The Journal of Experimental Biology 214, 2544-2559

doi:10.1242/jeb.051086

RESEARCH ARTICLE

The kinematic consequences of locomotion on sloped arboreal substrates in a generalized (*Rattus norvegicus*) and a specialized (*Sciurus vulgaris*) rodent

André Schmidt[1,*] and Martin S. Fischer[2]

[1]Ohio University, College of Osteopathic Medicine, Department of Biomedical Sciences, 120 Life Sciences Building, Athens, OH 45701, USA and [2]Institut für Spezielle Zoologie und Evolutionsbiologie mit Phyletischem Museum, Friedrich-Schiller-Universität Jena, Erbertstraße 1, 07743 Jena, Germany

*Author for correspondence (schmidta@ohio.edu)

Accepted 18 April 2011

SUMMARY

Small mammals must negotiate terrains that consist of numerous substrates that vary in diameter, surface structure, rigidity and orientation. Most studies on mammals have focused on the effects of substrate diameter during horizontal locomotion, especially in small- to medium-sized primates and marsupials. Locomotion across sloped arboreal substrates, however, is poorly understood. Here, in order to determine which locomotor parameters a terrestrial mammal, the rat, and a tree-dwelling mammal, the European red squirrel, modify in response to differences in substrate orientation, three-dimensional kinematics were examined using biplanar videoradiography as the animals moved on 30 and 60 deg inclined branches. Our results revealed that to maintain stability and friction as well as balance during inclined branch locomotion, these species utilize comparable locomotor adjustments despite significant differences in travel speed and gait. Rats and European red squirrels increased limb flexion and retraction in order to bring the center of mass as close as possible to the substrate surface and to achieve maximum propulsion. Additionally, forelimbs were placed more laterally and underneath the branch whereas the hindlimbs were placed approximately on the top of the branch. These locomotor adjustments, which have also been observed in primates and marsupials, are independent of speed, morphological adaptations and limb proportions and thus might be strategies used by early mammals. Our results also suggest that mammals that lack, or have reduced, grasping abilities try to maintain the locomotor mode used during horizontal branch locomotion on inclined branches for as long as possible.

Key words: incline, climbing, 3-D kinematics, X-ray imaging.

INTRODUCTION

Most studies on locomotion of small mammals to date have examined movements on horizontal and flat surfaces such as treadmills (e.g. Gillis and Biewener, 2001; Fischer et al., 2002; Schilling, 2005; Koopmans et al., 2007). In reality, small mammals must negotiate terrains that consist of numerous available substrates such as fallen logs and branches. The latter vary in diameter, surface structure, rigidity and orientation, and are often used as runways because they permit much more rapid locomotion than through heterogeneous terrain, especially in escape situations (Montgomery, 1980; Clarke, 1992; Ladine and Kissel, 1994). In addition, rodents, such as the small harvest mouse (*Micromys minutus*) and the large Japanese field mouse (*Apodemus speciosus*), frequently use small branches at different inclinations for foraging and nest-building activities (Piechocki, 1958; Imaizumi, 1978).

Several authors have focused on the locomotion and behavior of primates and their close relatives because of the assumption that they evolved in a ‘fine-branch’ environment (e.g. Jenkins, 1974; Schmitt and Lemelin, 2002; Gebo, 2004). When primates and marsupials with prehensile extremities move on horizontal branches, they reduce travel speed, stride frequency and substrate reaction forces compared with locomotion on the ground (Schmitt, 1994; Schmitt, 1999; Schmitt and Lemelin, 2002; Schmitt, 2003a; Schmitt, 2003b; Lammers and Biknevicius, 2004; Schmitt and Hanna, 2004; Carlson et al., 2005; Franz et al., 2005; Demes and Carlson, 2009; Carlson and Demes, 2010; Lemelin and Cartmill, 2010). By contrast, rodents, clawed primates and terrestrial marsupials are less capable of grasping small branches [e.g. the common marmoset, *Callithrix jacchus* (Schmitt, 2003b); the gray short-tailed opossum, *Monodelphis domestica* (Lammers and Biknevicius, 2004); and the brown rat *Rattus norvegicus* (Schmidt and Fischer, 2010)]. Instead, the latter group maintains dynamic stability *via* speed, with both limb pairs fulfilling similar functions. Despite these differences in locomotor styles, crouched postures and reductions in vertical oscillations were observed in mammals both with and without prehensile extremities (Schmitt, 1994; Schmitt and Lemelin, 2002; Schmitt, 2003b; Lammers and Biknevicius, 2004; Schmitt and Hanna, 2004; Carlson et al., 2005; Franz et al., 2005; Demes and Carlson, 2009; Schmidt and Fischer, 2010). This may be due to the similar biomechanical constraints to which they are subjected during locomotion on small horizontal branches.

When moving across sloped substrates, the challenge of maintaining stability increases enormously (Cartmill, 1974; Cartmill, 1985; Preuschoft, 2002; Alexander, 2003). On inclined substrates, a shear force component acts against the direction of movement (Preuschoft, 2002). With increasing inclination, the gravitational force is shifted towards the hindlimbs, which carry most of the body weight (Preuschoft, 2002). The higher forces that are required for propulsion, together with the shift in body weight distribution, increase the risk of toppling backwards if an animal is not able to make substrate contact as long and secure as possible. Clawless animals, such as primates, overcome this biomechanical constraint by using their grasping extremities, whereas rodents use their claws to interlock with the substrate or display facultative hallucial opposability and coordinated tail use (Cartmill, 1974; Stevens, 2003; Kivell et al., 2010; Byron et al., 2011).

Studies dealing with the effects of sloped substrates on locomotion in mammals have focused on locomotor behavior in general (Zwahlen, 1975; Youlatos, 1999; Essner, 2007) and specifically on inclined terrestrial substrates [treadmill (Prost and Sussman, 1969; Vilensky et al., 1994); trackway (Carlson-Kuhta et al., 1998; Lammers et al., 2006; Kivell et al., 2010)], kinetics (Lammers, 2007), the cost of transport (e.g. Wickler et al., 2000; Hanna et al., 2008), metabolism (Wunder and Morrison, 1974; Armstrong et al., 1983), rates of oxygen consumption (Snyder and Carello, 2008) and muscle activity (Carlson-Kuhta et al., 1998; Gillis and Biewener, 2001; Nakano, 2002). Little data, however, exist on the subject of metrics and kinematics during locomotion on inclined branches. Most primates and marsupials investigated so far do not reduce travel speed during locomotion on the shallow inclined branches (Stevens, 2003; Lammers, 2007; Nyakatura et al., 2008). Yet, lemurids tend to move more rapidly on inclined branches whereas cheirogaleids tend to move more slowly (Stevens, 2003). The primates investigated so far increase limb flexion (crouched posture) during locomotion on sloped branches in order to bring the center of mass as close as possible to the substrate (Stevens, 2003). During locomotion on inclined branches, the relative touch-down positions of the forelimbs and hindlimbs change considerably. The position of both limb pairs at touch-down and lift-off is more posterior than during locomotion on horizontal branches (Nyakatura et al., 2008). The need to increase stability on inclined branches also results in differences in limb placement. The aye-aye, for example, uses primarily 'full grip' rather than 'full curl' or 'digit-2-curl' hand postures during locomotion on shallow inclines (Krakauer et al., 2002). The forelimbs of the white-handed gibbon and the Japanese macaque are placed underneath the branch whereas the hindlimbs make contact with the top, in order to permit maximum propulsion (Nakano, 2002). Nakano suggested that this shift in the touch-down position of the forelimbs depends on substrate inclination in each species, which gives rise to the assumption of a significant correlation between limb proportion, inclination and the position of the center of mass (Nakano, 2002). Yet, between 0 and 30 deg substrate orientation, forelimbs and hindlimbs in the gray short-tailed opossum are placed in a comparable way (Lammers et al., 2004; Lammers, 2007), and an unmodified body position was also observed in the aye-aye (Krakauer et al., 2002). The influence of limb proportions on locomotor performance is supported by observations regarding differences in limb flexion; elbow and knee joints, for example, are consistently more flexed at touch-down and midstance on sloped branches in primates that possess long limbs (lemurids and lorisids) than in cheirogaleids, which possess short limbs (Stevens, 2003).

The increase in limb flexion and the decrease in limb protraction were also observed in most mammals moving on shallow inclined trackways and treadmills (Prost and Sussman, 1969; Vilensky et al., 1994; Carlson-Kuhta et al., 1998; Gillis and Biewener, 2001; Lammers et al., 2006; Kivell et al., 2010). The gray short-tailed opossum, however, exhibits greater limb protraction angles on 30 deg inclines than during horizontal locomotion (Lammers et al., 2006).

Despite this large amount of data pertaining to movements on inclined substrates, climbing definitions are exclusively theoretical (Cartmill, 1985; Hunt et al., 1996; Preuschoft, 2002). According to these theories, the transition between non-climbing and climbing locomotor modes occurs at approximately 45 deg substrate orientations (Hunt et al., 1996) and is highly correlated to the transmitted substrate reaction forces (Preuschoft, 2002). However, there are no definitions of climbing based on experimental data that include kinematic and metric parameters.

In this paper, we examine metric gait parameters (speed, stance duration) and kinematics [element and three-dimensional (3-D) joint angles] of rats and European red squirrels as they move on slightly (30 deg) and steeply (60 deg) inclined branches. Subjects were selected on the basis of the comparability of their body weight (approximately 315 g) and intralimb and interlimb proportions (Vasquez, 2004; Schmidt, 2005; Schmidt, 2008). Despite these similarities, European red squirrels, which prefer arboreal habitats, display relatively longer forelimbs and hindlimbs than rats. Rats are fairly unspecialized animals that occupy a wide range of diverse habitats. The overall questions were: (1) do clawed mammals show comparable locomotor adjustments during locomotion on shallow inclines to those utilized by mammals possessing prehensile extremities; and (2) do rats and European red squirrels show comparable locomotor adjustments despite different gaits during locomotion on inclined branches? The following hypotheses will be examined:

(1) In both rats and European red squirrels, limb flexion increases significantly in order to bring the center of mass as close as possible to the substrate surface.

(2) Touch-down positions in forelimbs and hindlimbs in both rodents do not differ from those of mammals possessing prehensile extremities. Forelimbs should be placed more laterally on the branch in order to increase stability whereas hindlimbs should be placed on the top of the branch in order to generate maximum propulsion.

(3) Protraction angles should decrease as the substrate inclines because of our expectations of an increase in limb flexion as well as stance duration. Both strongly flexed limbs and longer stance durations should limit the time for the swing phase.

(4) Lateral flexion of the vertebral column in rats increases with an increase in inclination. Lateral spine movements permit the placement of the hindlimbs underneath the body (Schmidt and Fischer, 2010), and should increase on sloped substrates if, as expected, the limbs are more flexed than on the horizontal branch.

(5) The transition between climbing and non-climbing locomotor modes occurs between 30 and 60 deg substrate orientation rather than between 0 and 30 deg.

Finally, we will try to determine kinematic and metric parameters that are helpful in defining the term 'climbing' in more detail.

MATERIALS AND METHODS

Animals and experimental setup

Metrics and kinematics of forelimbs and hindlimbs were obtained from two female rats [*Rattus norvegicus* (Berkenhout 1769)] and one female and one male European red squirrel (*Sciurus vulgaris* Linnaeus 1758), each weighing 315±10 g. The Committee for Animal Research of the Freistaat Thüringen, Germany, approved all procedures. All animals were kept in large cages with a wide range of substrates varying in diameter and orientation. Rats were motivated to move towards a box placed at the end of the branch. European red squirrels moved freely within a Plexiglas® enclosure during the experiments. We used branches with a diameter of 20 mm for rats and 25 mm for European red squirrels, which corresponds to 50% of the animal's body width (Schmidt and Fischer, 2010; Schmidt, 2011). The branches were 200 cm long and covered with cork to enable claw penetration.

