RESEARCH ARTICLE | *Control of Movement*

Kinematics of forward and backward locomotion performed in different environmental conditions

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Submitted 12 April 2019; accepted in final form 7 October 2019

Vemula MG, Deliagina TG, Zelenin PV. Kinematics of forward and backward locomotion performed in different environmental conditions. *J Neurophysiol* 122: 2142–2155, 2019. First published October 9, 2019; doi:10.1152/jn.00239.2019.—Mice are frequently used in analyses of the locomotor system. Although forward locomotion (FWL) in intact mice has been studied previously, backward locomotion (BWL) in mice has never been analyzed. The aim of the present study was to compare kinematics of FWL and BWL performed in different environmental conditions (i.e., in a tunnel, on a treadmill, and on an air-ball). In all setups, the average speed and step amplitude during BWL were significantly reduced compared with FWL. The cycle duration varied greatly during both FWL and BWL. The average swing duration during BWL was twice shorter than during FWL on each setup. Mice exhibited different interlimb coordinations (trot and walk with lateral or diagonal sequence) during BWL but only one gait (walk with lateral sequence) during FWL. Location of the rostro-caudal paw trajectory in relation to the hip projection to the surface (HP) depended on hip height. With low hip height, the trajectory was displaced either rostrally (anterior steps) or caudally (posterior steps) to HP. With high hip height, HP was near the middle of the trajectory (middle steps). During FWL, all three forms of steps were observed in the tunnel and predominantly anterior and posterior steps on the treadmill and air-ball, respectively. During BWL, only anterior steps were observed. Intralimb coordination depended on the form of stepping. Limb joints were coordinated to keep the hip at approximately constant height during stance and to have the smallest functional limb length during swing when the limb passed under the hip.

NEW & NOTEWORTHY Mice are extensively used for the analysis of the locomotor system. This study is the first examination of the kinematics of forward and backward locomotor movements in different environmental conditions in mice. Obtained results represent a benchmark for studies based on manipulations of activity of specific populations of neurons to reveal their roles in control of specific aspects of locomotion.

body configuration; intralimb coordination; kinematics; locomotor gait; mouse

INTRODUCTION

Locomotion is a motor function essential for animals and humans to interact with the environment. Although forward (FW) locomotion is the main form of progression for a majority of living beings, both humans and animals are capable of locomotion in other directions (backward, sideways) as well as stepping in place (Ashley-Ross and Lauder 1997; Axon et al. 1987; Buford et al. 1990; Eilam and Shefer 1992; Musienko et al. 2012; Thorstensson 1986; Vilensky and Cook 2000).

Describing the kinematics of locomotor movements is an important step in the analysis of the locomotor system, as it characterizes different aspects of locomotor movements and allows the correlation of these aspects with neuronal activity as well as with corresponding aspects of locomotor movements observed in patients with locomotor disorders, mutants, etc. Although the kinematics of FW locomotor movements have been studied in considerable detail in both humans and terrestrial quadrupeds (Barrey et al. 1993; Bellardita and Kiehn 2015; Carlson-Kuhta et al. 1998; Gambaryan 1974; Herbin et al. 2007; Leblond et al. 2003; Miller et al. 1975; Nilsson et al. 1985; Riley et al. 2007; Smith et al. 1998; Trank et al. 1996), backward (BW) locomotor movements have received relatively less attention (Ashley-Ross and Lauder 1997; Axon et al. 1987; Buford et al. 1990; Eilam et al. 1995; Grasso et al. 1998; Hoogkamer et al. 2014; Lee et al. 2013; Perell et al. 1993; Thorstensson 1986; Zelik et al. 2014). In terrestrial quadrupeds, comparisons of kinematics of FW and BW locomotor movements have been performed in cats, mole rats, and salamanders (Ashley-Ross and Lauder 1997; Buford et al. 1990; Eilam and Shefer 1992). These studies have demonstrated that the locomotor cycle is shorter and the speed of progression is lower during BW locomotion than during FW locomotion; that with an increase in locomotor speed changes in structure of the locomotor cycle during FW and BW locomotion are similar; and that interlimb coordination during BW locomotion corresponds to walking with a lateral sequence. It was also found that cats walk forward and backward using different body configurations, resulting in different intralimb coordination during these two forms of locomotion.

Mice are particularly useful for studying the locomotor system because they allow the use of rapidly developing and highly promising genetic, chemogenetic, and optogenetic methods. In these investigations, different setups (such as a treadmill, an air-ball, a solid immovable surface) providing different environmental conditions are used (Bellardita and Kiehn 2015; Herbin et al. 2007; Leblond et al. 2003). Thus, a treadmill determines the locomotor speed of the animal, and an air-ball provides an unstable support surface; in both situations, the animal locomotes in static surroundings. The differences in the sensorimotor and exteroceptive information during the

treadmill and air-ball locomotion compared with those during overground locomotion could lead to differences in some aspects of locomotor movements. However, an earlier comparison of FW treadmill and overground locomotion in mice did not reveal any clear differences (Herbin et al. 2007). Comparisons of kinematics of FW and BW locomotor movements in each of three setups (a treadmill, an air-ball, a flat immovable surface) have never been performed.

The aim of the present study was to compare the kinematics of FW and BW locomotion performed by mice in different environmental conditions. For this purpose, mice performing FW and BW locomotion in a tunnel, on a treadmill, and on an air-ball were video recorded and the hindlimb kinematics were analyzed. In particular, we studied the basic parameters of locomotion (speed, step amplitude, and cycle structure), interlimb and intralimb coordination, and the relationship between body configuration and intralimb coordination.

A brief account of this study has been published in abstract form (Vemula et al. 2016).

