic energies yields the total kinetic energy of the COM (E_{K-TOT}). Changes in the vertical displacement of the COM (h) were determined by integrating vertical velocity (integration constant estimated as the mean vertical record) and were used to determine changes in gravitational potential energy during the step ($E_P = mgh$, where g is gravitational acceleration, $9.81 m s^{-2}$). The sum of E_{K-TOT} and E_P yields the total external mechanical energy (E_{M-TOT}). Finally, we calculated percentage energy recoveries and phase shifts of the fluctuations between E_K and E_P (Cavagna et al., 1977).

Gait determination

Duty factor (S) and limb phase [synonymous to diagonality (Cartmill et al., 2002)] were used to identify running gaits used by the rats (Hildebrand, 1966). Duty factor for one limb refers to the percentage of stance duration of one complete stride cycle whereas limb phase refers to the time from the forelimb touchdown to the touchdown of the ipsilateral hindlimb (in percentage of the hindlimb stride cycle). Forelimb and hindlimb duty factors were used to calculate the duty factor index [$S_{Index} = 100S_{Hindlimb}/S_{Forelimb}$ (Cartmill et al., 2002)].

Statistical analyses

SPSS (v16.0; IBM, Somers, NY, USA) was used for statistical analysis. We performed two separate discriminant function analyses on s_0 and s_1

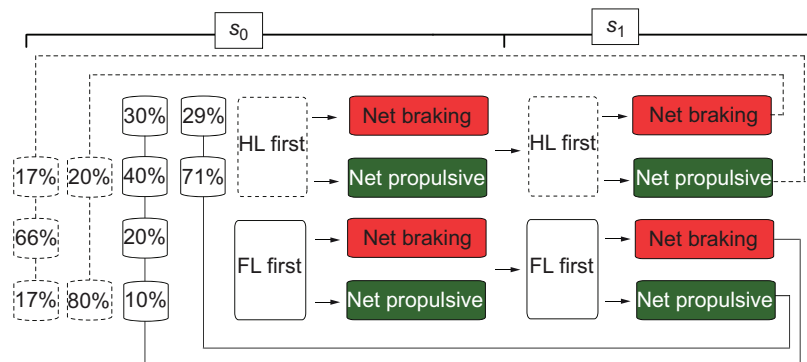


Fig. 5. Proposed model of the relationship between touchdown order, inter-step speed adjustments and net anteroposterior impulses (synchronous touchdowns are not included). For example, if s_1 was net propulsive and had a forelimb touchdown that occurred before the hindlimb touchdown (FL first) then the preceding step s_0 would have been a hindlimb touchdown first (HL first) regardless of whether s_0 was net braking (29%) or net propulsive (71%).

locomotor parameters, including stance duration, duty factor index, relative limb protraction at touchdown, relative limb retraction at lift-off, limb extension at touchdown and lift-off, phase shift, percentage energy recovery, and whole-body peak vertical, braking and propulsive force and associated impulses, to identify the parameters that discriminate between all four groups (G1–G4). Variables that were considered significantly different displayed structure matrix coefficient loadings greater than 0.30. The significance level for all analyses was set at $P < 0.05$.

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Competing interests

The authors declare no competing financial interests.

Author contributions

A.S. and A.R.B. conceived and designed the experiments and prepared the manuscript. A.S. performed the experiments and data analysis.

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References

- Daley, M. (2008). Biomechanics: running over uneven terrain is a no-brainer. *Curr. Biol.* **18**, R1064–R1066.
- Biewener, A. A. and Daley, M. A. (2007). Unsteady locomotion: integrating muscle function with whole body dynamics and neuromuscular control. *J. Exp. Biol.* **210**, 2949–2960.
- Blickhan, R., Seyfarth, A., Geyer, H., Grimmer, S., Wagner, H. and Günther, M. (2007). Intelligence by mechanics. *Philos. Trans. R. Soc. A* **365**, 199–220.
- Cartmill, M., Lemelin, P. and Schmitt, D. (2002). Support polygons and symmetrical gaits in mammals. *Zool. J. Linn. Soc.* **136**, 401–420.
- Cavagna, G. A., Heglund, N. C. and Taylor, R. C. (1977). Mechanical work in terrestrial locomotion: two basic mechanisms for minimizing energy expenditure. *Am. J. Physiol.* **233**, R243–R261.
- Daley, M. A. and Biewener, A. A. (2006). Running over rough terrain reveals limb control for intrinsic stability. *Proc. Natl. Acad. Sci. USA* **103**, 15681–15686.
- Eilam, D. (2004). Locomotor activity in common spiny mice (*Acomys cahirinuse*): the effect of light and environmental complexity. *BMC Ecol.* **4**, 16–26.
- Farley, C. T., Glasheen, J. and McMahon, T. A. (1993). Running springs: speed and animal size. *J. Exp. Biol.* **185**, 71–86.
- Fischer, M. S., Schilling, N., Schmidt, M., Haarhaus, D. and Witte, H. (2002). Basic limb kinematics of small therian mammals. *J. Exp. Biol.* **205**, 1315–1338.
- Full, R. J. and Koditschek, D. E. (1999). Templates and anchors: neuromechanical hypotheses of legged locomotion on land. *J. Exp. Biol.* **202**, 3325–3332.
- Full, R. J., Kubow, T., Schmitt, J., Holmes, P. and Koditschek, D. (2002). Quantifying dynamic stability and maneuverability in legged locomotion. *Integr. Comp. Biol.* **42**, 149–157.
- Geyer, H., Seyfarth, A. and Blickhan, R. (2005). Spring-mass running: simple approximate solution and application to gait stability. *J. Theor. Biol.* **232**, 315–328.
- Grimmer, S., Ernst, M., Günther, M. and Blickhan, R. (2008). Running on uneven ground: leg adjustment to vertical steps and self-stability. *J. Exp. Biol.* **211**, 2989–3000.
- Hiebert, G. W. and Pearson, K. G. (1999). Contribution of sensory feedback to the generation of extensor activity during walking in the decerebrate cat. *J. Neurophysiol.* **81**, 758–770.
- Hildebrand, M. (1966). Analysis of the symmetrical gaits of tetrapods. *Folia Biotheoretica* **5**, 9–22.
- Hooper, S. L. (2012). Body size and the neural control of movement. *Curr. Biol.* **22**, R318–R322.
- Nyakatura, J. A., Fischer, M. S. and Schmidt, M. (2008). Gait parameter adjustments of cotton-top tamarins (*Saguinus oedipus*, Callitrichidae) to locomotion on inclined arboreal substrates. *Am. J. Phys. Anthropol.* **135**, 13–26.
- Parchman, A. J., Reilly, S. M. and Biknevicius, A. R. (2003). Whole-body mechanics and gaits in the gray short-tailed opossum *Monodelphis domestica*: integrating patterns of locomotion in a semi-erect mammal. *J. Exp. Biol.* **206**, 1379–1388.
- Seyfarth, A., Geyer, H., Günther, M. and Blickhan, R. (2002). A movement criterion for running. *J. Biomech.* **35**, 649–655.
- Wainwright, P. C., Mehta, R. S. and Higham, T. E. (2008). Stereotypy, flexibility and coordination: key concepts in behavioral functional morphology. *J. Exp. Biol.* **211**, 3523–3528.