Data recordings included metrics and kinematics during locomotion on the horizontal (*S. vulgaris* hindlimbs) as well as on inclined branches (30 and 60 deg; *R. norvegicus* and *S. vulgaris* forelimbs and hindlimbs). In order to test the hypothesis that the transition between non-climbing and climbing locomotor modes occurs between 30 and 60 deg substrate orientation rather than between 0 and 30 deg substrate orientation, previously published metric and kinematic data were added to illustrations and statistical analyses [*R. norvegicus* forelimbs and hindlimbs (Schmidt and Fischer, 2010); *S. vulgaris* forelimbs (Schmidt, 2011)]. Rats preferred symmetrical gaits (walk) but they are also capable of using asymmetrical gaits (Andrada et al., 2010). However, for statistical comparisons between substrate orientations we decided to analyze symmetrical gaits only. Contrary to this, European red squirrels used asymmetrical gaits (0 and 30 deg: gallop; 60 deg: half-bound) regardless of the inclination of the substrate. Thus, trailing and leading forelimbs and hindlimbs were analyzed separately because of their time-delayed touch-down events. As European red squirrels placed their hindlimbs synchronously during locomotion on the steepest incline (60 deg), we merged the values in our analyses. In total, 336 strides (rats: 115; European red squirrels: 221) were analyzed (Table 1).

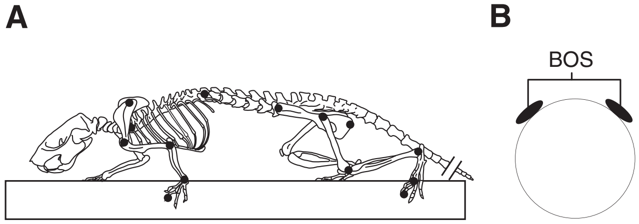

Fig. 1. (A) Illustration of the digitized skeletal landmarks (black dots) of the forelimbs, the hindlimbs and the pelvis (from Schmidt and Fischer, 2010). The proximal pivot of the forelimbs corresponds to the instantaneous center of scapular rotation (proximal end of the scapular spine). (B) Anterior view of the touch-down positions of two limbs (black circles) on a branch. The base of support (BOS) is defined as the distance between the centers of pressure (black circles) of both limbs.

X-ray motion recordings

Motion recordings and data analysis were carried out as described previously (Schmidt and Fischer, 2010) and are briefly summarized below. Movements were synchronously recorded at 1000 Hz using two normal light high-speed cameras (SpeedCam MiniVis e2 ECO, Weinberger, Karlsruhe, Baden-Württemberg, Germany) and a biplanar X-ray system (Neurostar, Siemens, Munich, Bavaria, Germany). Non-pulsed X-ray shots were applied (rats: 40 kV, 53 mA; European red squirrels: 60 kV, 55 mA). An internal 500 μs shutter was used to increase image sharpness.

X-ray motion analysis

The following skeletal landmarks were captured interactively in every tenth X-ray frame per plane using SimiMotion 3D (v7.5, Simi Reality Motion Systems GmbH, Unterschleissheim, Germany): proximal pivot of the forelimb, shoulder joint, elbow joint, wrist joint, distal tip of the third manual digit, hip joint (acetabulum), knee joint, ankle joint, metatarsophalangeal joint and distal tip of the third pedal digit (Fig. 1A) (Schmidt and Fischer, 2010). The proximal pivot of the forelimb is assumed to be the instantaneous center of scapular rotation and can be estimated to be the proximal end of the scapular spine (Fig. 1A) (Jenkins and Weijs, 1979; Fischer, 1994). The thoracic cage was determined by connecting the skeletal landmarks positioned in the center of thoracic vertebra 1 and 13. The position of the pelvis was determined by connecting two skeletal landmarks on the pelvis – the middle of the crista iliaca and the middle of the tuber ischiadicum. To test for accuracy of landmark identification, one randomly chosen sequence was independently digitized 10 times. Digitizing error was less than 1.5 deg for all element angles and joint angles in both species. The variations in the identification of the exact touch-down and lift-off events in X-ray recordings (1000 Hz) were five frames (*S. vulgaris*) and 10 frames (*R. norvegicus*), respectively.

After 3-D calibration in SimiMotion 3D, the single plane coordinates were spline interpolated and subsequently transformed into 3-D coordinates. Following Wu and Cavanagh (Wu and Cavanagh, 1995), the x-axis corresponds to the direction of movement. The y-axis was perpendicular to the substrate surface and the z-axis was perpendicular to the x–y plane. In order to verify the accuracy of the 3-D calibration, the calibration object was reconstructed in SimiMotion 3D and calculated distances between 3-D coordinates of points on the calibration object were compared (position error of max. 1 mm).

Three-dimensional joint angles were defined anatomically with one center of rotation and correspond to the flexor side of each joint investigated (Fig. 1A). They are always the smallest angles between two elements and can range between 0 and 180 deg. The wrist joint, however, was measured on the dorsal side of the joint (dorsiflexion).

Table 1. Speed and stance duration of forelimbs (FL) and hindlimbs (HL) of *Rattus norvegicus* and *Sciurus vulgaris* at different substrate orientations

	0 deg	30 deg	60 deg
Rattus norvegicus			
Speed (m s^{-1})	0.74±0.07 (25)[a]	0.56±0.08 (38)	0.26±0.04 (52)
Stance duration FL (s)	0.13±0.02 (11)[a]	0.16±0.02 (19)	0.32±0.07 (26)
Stance duration HL (s)	0.13±0.02 (14)[a]	0.19±0.03 (19)	0.36±0.08 (26)
Sciurus vulgaris			
Speed (m s^{-1})	1.46±0.27 (38)[b]	1.68±0.36 (33)	1.71±0.52 (45)
Trailing limb			
Stance duration FL (s)	0.08±0.02 (16)[b]	0.07±0.02 (18)	0.07±0.03 (17)
Stance duration HL (s)	0.08±0.02 (22)	0.09±0.02 (14)	0.08±0.02 (28)
Leading limb			
Stance duration FL (s)	0.07±0.02 (14)[b]	0.06±0.02 (19)	0.06±0.03 (17)
Stance duration HL (s)	0.09±0.01 (14)	0.09±0.02 (14)	0.08±0.02 (28)

Data are means ± s.d. The numbers of analyzed strides are given in parentheses.

[a] Data were published previously (Schmidt and Fischer, 2010).

[b] Data were published previously (Schmidt, 2011).

Two-dimensional (2-D) element angles were measured in relation to the substrate surface and calculated using *x*- and *y*-coordinates (lateral view). Lateral rotations of the thoracic cage and the pelvis during stance phase were quantified by calculating the 2-D angle (*x*- and *z*-coordinates, dorsal view) formed by the substrate and the thoracic cage or the pelvis, respectively (Jenkins and Camazine, 1977; Schmidt and Fischer, 2010). In order to eliminate possible effects of the diameter of support on the angle formed by the substrate surface and the thoracic cage or the pelvis, we divided these angles by the base of support (BOS). The BOS is defined as the distance between limb pairs perpendicular to the trajectory of movement (Koopmans et al., 2007). Given that the right and left limbs were placed in a comparable way, the BOS is the distance (*z*-coordinate) between the centers of pressure (COP) of both forelimbs and hindlimbs (Fig. 1B). In order to calculate the COP, we used the *y*- and *z*-coordinates of the wrist joint and the metatarsophalangeal joint at touch-down.

Protraction and retraction angles (αPA, αRA) were examined at touch-down and lift-off (Schmidt, 2008; Schmidt and Fischer, 2010). Both αPA and αRA were calculated by measuring the angle formed by a line perpendicular to the pole at the COP and a line connecting the proximal pivot (scapula, hip joint) with the COP.

At touch-down, midstance and lift-off we calculated limb position (αLP) in the transverse plane using the *y*- and *z*-coordinates of the wrist and the shoulder joint (forelimb) and the metatarsophalangeal and the hip joint (hindlimb). Values below zero indicate an abducted limb (the distal joints lie laterally to the proximal ones); values above zero indicate an adducted limb (the distal joints lie medially to the proximal ones).

Metric parameters include stance duration and mean velocity. To determine an animal's mean velocity, the diaphragm was digitized and plotted against time.

Statistical analyses

Data are expressed as means ± s.d. All variables were tested for individual differences using a two-way mixed-model ANOVA with individuals as random factor and orientation as fixed factor (SPSS v18.0; IBM, Somers, NY, USA). In addition, all variables were tested for speed correlation using GraphPad Prism 4.0c for Macintosh (La Jolla, CA, USA). Because speed effects were much more profound than individual effects, we decided to test against the potential effects of different substrate orientation by using an ANOVA if variables were independent of speed. If one or more significant correlations with speed was observed, heterogeneity was tested using SMATR (Version 2.0) (Warton and Weber, 2002; Falster et al., 2003). We first tested for significant differences in slopes. If slopes did not differ between substrate orientations we then tested for significant shifts in elevation and for significant shifts along common slopes. *Post hoc* tests for all variables were used to compare substrate orientations (0 *vs* 30 deg, 0 *vs* 60 deg and 30 *vs* 60 deg). The significance level was $P<0.05$.

RESULTS

Metric gait parameters

In rats, mean velocities decreased significantly as substrate inclination increased (Fig. 2; Tables 1, 2). By contrast, contact times of forelimbs and hindlimbs increased with increasing substrate inclination (Tables 1, 2). Forelimbs and hindlimbs displayed similar contact times during locomotion, regardless of orientation (ANOVA, $P>0.05$). Interestingly, standard deviations of forelimbs and hindlimbs both decreased (speed) and increased (stance duration) with increasing inclination (Fig. 2, Table 1).

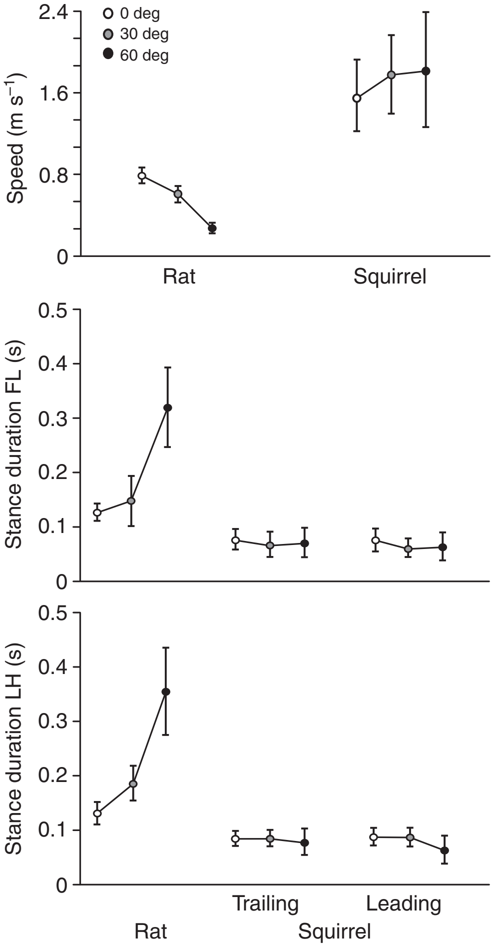

Fig. 2. Mean (±s.d.) values of speed and stance duration in forelimbs (FL) and hindlimbs (HL) in the brown rat (*Rattus norvegicus*) and the European red squirrel (*Sciurus vulgaris*) at three substrate orientations (0, 30 and 60 deg).

European red squirrels, however, increased travel speed by approximately 15% as substrate inclination increased (Fig. 2; Table 1). Contact times in the hindlimbs decreased significantly during locomotion on the steepest incline (*post hoc* test, Table 2). As in rats, contact times in forelimbs and hindlimbs were similar across the range of substrate inclination (ANOVA, $P>0.05$). However, the standard deviation of speed was twice as high during locomotion on the horizontal branch than during locomotion on the steepest incline (Fig. 2; Table 1).