MATERIALS AND METHODS

Animals

Experiments were performed on 21 adult male and female wild-type (C57) mice (20–30 g, 5–10 wk old). They were housed in standard cages with food and water ad libitum on a 12:12-h light-dark cycle. All experiments were conducted with approval of the local ethical committee (Norra Djurförsöksetiska Nämden) in Stockholm and followed the European Community Council Directive (2010/63EU) and the guidelines of the National Institutes of Health *Guide for the Care and Use of Laboratory Animals*.

Surgical Procedures

All surgical procedures were performed under general anesthesia and aseptic conditions. General anesthesia consisted of ketamine (75 mg/kg) in combination with medetomidine (1 mg/kg) administered intraperitoneally.

Eight animals were subjected to implantation of a head plate, which was used for head fixation during locomotion performed on an air-ball. The head fixation was done to prevent the animal from falling off the ball. A custom-fabricated aluminum head plate (1 cm × 0.4 cm, ~1 g) was cemented to the skull with dental adhesive (OptiBond, Kerr, CA).

For analgesia, buprenorphine (0.1 mg/kg) was administered subcutaneously postoperatively twice daily for 2 days to all animals subjected to surgery. Experiments started 1–2 days after the surgery.

Experimental Design

Locomotion in the tunnel. The tunnel setup (Fig. 1, *A* and *B*) was made from transparent Plexiglas and imitated the narrow tunnels of the burrows in which mice live in nature. It consisted of a narrow tunnel (length 50 cm, height 4 cm, width 2.5–3.5 cm) with a small box (7 × 7 × 4 cm each) at each end of the tunnel (one of these boxes is shown in Fig. 1*B*). Each box had a removable top and a door that closed the entrance to the tunnel. One of the side walls of the tunnel was movable so that the width of the tunnel could be adjusted to the size of a mouse in such a way that the animal could easily walk in the tunnel straight FW or straight BW but could not turn around. The animal was placed in the entrance box through the removable top, the top was closed, and then the door to the tunnel was opened and the animal performed FW locomotion. When the mouse reached the end of the tunnel, it performed BW locomotion until it reached the door closing the tunnel entrance.

Usually the animal spontaneously exhibited three or four sequential episodes of FW and BW locomotion. Each episode of FW and BW walking in the tunnel consisted of four to six and four to nine steps, respectively.

Locomotion on the treadmill. To evoke locomotion straight FW or straight BW, a mouse was placed on the treadmill with a transparent belt in a corridor formed by two transparent Plexiglas plates. The mice did not require specific training to perform FW locomotion on the treadmill belt moving backward in relation to the animal. In contrast, reversing the direction of the treadmill belt evoked a few episodes of BW locomotion in only two of eight mice. The speed of the treadmill belt was adjusted so that an animal performed steady locomotion, staying close to the middle of the belt. The locomotor speed was thus chosen by the animal, and it varied for different episodes in individual animals (Table 1).

Locomotion on the air-ball. One to two days after implantation of the head plate, the mice were trained for 2–3 days to accustom them to head fixation on the floating air-ball (one 20-min session per day) before they were used in experiments. An animal was placed on the fixed air-ball. The height of the head fixator was adjusted to the size of each individual animal so that the animal stood on the air-ball with a normal body configuration. To encourage an animal with a fixed head to perform locomotion straight FW or straight BW on the floating air-ball, two transparent plastic plates were positioned on the left and right sides of the animal, forming a corridor (Fig. 1*D*) with a width of 4–6 cm. In such an environment, the mouse spontaneously performed locomotion straight FW without touching the walls of the corridor. BW locomotion either was performed spontaneously or was evoked by light tactile stimulation of the snout.

To characterize kinematics of locomotor movements, video recording (100 frames/s) was performed. During walking in the tunnel (Fig. 1, *A* and *B*) and on the treadmill (Fig. 1*C*) we simultaneously video recorded the side view and the bottom view (obtained with a mirror tilted by 45° placed under the tunnel and the treadmill) of animals, whereas only the side view was recorded for animals performing locomotion on the air-ball (Fig. 1*D*).

The animal's hindlimbs and trunk were shaved, and markers were drawn on the skin above the metatarsophalangeal (MTP) and ankle joints as well as on the rostral and caudal ends of the pelvis. The video camera was positioned at a distance of ~2 m from the mouse.

Recording and Data Analysis

The video recordings were analyzed frame by frame to calculate the parameters of the locomotor movements exhibited by animals on different setups. To determine interlimb coordination during FW and BW locomotion, as well as the structure of locomotor cycles, the moments of paw liftoff and touchdown were determined. The period between these moments was considered the swing phase and the rest of the cycle the stance phase.

For analysis of the phases, one hindlimb was selected as a reference limb, its step cycle was normalized to 1.0 (the onset of swing was taken as the cycle onset), and the phases of paw-off and paw-on moments of different limbs were calculated in the reference cycle. The difference between the swing onset phases of two limbs was called the "phase difference between limbs."

The phase difference within a pair of limbs (right and left limbs of the same girdle or homolateral fore- and hindlimbs) varied within a locomotor episode. The average and SD for each episode were calculated with circular statistics methods (Batschelet 1981). The averages of these SDs observed during FW locomotion and BW locomotion were compared.

The step cycle structure (i.e., the duration of the swing and stance phases) can vary within one episode of locomotion, between different episodes of the same behavior (e.g., different episodes of BW locomotion on the air-ball) performed by the same animal, between tests on different setups (e.g., FW tunnel locomotion and FW treadmill
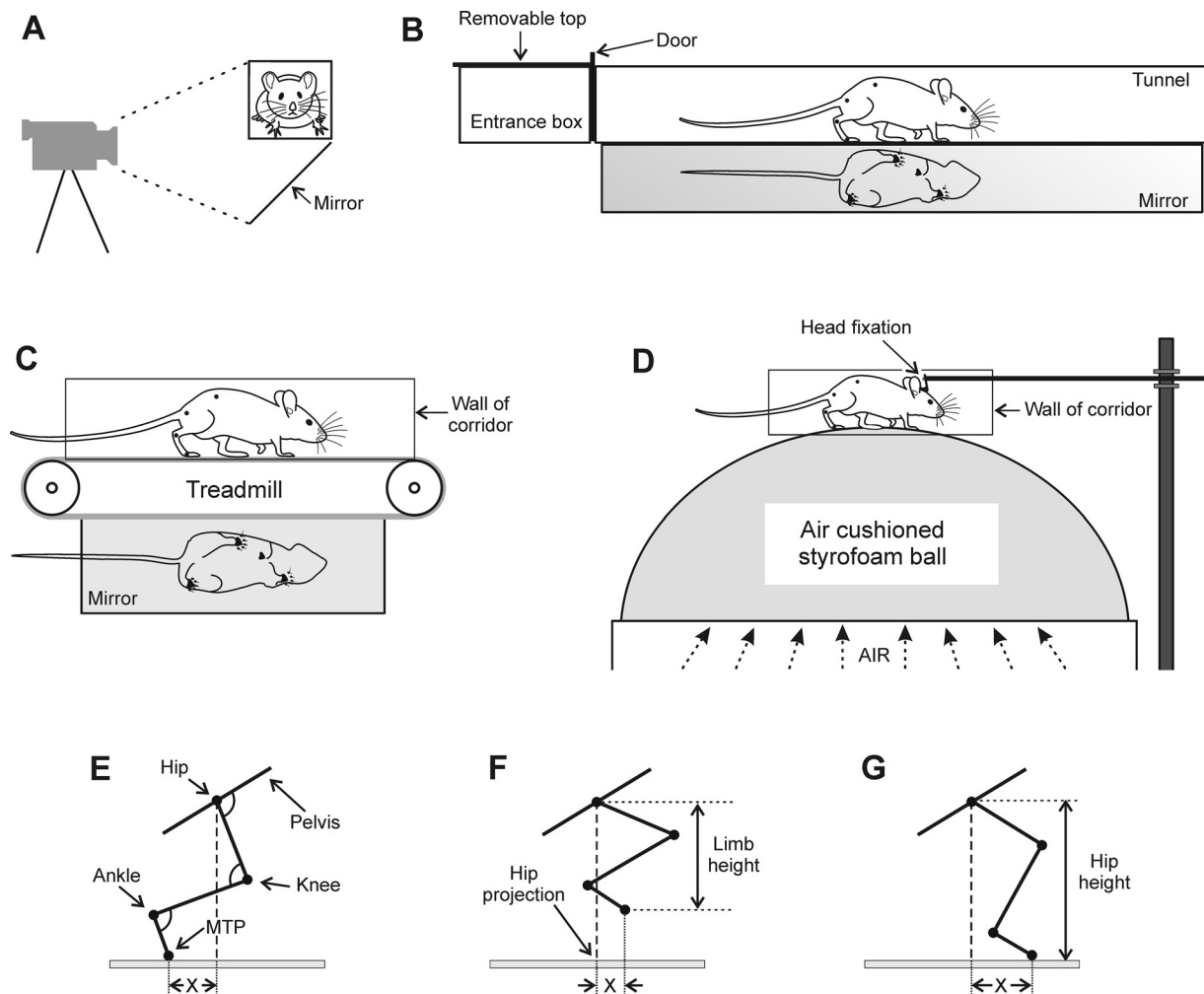
Fig. 1. Experimental designs. Mice performed forward (FW) and backward (BW) locomotion in a narrow tunnel (*A* and *B*), on a treadmill (*C*), and on a floating air-ball (*D*). Video recording (100 frames/s) from below (by means of a mirror) along with the side view of animals walking in the tunnel (*A* and *B*) and on the treadmill (*C*) was performed. Only the side view of the animals walking on the air-ball (*D*) was recorded. *A* and *B*: front view (*A*) and side view (*B*) of the tunnel setup. *C*: side view of the treadmill setup. The mouse performed locomotion on a transparent treadmill belt in a corridor between 2 transparent walls. *D*: side view of the air-ball setup. The mouse performed locomotion on a floating air-ball in a corridor between 2 transparent walls with its head fixed. *E–G*: configuration of the right hindlimb (lateral view) at the extreme caudal position at stance offset (*E*), in the middle of swing (*F*), and at the extreme rostral position at stance onset (*G*). The vertical dashed line indicates projection of the hip to the surface. Limb height (*F*), hip height (*G*), and the rostro-caudal position of the paw [metatarsophalangeal (MTP) joint] in relation to the hip (*X* in *E–G*) were measured.

locomotion) performed by the same animal, and for the same behavior performed by different animals. This is why, in our analysis of the intralimb coordination, we normalized the swing and stance phases separately. In each locomotor cycle, the swing phase was normalized to 0.3 parts of the cycle for FW locomotion and to 0.2 for BW, which represent the mean values of swing proportions observed, respectively, in FW and BW locomotor cycles in all mice. Correspondingly, the stance phase was normalized to 0.7 and 0.8 parts of the cycle. Such normalization allowed us to average profiles of the angle change at a particular joint during a definite phase of the cycle as well as to compare the averaged profiles during the same phase of the locomotor cycle in different episodes.