Kinematics

Limb placement

Rats gripped the substrate with their forelimbs significantly more laterally as substrate inclination increased (Fig. 3; Table 3). In both forelimbs and hindlimbs, touch-down positions and thus the BOS did not differ between 0 and 30 deg substrate orientation (Table 3). The same was observed in European red squirrels (Fig. 3; Table 3): touch-down positions of the trailing and leading hindlimbs in mediolateral direction (*z*-coordinates) were not affected by substrate orientation (Table 3).

Element and joint angles

In rats, several element and joint angles showed speed dependency at every substrate orientation, especially in the forelimbs (Table 4).

Table 2. Results of ANOVAs performed on metric parameters of the forelimbs (FL) and hindlimbs (HL) of *Rattus norvegicus* and *Sciurus vulgaris*

		P			
	F	0 *vs* 30 deg	0 *vs* 60 deg	30 *vs* 60 deg	Speed
Rattus norvegicus					
Speed (m s^{-1})[a]	277.9***	<0.001	<0.001	<0.001	
Stance duration FL (s)	21.1***	n.s.	<0.001	<0.001	s[c]
Stance duration HL (s)	30.3***	n.s.	<0.01	<0.001	s[c]
Sciurus vulgaris					
Trailing limb					
Speed FL (m s^{-1})[a]	0.9	<0.05	<0.01	n.s.	
Speed HL (m s^{-1})[a]	0.7	n.s.	<0.05	n.s.	
Stance duration FL (s)	0.5	n.s.	n.s.	n.s.	s[b]
Stance duration HL (s)	21.4***	<0.001	n.s.	<0.001	s[c]
Leading limb					
Speed FL (m s^{-1})[a]	2.1	n.s.	n.s.	n.s.	
Speed HL (m s^{-1})[a]	0.9	<0.05	<0.05	n.s.	
Stance duration FL (s)[a]	0.3	n.s.	n.s.	n.s.	s[b]
Stance duration HL (s)[a]	8.9*	n.s.	n.s.	<0.01	s[c]

Bonferroni *post hoc* tests (*P*-values) were used to compare substrate orientations (0, 30 and 60 deg). *P<0.05; ***P<0.001.

n.s., not significant; s, correlation with speed, without significant differences in slopes, shifts in elevations or shifts along common slopes between substrate orientations.

[a] Significant differences between individuals.

[b] No significant differences in slope, shift in elevation and shift along common slope.

[c] Significant shift in elevation.

At touch-down, forelimb flexion increased significantly with increasing substrate inclination (Fig. 4; Table 4). However, the wrist joint achieved similar touch-down angles across all substrate orientations. Shortly after touch-down, the angular excursions of the shoulder and wrist joints differed significantly between locomotion at 0 and 30 deg and locomotion at 60 deg; the shoulder and wrist joints were much more extended at midstance and lift-off (Fig. 4). Angular excursions of the elbow joint, in contrast, were relatively uniform across all slopes (Fig. 4). With the exception of the upper arm (at lift-off, Table 4), the positions of the elements of the forelimb were significantly affected by the incline, in particular at 60 deg (Fig. 5). As described for the wrist joint, the touch-down positions of the upper arm, the lower arm and the hand did not differ between locomotion on the horizontal branch and locomotion on the 30 deg incline (Fig. 5; Table 4). Elements that are in matched motion (scapula and lower arm) were placed more perpendicular to the substrate the more the inclination increased, whereas the upper arm was positioned more and more horizontally (Fig. 5). There was no difference between upper arm and hand positions at 0 and 30 deg substrate orientation throughout the complete stance phase (Fig. 5; Table 4).

Like the forelimbs, hindlimb joint angles in rats were significantly affected by the incline (Table 4). With increasing substrate orientation, the knee and metatarsophalangeal joints were much more

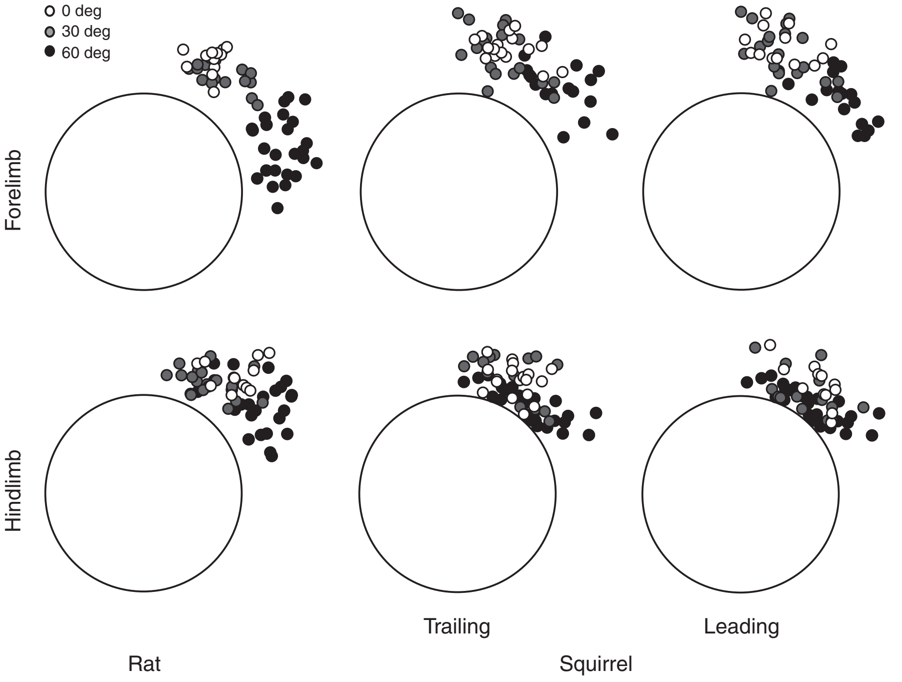

Fig. 3. Anterior view of the touch-down positions of the wrist joint and the metatarsophalangeal joint relative to the substrate orientation (N=336). In rats, the BOS of the forelimbs increased from 13±4 mm (mean±s.d.) on the horizontal branch to 16±6 and 29±4 mm on both inclines. The BOS of the hindlimbs increased from 17±4 mm (0 deg) and 13±5 mm (30 deg) to 23±5 mm (60 deg). In European red squirrels, the BOS of the trailing forelimb was 13±6 mm (0 deg), 12±8 mm (30 deg) and 26±7 mm (60 deg) and that of the leading forelimb was 11±6 mm (0 deg), 11±7 mm (30 deg) and 25±7 mm (60 deg). In the trailing hindlimb, the BOS ranged from 14±5 mm (0 deg) to 14±8 mm (30 deg) and 18±8 mm (60 deg). In the leading hindlimb, the BOS was 20±6 mm on the horizontal branch, 16±8 mm on the shallow incline and 18±8 mm at 60 deg incline.

Table 3. Results of ANOVAs performed on *y*- and *z*-coordinates of the forelimbs (FL) and hindlimbs (HL) of *Rattus norvegicus* and *Sciurus vulgaris*

		P			
	F	0 *vs* 30 deg	0 *vs* 60 deg	30 *vs* 60 deg	Speed
Rattus norvegicus					
z-coordinate FL	36.8*	n.s.	<0.001	<0.001	
y-coordinate FL	68.1*	n.s.	<0.001	<0.001	
z-coordinate HL	6.8*	n.s.	n.s.	<0.05	s[c]
y-coordinate HL	11.0	n.s.	<0.01	<0.05	
Sciurus vulgaris					
Trailing limb					
z-coordinate FL	198.2**	n.s.	<0.001	<0.001	
y-coordinate FL[a]	15.9***	<0.05	<0.001	<0.05	s[b]
z-coordinate HL	5.7	n.s.	n.s.	n.s.	
y-coordinate HL	45.3*	n.s.	<0.001	<0.001	
Leading limb					
z-coordinate FL	152.5**	n.s.	<0.001	<0.001	
y-coordinate FL	34.7***	n.s.	<0.001	<0.001	s[b,c]
z-coordinate HL	0.8	n.s.	n.s.	n.s.	
y-coordinate HL	1.9	n.s.	<0.01	n.s.	

Bonferroni *post hoc* tests (*P*-values) were used to compare substrate orientations (0, 30 and 60 deg). *$P<0.05$; **$P<0.01$; ***$P<0.001$.

n.s., not significant; s, correlation with speed, without significant differences in slopes, shifts in elevations or shifts along common slopes between substrate orientations.

[a] Significant differences between individuals.

[b] Significant shift in elevation.

[c] Significant shift along common slope.

flexed at touch-down (Fig. 6). Touch-down positions of the hip and ankle joints were uniform between 0 and 30 deg substrate orientation (Table 4). With increasing substrate orientation, we observed greater joint extension in the hip joint at midstance and in the knee and hip joints at lift-off (Fig. 6; Table 4). In terms of hindlimb element angles, here too all touch-down positions were fairly uniform at 0 and 30 deg substrate orientation (Fig. 7; Table 4). At touch-down, the thigh was held parallel to the substrate surface regardless of substrate orientation (Fig. 7; Table 4).

Interestingly, at 30 deg inclination we found an intermediate 'position' in the 3-D angular and element excursions of both forelimbs and hindlimbs of rats (Figs 4–7). In several cases (e.g. lower arm, hip joint and metatarsus; Figs 4–7), mean values at touch-down were identical to those observed during locomotion on the horizontal branch and switched to those of the steepest incline at lift-off. This is supported by statistical analyses (Table 4). The touch-down angles of the hip joint, for example, did not differ between 0 and 30 deg or between 30 and 60 deg substrate inclination at lift-off.

In European red squirrels, speed dependency was observed in 21 (trailing limbs) and 18 (leading limbs) of 45 events (e.g. touch-down

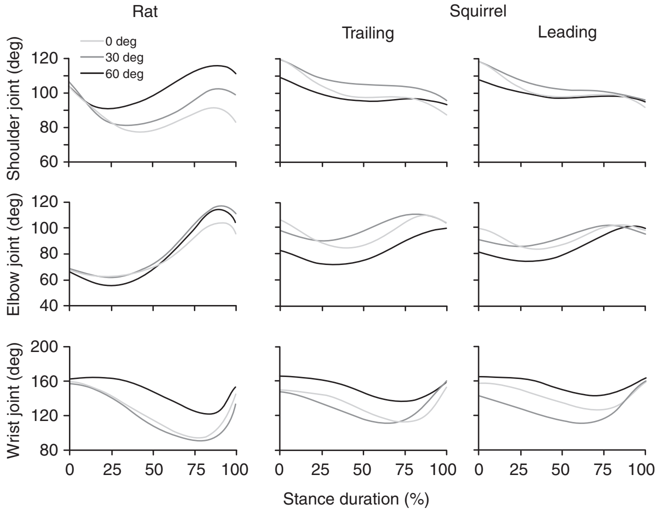

Fig. 4. Mean values of three-dimensional (3-D) joint angular excursions during the stance phase of the forelimbs during locomotion at different substrate orientations in the rat and the European red squirrel.