To assess intralimb coordination, the ankle, knee, and hip joint angles were determined. Because of substantial skin slippage above the hip and knee joints observed during walking, markers on the skin above these joints did not allow us to determine their exact positions at a definite moment of the locomotor cycle. To overcome these difficulties, the lengths of the femur and tibia of each individual mouse were measured postmortem, as was the position of the hip joint in relation to the rostro-caudal extent of the pelvis. With these measurements and markers on the rostral and caudal edges of the

pelvis from the side view, the position of the hip joint was determined. The position of the knee joint was triangulated from the positions of the hip and ankle joints with the measurements of the femur and tibia. The hip, knee, and ankle joint angles were calculated with the positions of the pelvis, hip, knee, ankle, and MTP joints.

To find out whether the caudal part of the trunk was maintained at a constant height during locomotion, hip height (Fig. 1*G*) was measured at different moments of the locomotor cycle. To qualitatively characterize the limb orientation in relation to the vertical during locomotor movements, at different moments of the locomotor cycle we measured the rostro-caudal paw position *X*, defined as the distance between the projections to the support surface of the MTP joint (the distal end of the limb) and the hip joint (Fig. 1, *E* and *F*). The hip projection to the surface was taken as "0," and thus positive and negative values of *X*, respectively, indicated the rostral and caudal positions of the paw in relation to the hip and therefore the rostral and caudal orientation of the hindlimb in relation to the vertical. The rostro-caudal orientation of the entire stride was characterized by the middle of the rostro-caudal paw excursions during stance (termed the "midstance position"). Then, to estimate the relationship between the limb orientation during the entire step cycle and hip height, the midstance position was

Table 1. *Speed of FW and BW locomotion on different setups*

	FW Locomotion			BW Locomotion		
	Tunnel	Treadmill	Air-ball	Tunnel	Treadmill	Air-ball
Speed, cm/s	23 ± 10 [7;45]	16 ± 4 [7;23]	30 ± 17 [6;57]	11 ± 5 [4;22]	7 ± 4 [1;17]	21 ± 13 [3;47]
Step amplitude, cm	5.9 ± 1.4 [2.7;8.1]	3.4 ± 0.6 [2.3;4.7]	4.4 ± 0.9 [2.8;6.4]	2.1 ± 0.6 [1.3;3.3]	1.6 ± 0.3 [0.7;2.1]	2.3 ± 0.6 [1.4;3.5]
N	5	3	2	3	2	2
n_{ep}	9	9	6	5	4	3
n_{cyc}	36	67	35	28	28	27

Mean ± SD values and range ([min;max]) of the speed during forward (FW) and backward (BW) locomotion in the tunnel, on the treadmill and on the air-ball. N, number of animals; n_{cyc}, number of locomotor cycles; n_{ep}, number of episodes. Significance of difference between the mean values of speed during FW and BW locomotion observed in the tunnel, on the treadmill, and on the air-ball (2-tailed t test with unequal variance): $P = 2 \times 10^{-6}$, 6×10^{-16}, and 3×10^{-2}, respectively. Significance of difference between the mean values of FW as well as BW locomotor speed observed on different setups (tunnel vs. treadmill, tunnel vs. air-ball, treadmill vs. air-ball): $P = 2 \times 10^{-6}$, 4×10^{-2}, 2×10^{-9}, respectively, for FW and $P = 5 \times 10^{-4}$, 4×10^{-4}, and 6×10^{-7}, respectively, for BW locomotion. Significance of difference between the mean values of FW and BW step amplitude observed in the tunnel, on the treadmill, and on the air-ball: $P = 4 \times 10^{-17}$, 3×10^{-26}, and 4×10^{-15}, respectively. Significance of difference between the mean values of FW as well as BW step amplitude observed on different setups (tunnel vs. treadmill, tunnel vs. air-ball, treadmill vs. air-ball): $P = 1 \times 10^{-18}$, 3×10^{-4}, and 4×10^{-15}, respectively, for FW and $P = 4 \times 10^{-4}$, 0.11, and 1×10^{-7}, respectively, for BW locomotion.

correlated with hip height (Fig. 6). To estimate changes of the functional length of the hindlimb, limb height (the distance between horizontal lines drawn through the hip and the paw; Fig. 1*F*) was measured at different moments of the locomotor cycle. To compare data obtained in mice of different sizes, the values of hip height, limb height, and the rostro-caudal paw position were normalized to the maximal limb length (that is, the sum of the femur, tibia, and foot lengths measured postmortem).

Statistical Analysis

All quantitative data in this study are presented as means ± SD. The phase difference SD was averaged across episodes. In all other cases, the average was calculated across all cycles pooled from all episodes recorded in all animals. Student's *t* test (2 tailed) was used to characterize statistical significance when comparing different means; the significance level was set at $P = 0.05$. Linear regression was used to test possible linear relationships between parameters (Fig. 3, Fig. 6); significant differences of the regression slope from 0 were tested with the *t* test.

RESULTS

Basic Parameters of FW and BW Locomotion Performed on Different Setups

First, we compared locomotor speed and step amplitude during locomotion performed in a particular direction on different setups, as well as during FW and BW locomotion performed on each of the three setups. We did not train animals to perform FW and BW locomotion with a definite speed on a particular setup. Thus, each animal chose a comfortable locomotor speed on each of three setups. As shown in Table 1, the speed of FW and BW locomotion in the tunnel, on the treadmill, and on the air-ball varied in a wide range, and these ranges overlapped considerably in all six conditions. On average, during locomotion in a particular direction the lowest speed was observed on the treadmill and the highest on the air-ball. The speed of FW as well as BW locomotion performed on different setups correlated positively with the step amplitude (that is, the distance between the extreme rostral and extreme caudal positions of the distal point of the paw during a cycle). On each of the three setups, both the mean values of the step amplitude and locomotor speed during BW locomotion were significantly (approximately twice) smaller than those observed during FW locomotion (Table 1).