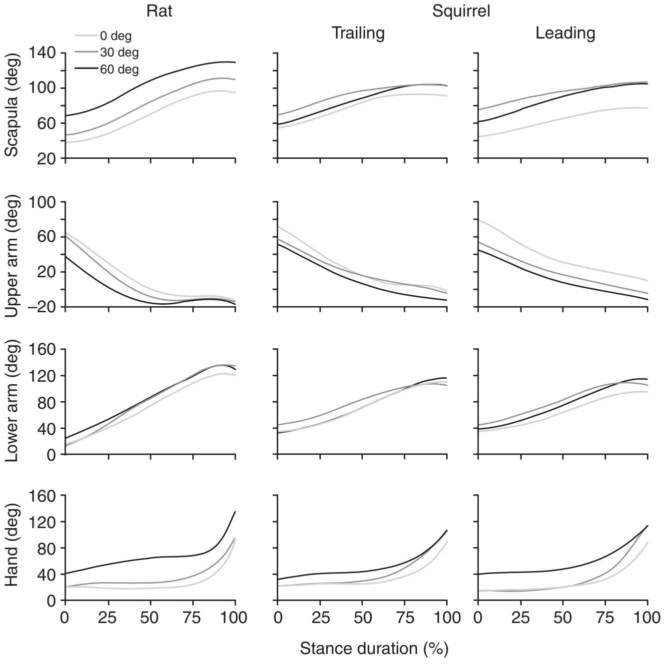

Fig. 5. Mean values of element angular excursions during the stance phase of the forelimbs during locomotion at different substrate orientations in the rat and the European red squirrel.

Table 4. Results of ANOVAs performed on three-dimensional joint angles and element angles of the forelimbs and hindlimbs of *Rattus norvegicus*

	Event	*F*	*P*			Speed
			0 *vs* 30 deg	0 *vs* 60 deg	30 *vs* 60 deg	
Shoulder joint	TD	64.6***	<0.001	<0.001	<0.001	s[b,c]
	MS	22.0*	<0.05	<0.001	<0.001	
	LO	7.5***	<0.001	<0.001	<0.001	s[b,c]
Elbow joint	TD	41.3***	0.01	<0.001	<0.001	s[b,c]
	MS	1.8	n.s.	n.s.	n.s.	
	LO	4.5	n.s.	n.s.	n.s.	
Wrist joint	TD	0.9	n.s.	n.s.	n.s.	
	MS[a]	193.5**	<0.001	<0.001	n.s.	
	LO	19.4*	n.s.	n.s.	<0.05	
Scapula	TD	121.2**	<0.05	<0.001	<0.001	
	MS	57.0*	<0.001	<0.001	<0.001	
	LO	158.4***	<0.001	<0.001	<0.001	s[b]
Upper arm	TD[a]	39.2*	n.s.	<0.001	<0.001	
	MS	30.7***	n.s.	<0.001	<0.001	s[b]
	LO	0.5	n.s.	n.s.	n.s.	
Lower arm	TD	78.7*	n.s.	<0.001	<0.001	
	MS	61.5*	<0.001	<0.001	n.s.	
	LO	265.9**	<0.001	<0.01	n.s.	
Hand	TD	35.1*	n.s.	<0.001	<0.001	
	MS	75.2*	n.s.	<0.001	<0.001	
	LO	30.6*	n.s.	<0.001	<0.001	
Hip joint	TD	13.3	n.s.	<0.001	<0.001	
	MS	82.6*	<0.001	<0.001	<0.001	
	LO	5.4	<0.01	<0.05	n.s.	
Knee joint	TD	134.7**	<0.001	<0.001	<0.001	
	MS[a]	1.5	n.s.	n.s.	n.s.	
	LO	93.7***	<0.001	<0.001	<0.001	s[b,c]
Ankle joint	TD	8.8	n.s.	<0.01	<0.01	
	MS	0.4	n.s.	n.s.	n.s.	
	LO	6.5	n.s.	<0.01	n.s.	
Metatarsophalangeal joint	TD	66.9***	<0.001	<0.001	<0.001	s[b,c]
	MS	4.5	<0.05	n.s.	n.s.	
	LO	1.3	n.s.	n.s.	n.s.	
Pelvis	TD	2.1	n.s.	<0.01	<0.05	
	MS[a]	14.6	n.s.	<0.05	n.s.	
	LO	49.3*	<0.05	<0.001	<0.001	
Thigh	TD	0.1	n.s.	n.s.	n.s.	
	MS	86.7*	<0.001	<0.001	<0.001	
	LO	56.7*	<0.001	<0.001	n.s.	
Lower leg	TD	12.4	n.s.	<0.001	<0.001	
	MS	217.9**	<0.001	<0.001	<0.01	
	LO	1.8	<0.05	n.s.	<0.05	
Metatarsus	TD	8.0	n.s.	<0.01	<0.001	
	MS	77.2*	<0.001	<0.001	<0.001	
	LO	38.8*	<0.001	<0.001	n.s.	

Bonferroni *post hoc* tests (*P*-values) were used to compare substrate orientations (0, 30 and 60 deg). *$P<0.05$; **$P<0.01$; ***$P<0.001$.

n.s., not significant; s, correlation with speed, without significant differences in slopes, shifts in elevations or shifts along common slopes between substrate orientations.

[a] Significant differences between individuals.

[b] Significant shift in elevation.

[c] Significant shift along common slope.

LO, lift-off; MS, midstance; TD, touch-down.

position of the shoulder joint; Tables 5, 6). As described for rats, European red squirrels flexed their forelimbs more as substrate incline increased (Fig. 4), whereas several touch-down positions of 3-D joints and elements in both trailing and leading forelimbs did not differ between 0 and 30 deg substrate orientation (Figs 4, 5; Tables 5, 6). However, in both trailing and leading forelimbs, the 3-D touch-down angles of the wrist joints differed significantly according to substrate orientation, in particular on the 60 deg inclines (Tables 5, 6). At lift-off, the degree of extension of the elbow and wrist joints in the trailing forelimb and of all joints in the leading forelimb was similar across all supports (Fig. 4; Tables 5, 6). The positions of almost all element angles in both trailing and leading forelimbs were significantly affected by inclination (Fig. 5; Tables 5, 6). The position of the hand at touch-down and the lower arm at lift-off was constant, regardless of the status of the limb and the substrate orientation (Fig. 5; Tables 5, 6).

The hindlimbs were more flexed the more the substrate inclined (Fig. 6; Tables 5, 6). *Post hoc* tests revealed that leading hindlimbs

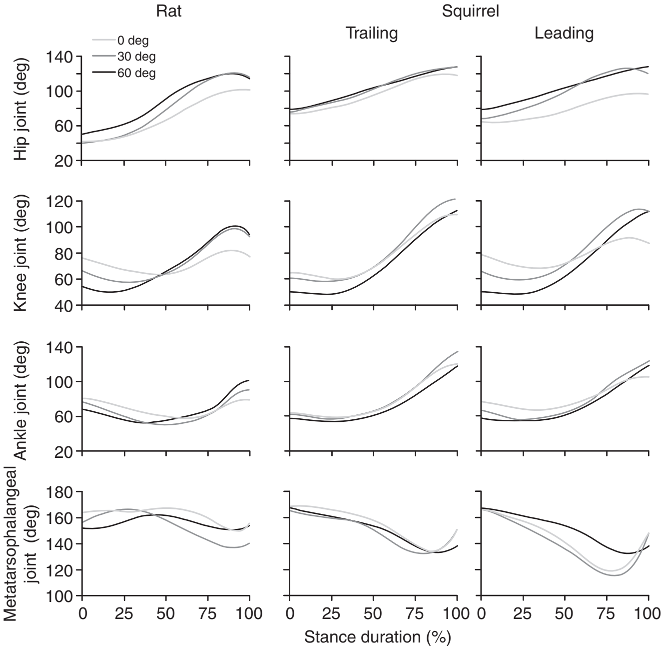

Fig. 6. Mean values of 3-D joint angular excursions during the stance phase of the hindlimbs during locomotion at different substrate orientations in the rat and the European red squirrel.

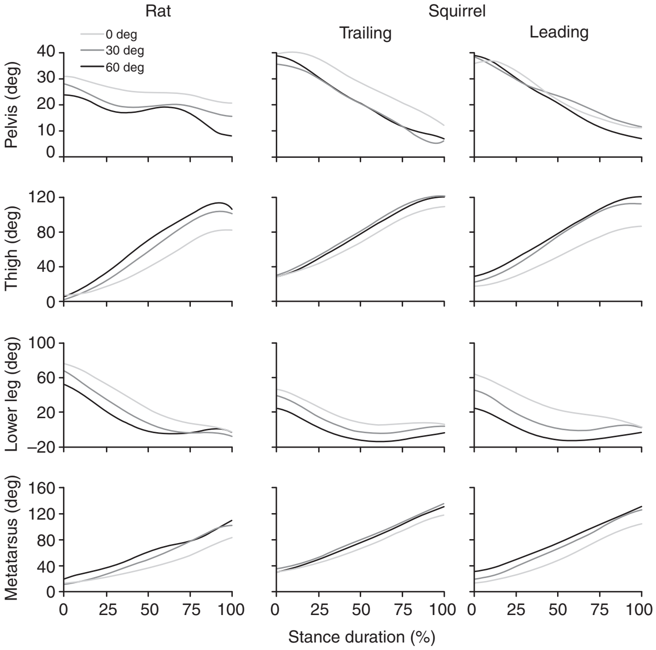

Fig. 7. Mean values of element angular excursions during the stance phase of the hindlimbs during locomotion at different substrate orientations in the rat and the European red squirrel.

were much more affected by substrate orientation than trailing hindlimbs (Figs 6, 7; Tables 5, 6). The differences in hindlimb joints between 0 and 60 deg substrate orientation were, for example, much more pronounced in the leading hindlimbs than in the trailing hindlimbs (see *P*-values in Tables 5 and 6). In European red squirrels, the intermediate 'position' in 3-D angular and element excursions was also observed on the shallow incline (Figs 4–7; Tables 5, 6).

Protraction and retraction angles

We observed significantly lower forelimb and hindlimb protraction in rats during locomotion on 60 deg inclines than during locomotion on 0 and 30 deg inclines. In contrast, forelimb and hindlimb retraction increased significantly with substrate inclination (Fig. 8; Table 7).

In European red squirrels, trailing and leading forelimbs as well as hindlimbs were significantly less protracted with increasing slopes (Fig. 8; Table 7). With one exception, the leading hindlimb, protraction angles of forelimbs and hindlimbs did not differ between 30 and 60 deg inclines (Table 7). In addition, forelimbs and hindlimbs showed greater limb retraction with increasing substrate inclination (Table 7).

Limb position

In rats, forelimb position at touch-down and midstance was significantly affected by substrate orientation (Fig. 9; Tables 7, 8). The comparison of limb positions at touch-down, midstance and lift-off on each substrate revealed that the forelimbs were placed in a constant and slightly adducted position from touch-down until midstance (Fig. 9). At touch-down, mean adduction angles in the forelimbs reached values of approximately 10 deg during locomotion on the horizontal and the shallow inclines. During locomotion on 60 deg inclines, forelimbs were placed parallel relative to the midline of the substrate surface. Maximum adduction angles across all substrate orientations were achieved at lift-off (Fig. 9).

In rats, hindlimb positions at touch-down differed significantly between the locomotion on the horizontal branch and 60 deg substrate inclination (Table 7). At midstance and lift-off, mean adduction angles were comparable across substrate inclinations but differed significantly because of their speed dependency (Fig. 9; Table 7). Contrary to the forelimbs, the hindlimbs were held almost slightly adducted (0–10 deg) throughout the stance phase, regardless of substrate orientation (Fig. 9; Table 8).

In European red squirrels, positions of leading hindlimbs in the transverse plane were completely unaffected by substrate inclination (Table 7). In trailing forelimbs and hindlimbs, substrate inclination was observed to have only minor effects on limb abduction and adduction (Table 7). In contrast, leading forelimbs were positioned almost parallel relative to the substrate during locomotion on the horizontal branch, but adducted slightly on the inclines (Fig. 9; Table 8). Leading forelimbs were significantly less adducted at lift-off than at touch-down during locomotion on inclined substrates (Table 8). This was also observed for the hindlimbs during locomotion on the horizontal branch (trailing hindlimb) and on the shallow incline (leading hindlimb).

Lateral rotations of the thoracic cage and the pelvis

Lateral rotations of the thoracic cage and the pelvis were calculated in relation to the BOS. In rats, the thoracic cage and the pelvis were positioned almost parallel to the direction of movement at touch-down on all substrates (Fig. 10). The lateral rotation of the thoracic cage increased with substrate inclination, with one maximum in the first third of the stance phase and one in the last third. Maximum lateral rotation of the pelvis was achieved in the first third (60 deg) and the last third (0 and 30 deg) of the stance phase. During locomotion on the horizontal branch and the shallow incline, the thoracic cage and the pelvis rotated in opposite directions, causing

Table 5. Results of ANOVAs performed on three-dimensional joint angles and element angles of the trailing forelimbs and hindlimbs of *Sciurus vulgaris*

	Event	*F*	*P*			Speed
			0 *vs* 30 deg	0 *vs* 60 deg	30 *vs* 60 deg	
Shoulder joint	TD	7.8*	<0.05	<0.05	n.s.	s[d]
	MS	8.9*	<0.01	n.s.	n.s.	s[e]
	LO	9.0*	<0.01	<0.05	n.s.	s[e]
Elbow joint	TD	8.9*	n.s.	<0.01	n.s.	s[c]
	MS	8.8	<0.01	<0.001	<0.001	
	LO	2.0	n.s.	n.s.	n.s.	
Wrist joint	TD	200.4**	n.s.	<0.001	<0.001	
	MS[a]	7.7	n.s.	<0.01	<0.001	
	LO	1.8	n.s.	n.s.	n.s.	
Scapula	TD	4.3	<0.001	n.s.	<0.05	
	MS	67.0***	<0.001	<0.05	<0.001	s[d,e]
	LO	59.5***	<0.001	<0.001	<0.05	s[d,e]
Upper arm	TD[a]	11.1*	<0.05	<0.01	n.s.	s[c]
	MS	9.3*	<0.05	<0.01	n.s.	s[d]
	LO	9.9**	<0.05	<0.01	n.s.	s[d]
Lower arm	TD	21.7*	<0.05	<0.01	n.s.	
	MS	10.4	<0.001	n.s.	<0.001	
	LO	34.1*	n.s.	n.s.	n.s.	
Hand	TD	1.9	n.s.	n.s.	n.s.	
	MS	3.2	n.s.	<0.05	<0.05	
	LO	1.0	<0.05	n.s.	n.s.	
Hip joint	TD	3.8	n.s.	n.s.	n.s.	s[b]
	MS	4.3	n.s.	<0.01	n.s.	
	LO	1.3	<0.05	<0.01	n.s.	
Knee joint	TD	6.3*	n.s.	<0.05	n.s.	s[c]
	MS[a]	15.8**	n.s.	<0.01	<0.05	s[c]
	LO	3.3	n.s.	n.s.	n.s.	s[b]
Ankle joint	TD	6.5	n.s.	n.s.	n.s.	
	MS	6.9*	n.s.	<0.01	n.s.	s[d]
	LO	58.8*	<0.01	n.s.	<0.001	
Metatarsophalangeal joint	TD	6.5	n.s.	n.s.	n.s.	
	MS	2.9	n.s.	n.s.	n.s.	
	LO	6.8	n.s.	<0.05	<0.05	
Pelvis	TD	16.5	n.s.	n.s.	n.s.	
	MS[a]	7.2	<0.05	<0.01	n.s.	
	LO	4.6	n.s.	n.s.	n.s.	
Thigh	TD	1.6	n.s.	n.s.	n.s.	s[b]
	MS	8.4*	<0.01	n.s.	n.s.	s[c]
	LO	3.4	n.s.	n.s.	n.s.	s[b]
Lower leg	TD	42.7***	<0.01	<0.001	<0.001	s[d,e]
	MS	7.1*	n.s.	<0.01	n.s.	s[c]
	LO	10.4**	<0.05	n.s.	<0.01	s[d,e]
Metatarsus	TD	5.9*	<0.05	n.s.	n.s.	s[d]
	MS	50.1*	<0.001	<0.001	<0.05	
	LO	35.3*	<0.001	<0.001	n.s.	

Bonferroni *post hoc* tests (*P*-values) were used to compare substrate orientations (0, 30 and 60 deg). *$P<0.05$; **$P<0.01$; ***$P>0.001$.

n.s., not significant; s, correlation with speed, without significant differences in slopes, shifts in elevations or shifts along common slopes between substrate orientations.

[a] Significant differences between individuals.

[b] No significant differences in slope, shift in elevation or shift along common slope.

[c] Significant differences in slope.

[d] Significant shift in elevation.

[e] Significant shift along common slope.

LO, lift-off; MS, midstance; TD, touch-down.

a lateral flexion of the spine. On the steepest incline, lateral rotation of the pelvis was restricted to the first third of the stance phase. Compared with the movements of the thorax, lateral pelvis rotation was relatively low on 60 deg inclines. In European red squirrels, the thoracic cage and the pelvis were held parallel relative to the substrate surface throughout the complete stance phase, regardless of the orientation of the substrate (Fig. 10).

DISCUSSION

The results of this study reveal the challenges that small animals face in their attempts to maintain balance during locomotion on inclined branches. European red squirrels and rats, which are different in their locomotor ecology (one is an arboreal specialist, the other a generalist), adopt very different strategies in response to increasing inclines. Both taxa, however, utilize comparable locomotor adjustments (e.g. strongly

Table 6. Results of ANOVAs performed on three-dimensional joint angles and element angles of the leading forelimbs and hindlimbs of *Sciurus vulgaris*

	Event	*F*	*P*			Speed
			0 *vs* 30 deg	0 *vs* 60 deg	30 *vs* 60 deg	
Shoulder joint	TD^{a}	9.3**	<0.05	<0.01	n.s.	s^{c}
	MS^{a}	2.3	n.s.	n.s.	n.s.	s^{b}
	LO	0.6	n.s.	n.s.	n.s.	
Elbow joint	TD^{a}	13.9***	<0.01.	<0.01	n.s.	s^{c}
	MS^{a}	7.9*	n.s.	<0.05	<0.01	s^{b}
	LO	0.1	n.s.	n.s.	n.s.	
Wrist joint	TD	77.5*	<0.001	<0.05	<0.001	
	MS	28.9*	<0.01	<0.05	<0.001	
	LO	0.4	n.s.	n.s.	n.s.	
Scapula	TD^{a}	13.0**	<0.001	<0.05	n.s.	s^{c,d}
	MS^{a}	68.8***	<0.001	<0.001	n.s.	s^{c,d}
	LO	12.9	<0.001	<0.001	n.s.	
Upper arm	TD	57.8*	<0.001	<0.001	n.s.	
	MS	26.9*	<0.001	<0.001	n.s.	
	LO	12.0	<0.001	<0.001	n.s.	
Lower arm	TD^{a}	2.0	<0.01	n.s.	n.s.	
	MS	12.6	<0.001	<0.05	<0.01	
	LO	65.4*	<0.05	<0.001	<0.05	
Hand	TD	11.9	n.s.	<0.001	<0.001	
	MS	4.6	n.s.	<0.001	<0.001	
	LO	8.2*	<0.05	<0.01	n.s.	s^{d}
Hip joint	TD	7.7*	n.s.	<0.01	n.s.	s^{b}
	MS	19.3*	<0.001	<0.001	n.s.	
	LO^{a}	77.4*	<0.001	<0.001	n.s.	
Knee joint	TD^{a}	40.1***	n.s.	<0.001	<0.001	s^{c,d}
	MS	8.1*	n.s.	<0.01	n.s.	s^{b}
	LO^{a}	22.9***	<0.001	<0.001	n.s.	s^{c,d}
Ankle joint	TD	8.1	<0.05	<0.001	<0.05	
	MS	14.0***	<0.05	<0.001	n.s.	s^{c}
	LO	15.9	<0.01	<0.01	n.s.	
Metatarsophalangeal joint	TD	0.4	n.s.	n.s.	n.s.	
	MS	10.7	n.s.	<0.01	<0.001	
	LO	1.4	n.s.	n.s.	n.s.	
Pelvis	TD	1.9	n.s.	n.s.	n.s.	
	MS	0.8	n.s.	n.s.	n.s.	
	LO	0.4	n.s.	n.s.	n.s.	
Thigh	TD	23.6***	<0.01	<0.001	n.s.	s^{c}
	MS^{a}	15.0**	<0.05	<0.01	n.s.	s^{b}
	LO	9.0**	<0.05	<0.01	n.s.	s^{b}
Lower leg	TD^{a}	21.5*	<0.001	<0.001	<0.001	
	MS	12.1**	n.s.	<0.01	<0.05	s^{b}
	LO^{a}	10.2**	n.s.	n.s.	<0.01	s^{c,d}
Metatarsus	TD	21.3**	n.s.	<0.01	<0.05	s^{b}
	MS	13.5	<0.001	<0.001	<0.05	
	LO	22.4*	<0.001	<0.001	n.s.	

Bonferroni *post-hoc* tests (*P*-values) were used to compare substrate orientations (0, 30 and 60 deg). *P<0.05; **P<0.01; ***P<0.001.

n.s., non significant; s, correlation with speed, without significant differences in slopes, shifts in elevations or shifts along common slopes between substrate orientations.

[a] Significant differences between individuals.

[b] No significant differences in slope, shift in elevation or shift along common slope.

[c] Significant shift in elevation.

[d] Significant shift along common slope.

LO, lift-off; MS, midstance; TD, touch-down.

flexed limbs, decrease in protraction angles and increase in retraction angles) during locomotion on inclined braches, which may be caused by similar biomechanical constraints. We suggest that these locomotor adjustments, which have also been observed in primates and marsupials, might be strategies used by early mammals.

Kinematic changes on inclined branches

Regardless of the way animals respond to differences in substrate orientation, numerous kinematic parameters are surprisingly similar and may be caused by similar biomechanical constraints during locomotion on inclined supports (Fig. 11). As described previously for various mammals, rats and European red squirrels flexed their limbs significantly with an increase in substrate orientation (Prost and Sussman, 1969; Vilensky et al., 1994; Carlson-Kuhta et al., 1998; Nakano, 2002; Stevens, 2003; Lammers, 2007; Nyakatura et al., 2008). The increase in limb flexion on inclines results in a center of mass positioned closer to the substrate surface, which consequently increases friction during the stance phase.

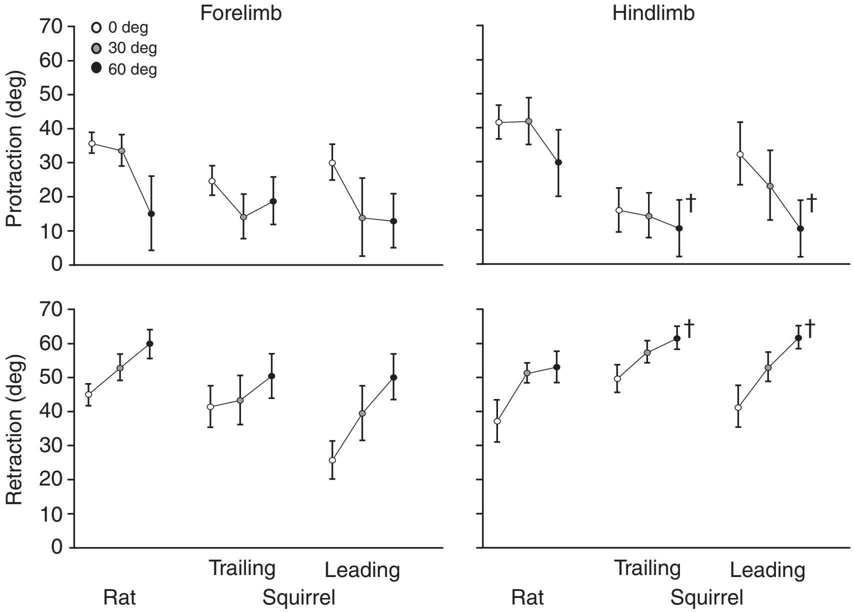

Fig. 8. Mean (±s.d.) values of protraction and retraction angles at touch-down and lift-off of the forelimbs and hindlimbs of the rat and the European red squirrel for each substrate orientation. Values of both hindlimbs were merged in the analyses because of the synchronous touch-down events (†).