Second, we compared the structure of locomotor cycles during locomotion performed in a particular direction on different setups, as well as during FW and BW locomotion performed on each of three setups. We found that ranges for cycle durations during FW and BW locomotion performed on different setups strongly overlapped. On average, cycle durations as well as the durations of the stance and swing phases were similar (no significant difference was observed) during locomotion performed in a particular direction on different setups (Fig. 2). However, on each of the three setups, the mean value of swing duration during BW locomotion was significantly (almost twice) smaller than that observed during FW locomotion (Fig. 2*A*). In addition, both stance and cycle durations during BW locomotion on the treadmill and cycle duration during BW locomotion on the air-ball were significantly shorter than those measured during FW locomotion performed on the corresponding setups (Fig. 2, *B* and *C*).

Finally, to reveal changes in the structure of the locomotor cycle with an increase in cycle duration during FW and BW locomotion performed on different setups, we plotted swing duration and stance duration versus cycle duration (Fig. 3). On each of the three setups, a strong (statistically significant) positive dependence of stance duration on cycle duration was revealed during both FW and BW locomotion. By contrast, the dependence of swing duration on cycle duration was much weaker or absent (statistically insignificant) during BW locomotion and FW locomotion on any of the three setups. Thus, during both FW and BW locomotion performed on each of the three setups, with an increase in cycle duration stance duration increased, whereas swing duration remained relatively constant.

Interlimb Coordination During FW and BW Locomotion

Next, we analyzed interlimb coordination during FW and BW locomotion performed in the tunnel, as this setup allowed clear visualization of all four limbs during the whole locomotor episode. Examples illustrating the different types of interlimb coordination during FW and BW locomotion are shown in Fig. 4*A* and Fig. 4, *B–D*, respectively. To build these diagrams, the left hindlimb was selected as a reference limb and the phases of onset and offset of each limb stance were normalized to the reference limb cycle. The normalized phases of stance onsets
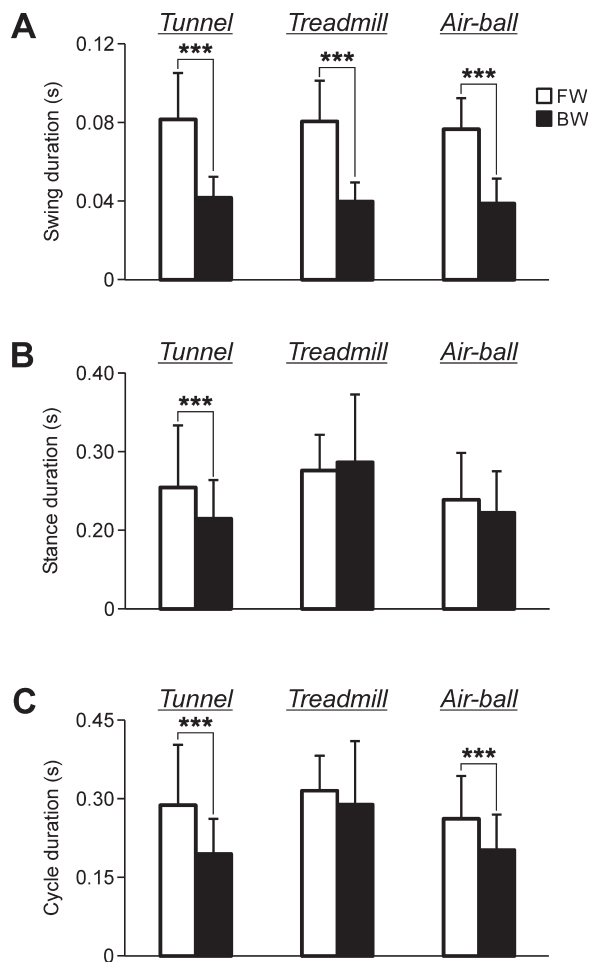
Fig. 2. Characteristics of forward (FW) and backward (BW) locomotor cycles on different setups: mean (±SD) values of swing (*A*), stance (*B*) and cycle (*C*) durations during FW and BW locomotion on different setups. In *A*, *B*, and *C*, respectively: during FW locomotion number of animals $N = 6, 6, 5$, number of cycles $n = 60, 120, 94$; during BW locomotion, $N = 6, 2, 5$, $n = 90, 41, 49$. ***Statistically significant differences between mean values of a specific parameter obtained during FW and BW locomotion performed on a particular setup ($P < 0.001$).

and offsets in individual cycles were then averaged, and the sequence of locomotor movements of all four limbs was drawn.

In all episodes of FW and BW locomotion, swing duration was shorter than stance duration (Fig. 2), and the phase difference between the right and left limbs was close to 0.5 of the cycle. Taken together, these two criteria indicate that in our experiments the animals always exhibited walk or trot (Hildebrand 1980; Lemieux et al. 2016).

During FW locomotion in the tunnel, only one type of sequence of limb movements was observed. As shown in Fig. 4*A*, the mice moved their limbs in the following sequence: left hindlimb (HL), left forelimb (FL), right hindlimb (HR), right forelimb (FR). Such coordination corresponds to walk with a lateral sequence (Gambaryan 1974).

During BW locomotion three different locomotor gaits were observed. First, the walk with lateral sequence (Fig. 4*B*), that is, the same coordination that was observed during FW locomotion, was revealed. Second, the walk with diagonal sequence (Fig. 4*C*; Gambaryan 1974), which was characterized by a reverse sequence of limb movements (HL, FR, HR, FL)

compared with that observed during walk with a lateral sequence (HL, FL, HR, FR; Fig. 4*B*), was found. The last observed gait was the trot (Fig. 4*D*), which was characterized by simultaneous movements of a diagonal pair of limbs (HL and FR) alternating with simultaneous movements of the other pair (HR and FL). The same mouse could perform BW locomotion with different interlimb coordination and sometimes even switch from one coordination to another within one locomotor episode. We observed no dependence of the gait on the BW locomotion velocity.