As hypothesized, rats and squirrels do not differ in general limb placement from primates, which possesses prehensile extremities. All the animals investigated so far place their forelimbs more laterally and/or underneath steeply inclined branches compared with horizontal or 30 deg inclined branches in order to fix the body on the substrate by pulling on the substrate, to bring the center of mass closer to the substrate surface and thus to avoid toppling backwards (Fig. 2) (Nakano, 2002; Stevens, 2003).

However, available data on limb placement during inclined locomotion in various mammals imply significant differences in the way the forelimbs make contact with the branch, in particular the timing and position. Aye-ayes, which are able to grasp branches of 80 mm in diameter, switch from a 'full-curl' hand position during locomotion on a horizontal branch to a 'full-grip' hand position on slightly ascending branches (30–35 deg) (Krakauer et al., 2002). This may be due to the need to generate tensile forces as they move upwards. Differences in the positioning of the forelimbs (top *vs* laterally *vs* underneath) with increasing substrate inclination have been described for the gibbon and the macaque (Nakano, 2002). The gibbon is able to change limb position at lower inclines (40 deg) than the macaque (50 deg). Nakano proposed that limb placement on inclined substrates depends mainly on differences in limb proportions and the position of the estimated center of mass (Nakano, 2002).

The point at which the shift between forelimb positions on inclined branches (top *vs* laterally) in rats and squirrels takes place was not resolved in the present study. Our statistical analyses reveal that the shift occurs at an incline between 30 and 60 deg (Table 3). This is in agreement with previous estimations considering the point at which climbing begins (see below). It seems likely that gray short-tailed opossums (Lammers et al., 2004; Lammers, 2007), rats and squirrels, which are not able to grasp the branch, overcome the problem of generating the required tensile forces on steeper inclines by using their claws. On shallow inclines (approximately 30 deg), however, forelimb positions of these mammals are quite similar to horizontal locomotion. These unchanged forelimb positions (between 0 and 30 deg substrate orientation) probably characterize the locomotion of mammals without grasping extremities because of the need to maintain stability. We propose that animals without grasping extremities increase security during locomotion on shallow inclines by placing their limbs in the same position for as long as possible. Moreover, placing the limbs more laterally should increase the risk of sliding down the branch and thus increase the muscle effort required to generate mediolateral pressure to prevent toppling down. A highly standardized touch-down position during terrestrial locomotion has also been argued to be a mechanism for maintaining dynamic stability (Fischer et al., 2002; Schilling, 2005; Daley and Biewener, 2006; Fischer and Blickhan, 2006; Blickhan et al., 2007).

Larger protraction angles on inclined substrates should also be disadvantageous because of the expected increase in muscle work that is required to pull on the branch with a much more extended forelimb. Hence, animals that lack or have reduced grasping abilities try to maintain comparable functional roles of both forelimbs and hindlimbs for as long as possible during locomotion on inclined branches.

In previous studies, the hindlimbs of primates, rats and squirrels were placed on or near the top of steeply inclined branches, suggesting a relationship between hindlimb placement and propulsive function (Hirasaki et al., 1993; Nakano, 2002). The gray short-tailed opossum, however, placed its hindlimbs more laterally than the forelimbs as they moved on 30 deg inclines (Lammers et al., 2004; Lammers, 2007). The placement of the hindlimbs was comparable to their placement on horizontal branches.

The fact that, during locomotion on sloped branches, animals without grasping abilities try to place their forelimbs and hindlimbs in the same position for as long as possible explains the 'intermediate position' in the 3-D angular and element excursions on 30 deg inclines in rats and European red squirrels. This is supported by the similar contact times observed in the forelimbs and hindlimbs and the similar protraction/retraction and abduction/adduction angles seen during locomotion on the horizontal branch and the shallow inclined branch (see below). Despite these similarities in limb placement, further studies are needed in order to clarify whether the shift in forelimb positions depends on differences in limb proportions, the position of the center of mass, how mammals make contact with the substrate (e.g. claws and prehensile extremities) or a combination of these factors.

Another common pattern is the increase in limb extension at lift-off to push the body forward and thus to act against gravity (Fig. 11). The increase in limb extension at lift-off during inclined locomotion

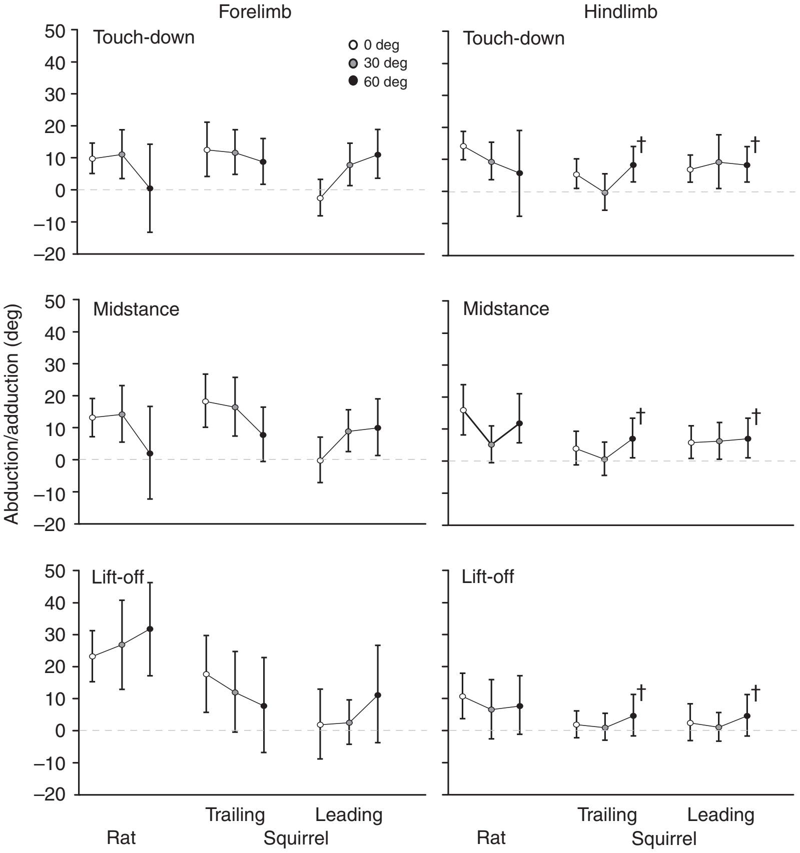

Fig. 9. Mean (±s.d.) values of the angles of adduction and abduction at touch-down, midstance and lift-off of the forelimbs and hindlimbs of the rat and the European red squirrel for each substrate orientation. Values of both hindlimbs were merged in the analyses because of the synchronous touch-down events (†). Values near zero indicate a parasagittal limb position, values higher than zero an adducted limb position and values below zero an abducted limb position.

leads to higher retraction angles and is common among a wide range of species (Vilensky et al., 1994; Carlson-Kuhta et al., 1998; Gillis and Biewener, 2001; Stevens, 2003; Nyakatura et al., 2008). It is related to the higher propulsive forces required on inclines (Preuschoft, 2002).

The decrease in protraction angles across sloping substrates also seems to be a common pattern, regardless of the chosen gait (asymmetrical or symmetrical) and the substrate (terrestrial or branch) (Carlson-Kuhta et al., 1998; Nyakatura et al., 2008).

Static *vs* dynamic stability during inclined locomotion

The comparison of all available data gives rise to the assumption that speed and gait adjustments, rather than the locomotor adjustments listed above, affect the selection of climbing strategies (static stability *vs* dynamic stability) (Fig. 11). Rats, for example, change their preferred locomotor strategy as substrate orientation increases and switch from a dynamic stable locomotion on horizontal branches (Schmidt and Fischer, 2010) to a more static one as they move on inclines. The latter is associated with a reduction in speed

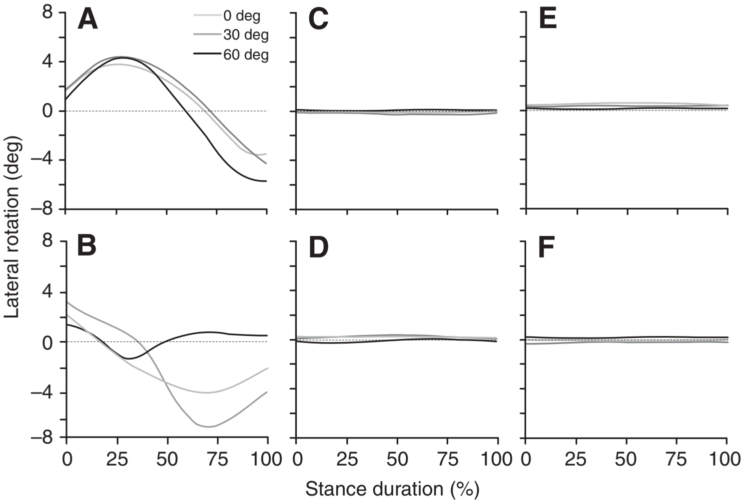

Fig. 10. Changes in the angles of the lateral rotation of the thoracic cage (top) and the pelvis (bottom) during the stance phase across different substrate orientations in the (A,B) rat; (C,D) European red squirrel, trailing limb; and (E,F) European red squirrel, leading limb.

Table 7. Results of ANOVAs performed on protraction angles (αPA), retraction (αRA) and limb position (αLP) of the forelimbs and hindlimbs of *Rattus norvegicus* and *Sciurus vulgaris*

		P			
	F	0 *vs* 30 deg	0 *vs* 60 deg	30 *vs* 60 deg	Speed
Rattus norvegicus					
Forelimbs					
αPA	29.4*	n.s.	<0.001	<0.001	
αRA	75.2*	<0.001	<0.001	<0.001	
αLP – TD	3.5	n.s.	<0.05	<0.01	
αLP – MS[a]	77.4*	n.s.	<0.05	<0.01	
αLP – LO	3.4	n.s.	n.s.	n.s.	
Hindlimbs					
αPA	3.5	n.s.	<0.001	<0.001	
αRA	40.2*	<0.001	<0.001	n.s.	
αLP – TD[a]	23.2*	n.s.	<0.05	n.s.	
αLP – MS	81.3***	<0.001	<0.001	<0.001	s[c]
αLP – LO	57.7***	<0.001	<0.001	<0.001	s[c]
Sciurus vulgaris					
Trailing forelimb					
αPA[a]	165.4**	<0.001	<0.05	n.s.	
αRA[a]	18.7***	<0.05	<0.001	<0.01	s[c]
αLP – TD	0.3	n.s.	n.s.	n.s.	
αLP – MS	3.0	n.s.	<0.01	<0.05	
αLP – LO	2.7	n.s.	n.s.	n.s.	
Leading forelimb					
αPA	9.9	<0.001	<0.001	n.s.	
αRA	233.0**	<0.001	<0.001	<0.001	
αLP – TD	18.9***	<0.001	<0.001	n.s.	s[c,d]
αLP – MS	11.3**	<0.01	<0.01	n.s.	s[d]
αLP – LO[a]	7.5*	n.s.	<0.01	n.s.	s[c]
Trailing hindlimb					
αPA	11.8**	n.s.	<0.01	n.s.	s[b]
αRA[a]	209.6**	<0.001	<0.001	<0.01	
αLP – TD	25.8*	<0.01	n.s.	<0.001	
αLP – MS	6.1	n.s.	n.s.	<0.01	
αLP – LO	2.9	n.s.	n.s.	n.s.	
Leading hindlimb					
αPA	59.3***	<0.01	<0.001	<0.001	s[c,d]
αRA[a]	24.8*	<0.001	<0.001	<0.001	
αLP – TD	0.7	n.s.	n.s.	n.s.	
αLP – MS	0.3	n.s.	n.s.	n.s.	
αLP – LO[a]	1.2	n.s.	n.s.	n.s.	

Bonferroni *post hoc* tests (P-values) were used to compare substrate orientations (0, 30 and 60 deg). *$P<0.05$, **$P<0.01$; ***$P<0.001$.

n.s., non significant; s, correlation with speed, without significant differences in slopes, shifts in elevations or shifts along common slopes between substrate orientations.

[a] Significant differences between individuals.

[b] No significant differences in slope, shift in elevation or shift along common slope.

[c] Significant shift in elevation.

[d] Significant shift along common slope.

LO, lift-off; MS, midstance; TD, touch-down.

and an increase in stance duration. A decrease in travel speed on inclines has also been observed in various primates (Nakano, 2002; Stevens, 2003). Interestingly, standard deviations of speed in these species always follow a pattern, decreasing as substrate inclination increases and *vice versa* (Nakano, 2002; Stevens, 2003). This does not happen in small-bodied and agile mammals such as European red squirrels, tamarins and lemurids, which show high locomotor speeds during inclined locomotion (Stevens, 2003; Nyakatura et al., 2008).

The ratio of stance duration between forelimbs and hindlimbs in rats and European red squirrels is similar across all inclines despite the observed differences in travel speed and climbing strategies. This is surprising because the hindlimbs support more body weight and have to contribute more to propulsion with increasing substrate orientation (Preuschoft, 2002). Therefore, it seems likely that forelimbs and hindlimbs fulfill different functions of equal importance during incline locomotion, particularly in mammals that lack prehensile extremities. The forelimbs provide substrate contact and prevent the animal from toppling backwards whereas the hindlimbs generate high propulsive forces. The significant increase (approximately 100%) in stance duration in rats ties in with the need for stability and security of this unspecialized rodent (static stability). European red squirrels, in contrast, achieve dynamic stability by maintaining travel speed and display relatively short contact times across all inclines (Fig. 11).

Lateral rotations of the thoracic cage and the pelvis

On the steepest incline, the thoracic cage and the pelvis in European red squirrels are positioned parallel to the substrate. Thus, both forelimbs and hindlimbs are slightly adducted throughout the stance

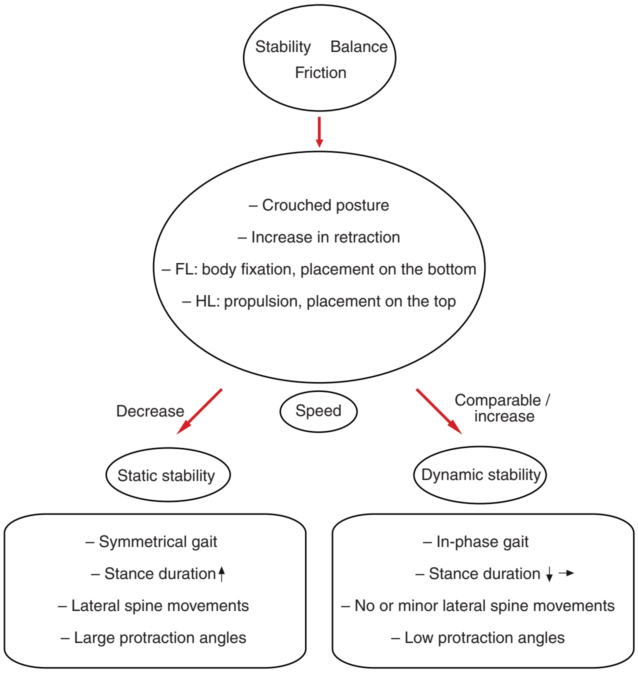

Fig. 11. Proposed relationships between biomechanical constraints, required locomotor adjustments, preferred locomotion strategies and travel speeds during inclined locomotion. FL, forelimbs; HL, hindlimbs.

phase, which should reduce the forces required for joint stabilization in the transverse plane (Witte et al., 2002; Schmitt, 2003a; Fischer et al., 2010). In rats, lateral rotations of the thoracic cage increase as substrate orientation increases, thereby enabling rats to grip the substrate as laterally and far underneath as possible with their relatively flexed limbs (Fig. 2). In comparison to the significant lateral rotations of the thoracic cage on the steepest incline, the pelvis remains relatively parallel to the substrate surface during the stance phase. This relates to the predominantly propulsive function of the hindlimbs. As mentioned above, joint stabilization in the transverse plane and thus the expenditure of energy is reduced if the hindlimb is positioned more parasagittally for the duration of the stance phase. The hindlimbs, which bear more and more weight as the stance phase progresses, relieve the forelimbs, giving them and the front body increased flexibility. In order to permit the lateral rotation of the thoracic cage while the pelvis is held in a relatively constant parallel position, huge tilting movements take place in the pelvis (A.S., unpublished data).

Implications for our understanding of climbing

To date, definitions of climbing have been based exclusively on theoretical advisements. Climbing "...refers to locomotion on supports with vertical or steeply sloping surfaces..." (Cartmill, 1985) and "...means performing locomotion connected with a gain or a controlled loss of height, in which the distribution of body weight substantially deviates from the situation on level substrates. Tensile forces, or rotational moments, are transmitted between an animal

Table 8. Results of ANOVAs performed on limb position αLP of the forelimbs (FL) and hindlimbs (HL) of *Rattus norvegicus* and *Sciurus vulgaris*

		P			
	F	TD *vs* MS	TD *vs* LO	MS *vs* LO	Speed
Rattus norvegicus					
αLP-FL (0 deg)[a]	122.6**	n.s.	<0.001	<0.01	
αLP-FL (30 deg)	9.8	n.s.	<0.01	<0.05	
αLP-FL (60 deg)	13.6	n.s.	<0.001	<0.001	
αLP-HL (0 deg)	4.3	n.s.	n.s.	n.s.	s[b]
αLP-HL (30 deg)	3.5	n.s.	n.s.	n.s.	
αLP-HL (60 deg)	2.6	n.s.	n.s.	n.s.	
Sciurus vulgaris					
Trailing forelimb					
αLP-FL (0 deg)	4.8	n.s.	n.s.	n.s.	
αLP-FL (30 deg)[a]	2.7	n.s.	n.s.	n.s.	
αLP-FL (60 deg)[a]	0.4	n.s.	n.s.	n.s.	
Leading forelimb					
αLP-FL (0 deg)	3.7	n.s.	n.s.	n.s.	s[b]
αLP-FL (30 deg)[a]	9.1*	n.s.	<0.05	<0.01	s[c]
αLP-FL (60 deg)[a]	5.9*	n.s.	<0.05	n.s.	s[b]
Trailing hindlimb					
αLP-HL (0 deg)	1.1	n.s.	n.s.	n.s.	
αLP-HL (30 deg)	0.3	n.s.	n.s.	n.s.	
αLP-HL (60 deg)	1.1	n.s.	n.s.	n.s.	
Leading hindlimb					
αLP-HL (0 deg)[a]	2.5	n.s.	n.s.	n.s.	
αLP-HL (30 deg)	16.5	n.s.	<0.01	n.s.	
αLP-HL (60 deg)	1.1	n.s.	n.s.	n.s.	

Bonferroni *post hoc* tests (*P*-values) were used to compare substrate orientations (0, 30 and 60 deg). $^*P<0.05$; $^{**}P<0.01$.

n.s., non significant; s, correlation with speed, without significant differences in slopes, shifts in elevations or shifts along common slopes between substrate orientations.

[a] Significant differences between individuals.

[b] No significant differences in slope, shift in elevation or shift along common slope.

[c] Significant shift in elevation.

LO, lift-off; MS, midstance; TD, touch-down.

and its substrate and these forces may well assume higher values than the compressive forces acting on the limbs...” (Preuschoft, 2002). Hunt and colleagues draw a theoretical distinction between various modes of climbing based on substrate orientation, namely that a substrate orientation of approximately 45 deg forms the boundary between climbing and non-climbing locomotor modes (e.g. walking) (Hunt et al., 1996). The results of the present study reveal that the transition between the two locomotor modes occurs between 30 and 60 deg regardless of the preferred gait and speed, in support of Hunt et al. (Hunt et al., 1996). The kinematic and metric parameters that are influenced by differences in substrate orientation and are thus helpful in defining the term ‘climbing’ in more detail include 3-D joint angles, element angles and limb placement, as well as protraction and retraction angles. The shift between the two modes is presumably caused by the decrease in compressive forces in the forelimbs during locomotion on steeper inclines and *vice versa* for the hindlimbs. As mentioned above, animals have to overcome the limitation of lower compressive forces in the forelimbs by the use of prehensile extremities and/or claws. As a result, tensile forces in the forelimbs increase and become the main force component as vertical forces decrease (A.S., unpublished data). This supports Preuschoft’s (Preuschoft, 2002) definition of climbing. The point at which climbing begins thus could be defined as the time at which the functional differentiation of both forelimbs and hindlimbs takes place (safety *vs* propulsive function). Thus, it seems likely that this transition is species specific and highly related to the position of the center of mass and limb proportions (Nakano, 2002). As the present results are restricted to one diameter of support and two substrate inclinations in two clawed species, it is impossible to draw general conclusions about climbing. In other words, the angle at which climbing starts remains unknown. Further studies are needed to pinpoint exactly the metric, kinematic and kinetic differences between locomotion on shallow and steep inclines (branches and trackways). The effect of the use of claws during incline locomotion on arboreal substrates and in trees remains, in particular, an unknown factor.

Evolutionary implications

According to Jenkins (Jenkins, 1974), the locomotor mode of early mammals includes a highly flexible vertebral column and flexed, abducted limbs and may have evolved as an adaptation to moving on uneven and irregular substrates. Studies dealing with the locomotion on inclined substrates support this hypothesis because most of the locomotor adjustments (increase in limb flexion and limb retraction, functional differentiation between forelimbs and hindlimbs) are independent of travel speed and morphological adaptations (e.g. prehensile extremities and limb proportions). Therefore, it seems likely that these locomotor adjustments on inclined substrates might be strategies used by early mammals as well. Our results also strongly imply that the need for a stable and secure locomotion in the ‘fine-branch’ environment benefits the development of prehensile extremities in early primates.

ACKNOWLEDGEMENTS

We thank Sebastian Köhring and Jörg Mämpel (Technical University Ilmenau) for the construction of the experimental setup. Thanks also go to Rommy Petersohn for her technical assistance with the X-ray facility and to Florian Wolf for digitizing the X-ray movies. We thank Lucy Cathrow, Lisa Cashmore and Jillian S. Davis (Ohio University) for editing the language of the text. We are grateful to Audrone R. Biknevicius, Nancy J. Stevens (Ohio University) and David V. Lee (University of Nevada) for helpful comments and constructive criticism. We also thank two anonymous reviewers for their important contributions to the final manuscript. This study was funded by the Federal Ministry of Education and Research (BMBF, “InspiRat” Fkz 01RI0633).

REFERENCES

Alexander, R. M. (ed.) (2003). Climbing and jumping. In *Principles of Animal Locomotion*, pp. 146-165. Princeton, NJ: Princeton University Press.

Andrada, E., Mämpel, J., Schmidt, A., Fischer, M. S. and Witte, H. (2010). Biomechanical analyses of rat locomotion during walking and climbing as a base for the design and construction of climbing robots. In: *Design and Nature V. Comparing Design in Nature with Science and Engineering* (ed. C. A. Brebbia and A. Carpi), pp. 165-178. Southampton: WIT.