We found that the mean SD values of the phase difference between movements of the reference hindlimb and contralateral hindlimb during FW locomotion and BW locomotion performed with different gaits were almost the same [0.089 ± 0.012 and 0.096 ± 0.016, respectively; number of animals (N) = 4, number of episodes (n) = 15 and $N = 6$, $n = 14$, respectively; no significant difference: $P = 0.72$, t test], suggesting that the strength of coupling between hindlimb movements was similar during FW and BW locomotion. By contrast, the mean SD value of the phase difference between movements of the reference hindlimb and ipsilateral forelimb during FW locomotion was significantly smaller than that during BW locomotion (0.064 ± 0.012 and 0.109 ± 0.012, respectively; $N = 4$ and 6, respectively; $n = 15$ and 14, respectively; $P = 0.003$, t test), suggesting that the strength of coupling between the pelvic and shoulder girdles was stronger during FW than during BW locomotion.

Body Configurations During FW and BW Locomotion on Different Setups

Mice can perform locomotion with different body configurations, i.e., with different positions of rostro-caudal paw displacement during stepping in relation to the trunk, as well as with different heights of the hindquarters. To determine whether the height of the hindquarters (characterized by the hip height) was maintained constant during locomotor episodes, we plotted hip height versus the rostro-caudal position of the paw (characterized by the distance between MTP and the projection of the hip to the surface; Fig. 1, *E–G*; see MATERIALS AND METHODS for details) for individual locomotor episodes. Such a plot for the FW locomotor episode performed on the treadmill is presented in Fig. 5. One can see that the vertical oscillations of the hip during sequential locomotor cycles were rather small and thus hip height remained almost constant during the locomotor episode (Fig. 5*A*). Similarly, maintenance of the hip at the same height throughout the locomotor episode was observed in all animals during both FW and BW locomotion performed on all setups [number of animals (N) = 6, number of episodes (n) = 25 and $N = 6$, $n = 23$, respectively].

To check whether the position of the rostro-caudal paw displacement during stepping in relation to the hip depended on the hip height, we plotted the limb height versus the rostro-caudal position of the paw, as during stance the limb height is equal to the hip height (Fig. 1, *F* and *G*; see MATERIALS AND METHODS for details). Such a plot (for the same episode as in Fig. 5*A*) is shown in Fig. 5*B*. One can see that the limb height was almost constant (and equal to hip height) during stance, whereas during swing it decreased from swing onset to midswing and then increased again from midswing to contact. In this locomotor episode, the hip was kept rather high above the
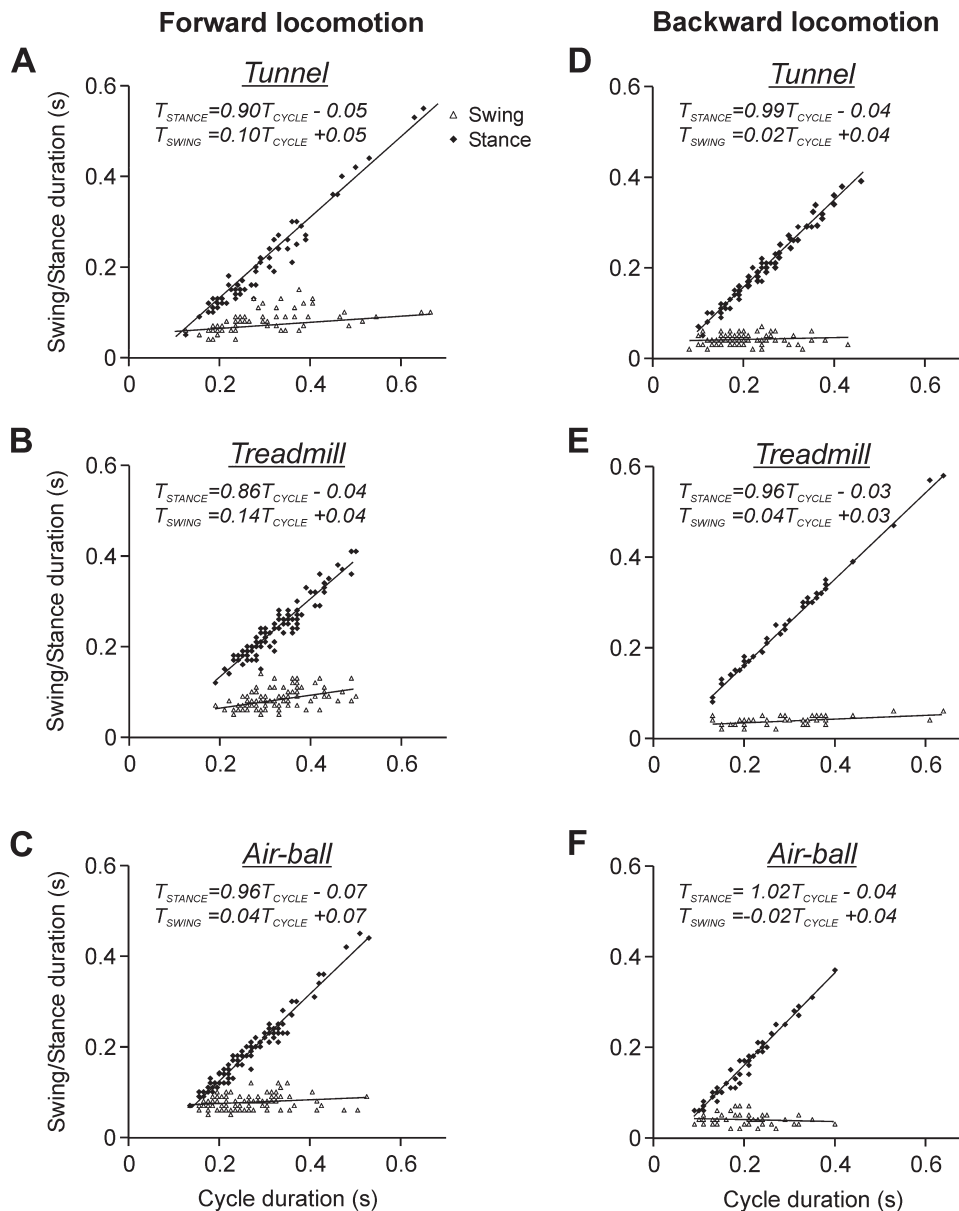
Forward locomotion

Backward locomotion

A *Tunnel*

$T_{STANCE}=0.90T_{CYCLE} - 0.05$
$T_{SWING} =0.10T_{CYCLE} +0.05$

△ Swing
◆ Stance

B *Treadmill*

$T_{STANCE}=0.86T_{CYCLE} - 0.04$
$T_{SWING} =0.14T_{CYCLE} +0.04$

C *Air-ball*

$T_{STANCE}=0.96T_{CYCLE} - 0.07$
$T_{SWING} =0.04T_{CYCLE} +0.07$

D *Tunnel*

$T_{STANCE}=0.99T_{CYCLE} - 0.04$
$T_{SWING} =0.02T_{CYCLE} +0.04$

E *Treadmill*

$T_{STANCE}=0.96T_{CYCLE} - 0.03$
$T_{SWING} =0.04T_{CYCLE} +0.03$

F *Air-ball*

$T_{STANCE}= 1.02T_{CYCLE} - 0.04$
$T_{SWING} =-0.02T_{CYCLE} +0.04$

Fig. 3. Characteristics of forward (FW) and backward (BW) locomotor cycles observed on different setups: swing duration and stance duration plotted vs. cycle duration during FW (*A–C*) and BW (*D–F*) locomotion performed in the tunnel (*A* and *D*), on the treadmill (*B* and *E*), and on the air-ball (*C* and *F*). Note positive dependence of stance duration T_{STANCE} on cycle duration T_{CYCLE} as well as weak or absent dependence of swing duration T_{SWING} on cycle duration T_{CYCLE} during both FW and BW locomotion performed on any of the 3 setups (formulas for the linear regression are presented on each panel). In *A, B, C, D, E, F*, respectively: number of animals $N = 6, 6, 5, 6, 2, 5$, number of cycles $n = 60, 120, 94, 90, 41, 49$, coefficient of determination for swings $R^2 = 0.24, 0.19, 0.04, 0.01, 0.26, 0.01$ and for stances $R^2 = 0.96, 0.90, 0.96, 0.97, 0.99, 0.99$, P value for linear regression slope for swing durations $P = 8 \times 10^{-5}, 5 \times 10^{-7}, 0.04, 0.36, 6 \times 10^{-4}, 0.43$ and for stance durations $P = 4 \times 10^{-42}, 5 \times 10^{-60}, 3 \times 10^{-65}, 5 \times 10^{-69}, 2 \times 10^{-45}, 2 \times 10^{-36}$.

ground (the limb height during stance was ~0.6) and the rostro-caudal excursions of the paw were slightly displaced caudally in relation to the hip projection to the surface. Figure 5, *C–F*, show such graphs for four other episodes of FW locomotion performed with different hip heights. One can see that when locomotion was performed with a high hip height (*episode C*) the rostro-caudal excursions of the paw were almost symmetric in relation to the hip (we considered the excursions perfectly "symmetric" if the middle of the excursion range was exactly under the hip). In the episode in which the hip was maintained at lower height (*episode D*) similar to that in the episode shown in Fig. 5*B*, the trajectories were slightly displaced rostrally, whereas in *episode B* they were displaced caudally. Finally, during episodes with the lowest hip height, the posterior (*episode E*) and anterior (*episode F*) biases in the rostro-caudal paw excursions were substantial. Thus, in these episodes, with a decrease in the hip height a gradual displacement of the rostro-caudal paw excursions from the middle to either anterior or posterior position in relation to the hip was

observed. It should be noted that the amplitude of the rostro-caudal paw movements was similar in all of these episodes of FW locomotion.

Figure 5, *G* and *H*, show the same type of plots for two episodes of BW locomotion performed with different hip heights. In both episodes, almost all paw trajectories were located rostrally to the hip. However, in the episode with higher (~0.5) hip height (*episode G*), the trajectories were closer to the hip compared with those in the episode with lower (~0.4) hip height (*episode H*). It should be noted that the amplitude of the rostro-caudal paw excursion during BW locomotion was approximately half that observed during FW locomotion (compare Fig. 5, *G* and *H*, to Fig. 5, *B–F*). In addition, rather often in some cycles of BW locomotion limb height did not decrease during swing (the paw was dragged along the surface), and thus the paw trajectories during stance and swing coincided (Fig. 5*H*).