Armstrong, R. B., Laughlin, M. H., Rome, L. and Taylor, C. R. (1983). Metabolism of rats running up and down an incline. *J. Appl. Physiol.* **55**, 518-521.

Blickhan, R., Seyfarth, A., Geyer, H., Grimmer, S., Wagner, H. and Günther, M. (2007). Intellgence by mechanics. *Philos. Trans. R. Soc. A* **365**, 199-220.

Byron, C., Kunz, H., Matuszek, H., Lewis, S. and Van Valkinburgh, D. (2011). Rudimentary pedal grasping in mice and implications for terminal branch arboreal quadrupedalism. *J. Morphol.* **272**, 230-240.

Carlson, K. J. and Demes, B. (2010). Gait dynamics of *Cebus apella* during quadrupedalism on different substrates. *Am. J. Phys. Anthropol.* **142**, 273-286.

Carlson, K. J., Demes, B. and Franz, T. M. (2005). Mediolateral forces associated with quadrupedal gaits of lemurids. *J. Zool. (Lond.)* **266**, 261-273.

Carlson-Kuhta, P., Trank, T. V. and Smith, J. L. (1998). Forms of forward quadrupedal locomotion. II. A comparison of posture, hindlimb kinematics, and motor patterns for upslope and level walking. *J. Neurophysiol.* **79**, 1687-1701.

Cartmill, M. (1974). Pads and claws in arboreal locomotion. In *Primate Locomotion* (ed. F. A. Jenkins), pp. 45-83. New York: Academic Press.

Cartmill, M. (1985). Climbing. In *Functional Vertebrate Morphology* (ed. M. Hildebrand, D. M. Bramble, K. F. Liem and D. B. Wake), pp. 73-88. Cambridge, MA: Harvard University Press.

Clarke, K. A. (1992). A technique for the study of spatiotemporal aspects of paw contact patterns, applied to rats treated with a TRH analogue. *Behav. Res. Methods* **24**, 407-411.

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.

Demes, B. and Carlson, K. J. (2009). Locomotor variation and bending regimes of capuchin limb bones. *Am. J. Phys. Anthropol.* **139**, 558-571.

Essner, R. L., Jr (2007). Morphology, locomotor behavior and microhabitat use in North American squirrels. *J. Zool.* **22**, 101-109.

Falster, D. S., Warton, D. I. and Wright, I. J. (2003). (S)MATR: Standardised major axis tests and routines. Version 2.0. http://www.bio.mq.edu.au/ecology/SMA TR/.

Fischer, M. S. (1994). Crouched posture and high fulcrum, a principle in the locomotion of small mammals: the example of the rock hyrax (*Procavia capensis*) (Mammalia: Hyracoidea). *J. Hum. Evol.* **26**, 501-524.

Fischer, M. S. and Blickhan, R. (2006). The tri-segmented limbs of therian mammals: kinematics, dynamics, and self-stabilization – a review. *J. Exp. Zool.* **305**, 935-952.

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.

Fischer, M. S., Krause, C. and Lilje, K. E. (2010). Evolution of chameleon locomotion, or how to become arboreal as a reptile. *Zoology* **113**, 67-74.

Franz, T. M., Demes, B. and Carlson, K. J. (2005). Gait mechanics of lemurid primates on terrestrial and arboreal substrates. *J. Hum. Evol.* **48**, 199-217.

Gebo, D. L. (2004). A shrew-sized origin for primates. *Yearb. Phys. Anthropol.* **47**, 40-62.

Gillis, G. B. and Biewener, A. A. (2001). Effects of surface grade on proximal hindlimb muscle strain and activation during rat locomotion. *J. Appl. Physiol.* **93**, 1731-1743.

Hanna, J. B., Schmitt, D. and Griffin, T. M. (2008). The energetic cost of climbing in primates. *Science* **320**, 898.

Hirasaki, E., Kumakura, H. and Matano, S. (1993). Kinesiological characteristics of vertical climbing in *Ateles geoffroyi* and *Macaca fuscata*. *Folia Primat.* **61**, 148-156.

Hunt, K. D., Cant, J. G. H., Gebo, D. L., Rose, M. D., Walker, S. E. and Youlatos, D. (1996). Standardized descriptions of primate locomotor and postural modes. *Primates* **37**, 363-387.

Imaizumi, Y. (1978). Climbing behavior of *Apodemus argenteus* and *Apodemus speciosus* (Rodentia: Muridae). *Appl. Entomol. Zool.* **13**, 304-307.

Jenkins, F. A., Jr (1974). Tree shrew locomotion and the origins of primate arborealism. In *Primate Locomotion* (ed. F. A. Jenkins, Jr), pp. 85-115. New York: Academic Press.

Jenkins, F. A., Jr and Camazine, S. M. (1977). Hip structure and locomotion in ambulatory and cursorial carnivores. *J. Zool. (Lond.)* **181**, 351-370.

Jenkins, F. A., Jr and Weijs, W. A. (1979). The functional anatomy of the shoulder in the Virginia opossum (*Didelphis virginia*). *J. Zool. (Lond.)* **188**, 379-410.

Kivell, T. L., Schmitt, D. and Wunderlich, R. E. (2010). Hand and foot pressures in the aye-aye (*Daubentonia madagascariensis*) reveal novel biomechanical trade-offs required for walking on gracile digits. *J. Exp. Biol.* **213**, 1549-1557.

Koopmans, G. C., Deumens, R., Brook, G., Gerver, J., Honig, W. M. M., Hamers, F. P. T. and Joosten, E. A. J. (2007). Strain and locomotor speed affect over-ground locomotion in intact rats. *Physiol. Behav.* **92**, 993-1001.

Krakauer, E., Lemelin, P. and Schmitt, D. (2002). Hand and body position during locomotor behavior in the aye-aye (*Daubentonia madagascariensis*). *Am. J. Primat.* **57**, 105-118.

Ladine, T. A. and Kissel, R. E., Jr (1994). Escape behavior of Virginia opossums. *Am. Mid. Nat.* **132**, 234-238.

Lammers, A. R. (2007). Locomotor kinetics on sloped arboreal and terrestrial substrates in a small quadrupedal mammal. *Zoology* **110**, 93-103.

Lammers, A. R. and Biknevicius, A. R. (2004). The biodynamics of arboreal locomotion: the effects of substrate diameter on locomotor kinetics in the gray short-tailed opossum (*Monodelphis domestica*). *J. Exp. Biol.* **207**, 4325-4336.

Lammers, A. R., Earls, K. D. and Biknevicius, A. R. (2006). Locomotor kinetics and kinematics on inclines and declines in the gray short-tailed opossum *Monodelphis domestica*. *J. Exp. Biol.* **209**, 4154-4166.

Lemelin, P. and Cartmill, M. (2010). The effect of substrate size on the locomotion and gait patterns of the kinkajou (*Potos flavus*). *J. Exp. Zool.* **313**, 157-168.

Montgomery, W. I. (1980). The use of arboreal runways by woodland rodents, *Apodemus sylvaticus* (L.), *A. flavicollis* (Melchoir) and *Clethrionomys glareolus* (Schreber). *Mammal Rev.* **10**, 189-195.

Nakano, Y. (2002). The effects of substratum inclination on locomotor patterns in primates. *Z. Morphol. Anthropol.* **83**, 189-199.

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**, 13-25.

Piechocki, R. (1958). Die Zwergmaus, *Micromys minutus* Pallas. Vol. 222. Wittenberg, Lutherstadt: Neue Brehm-Bücherei.

Preuschoft, H. (2002). What does “arboreal locomotion” mean exactly and what are the relationships between “climbing”, environment and morphology? *Z. Morphol. Anthropol.* **83**, 171-188.

Prost, J. H. and Sussman, R. W. (1969). Monkey locomotion on inclined surfaces. *Am. J. Phys. Anthropol.* **31**, 53-58.

Schilling, N. (2005). Ontogenetic development of locomotion in small mammals – a kinematic study. *J. Exp. Biol.* **208**, 4013-4034.

Schmidt, A. (2011). Functional differentiation of trailing and leading forelimbs during locomotion on the ground and on a horizontal branch in the European red squirrel (*Sciurus vulgaris*, Rodentia). *Zoology*, doi:10.1016/j.zool.2011.01.001.

Schmidt, A. and Fischer, M. S. (2010). Arboreal locomotion in rats – the challenge of maintaining stability. *J. Exp. Biol.* **213**, 3615-3624.

Schmidt, M. (2005). Hind limb proportions and kinematics: are small primates different from other small mammals? *J. Exp. Biol.* **208**, 3367-3383.

Schmidt, M. (2008). Forelimb proportions and kinematics: how are small primates different from other small mammals? *J. Exp. Biol.* **211**, 3775-3789.

Schmitt, D. (1994). Forelimb mechanics as a function of substrate type during quadrupedalism in two anthropoid primates. *J. Hum. Evol.* **26**, 441-457.

Schmitt, D. (1999). Compliant walking in primates. *J. Zool. (Lond.)* **248**, 149-160.

Schmitt, D. (2003a). Mediolateral reaction forces and forelimb anatomy in quadrupedal primates: implications for interpreting locomotor behavior in fossil primates. *J. Hum. Evol.* **44**, 47-58.

Schmitt, D. (2003b). Evolutionary implications of the unusual walking mechanics of the common marmoset (*C. jacchus*). *Am. J. Phys. Anthropol.* **122**, 28-37.

Schmitt, D. and Hanna, J. B. (2004). Substrate alters forelimb to hindlimb peak force ratios in primates. *J. Hum. Evol.* **46**, 239-254.

Schmitt, D. and Lemelin, P. (2002). Origins of primate locomotion: gait mechanics of the woolly opossum. *Am. J. Phys. Anthropol.* **118**, 231-238.

Snyder, G. and Carello, C. (2008). Body mass and the energy efficiency of locomotion: lessons from incline running. *Comp. Biochem. Physiol.* **150A**, 144-150.

Stevens, N. J. (2003). The influence of substrate size, orientation and compliance upon prosimian arboreal quadrupedalism. PhD thesis, Stony Brook University, Stony Brook, NY, USA.

Vasquez, M. R. (2004). Comparative anatomy of *Henkelotherium guimarotae* (Holotheria), a late Jurassic small mammal, and its relevance for the evolution of the mode of locomotion of modern mammals. PhD thesis, Freie Universität, Berlin, Germany.

Vilensky, J. A., Moore, A. M. and Libii, J. N. (1994). Squirrel monkey locomotion on an inclines treadmill: implications for the evolution of gaits. *J. Hum. Evol.* **26**, 375-386.

Warton, D. I. and Weber, N. C. (2002). Common slope tests for bivariate structural relationships. *Biometr. J.* **44**, 161-174.

Wickler, S. J., Hoyt, D. F., Cogger, E. A. and Hirschbein, M. H. (2000). Preferred speed and cost of transport: the effect of incline. *J. Exp. Biol.* **203**, 2195-2200.

Witte, H., Preuschoft, H. and Fischer, M. S. (2002). The importance of the evolutionary heritage of locomotion on flat ground in small mammals for the development of arboreality. *Z. Morphol. Anthropol.* **83**, 221-233.

Wu, G. and Cavanagh, P. R. (1995). ISB recommendations for standardization in the reporting of kinematic data. *J. Biomech.* **28**, 1257-1261.

Wunder, B. A. and Morrison, P. R. (1974). Red squirrel metabolism during incline running. *Comp. Biochem. Physiol.* **48**, 153-161.

Youlatos, D. (1999). Locomotor and postural behavior of *Sciurus igniventris* and *Microsciurus flaviventer* (Rodentia, Sciuridae) in eastern Ecuador. *Mammalia* **63**, 405-416.

Zwahlen, R. (1975). Die lokomotorische Aktivität des Eichhörnchens (*Sciurus vulgaris*). *Oecologica* **22**, 79-98.