To reveal the relationship between hip height and the position of the rostro-caudal paw excursions during FW and BW
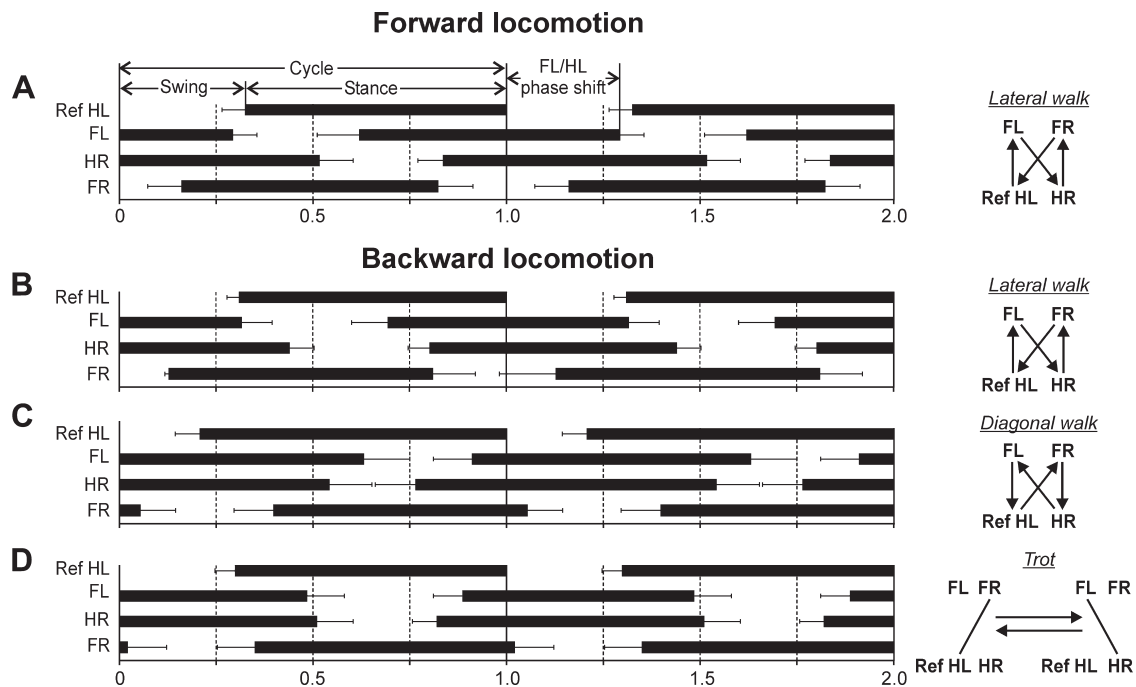
Fig. 4. Interlimb coordination during forward (FW) and backward (BW) locomotion in the tunnel. *A*: pattern of interlimb coordination ("walk with lateral sequence") observed during FW locomotion. *B–D*: patterns of interlimb coordination ["walk with lateral sequence" (*B*), "walk with diagonal sequence" (*C*), and "trot" (*D*)] observed during BW locomotion. Horizontal bars represent average stance phases of individual limbs (±SDs of stance onset and offset) normalized to the cycle of the reference left hindlimb (Ref HL). Two cycles are shown. *A–D*, *right*, represent diagrams of step sequences. The cycle, swing, stance, and phase difference between the left forelimb (FL) and the left hindlimb (reference limb) are indicated in *A*. HR, right hindlimb; FR, right forelimb.

locomotion performed on different setups, for episodes of locomotion performed in the same direction by all animals tested on a particular setup, we plotted hip height against the middle of the rostro-caudal paw excursions (midstance position) in individual cycles (Fig. 6). As can be seen in Fig. 6, *A–C*, on each of the three setups the mice could perform FW locomotion with a different hip height and on each of three setups we observed a correlation between hip height and midstance position. However, the preferred body configuration during FW locomotion differed on different setups. During locomotion in the tunnel (Fig. 6*A*), about half of the midstance positions had positive values and half had negative values, suggesting that on this setup the mice performed stepping both rostrally and caudally in relation to the hip. By contrast, locomotion on the treadmill (Fig. 6*C*) was performed with anterior stepping (the majority of midstance positions had positive values), whereas on the air-ball (Fig. 6*B*) it was performed with posterior stepping (the majority of midstance positions had negative values).

In episodes of BW locomotion performed by mice in the tunnel and on the air-ball (Fig. 6, *D* and *E*), the range of hip heights was similar to that observed in episodes of FW locomotion. We found some correlation between hip height and the midstance position for BW locomotion on the air-ball and a weaker correlation in the tunnel. In contrast to FW locomotion (Fig. 6, *D* and *E*), mice always performed BW locomotion with anterior stepping (all midstance values were positive).

To conclude, we found that at low hip height FW locomotion was performed with either anterior or posterior (in relation to the hip) steps depending on setup, whereas BW locomotion was always performed with anterior steps regardless of setup. With an increase in hip height, stepping movements became progressively more symmetric in relation to the hip during both FW and BW locomotion.

Intralimb Coordination During FW and BW Locomotion Performed with Different Body Configurations

To compare intralimb coordination during FW and BW locomotion performed with different body configurations, in corresponding locomotor episodes motions at different joints were analyzed.

Figure 7, *D–F*, show the average profiles of angle changes at the hip, knee, and ankle joints (Fig. 7, *D*, *E*, and *F*, respectively) in three representative episodes of FW locomotion performed with different body configurations shown in Fig. 7, *A–C*. In two episodes, the hip was maintained at a low height and the steps were anterior ("anterior stepping" in Fig. 7*A*) or posterior ("posterior stepping" in Fig. 7*C*). In contrast, in the episode shown in Fig. 7*B* the hip was maintained at a high height and the steps were symmetric in relation to the hip ("middle stepping"). Motions at the hip joint were similar in all three episodes and characterized by flexion and extension during swing and stance, respectively (Fig. 7*D*). There was also no substantial difference in the amplitude of the hip motions. However, the range of angle changes during the locomotor cycle was different in each of the three episodes: the mean values of the hip joint angle during anterior, middle, and posterior stepping were 52° (flexed hip), 78° (slightly flexed), and 108° (slightly extended), respectively.

In contrast to motions at the hip joint, motions at the knee and ankle joints (Fig. 7, *E* and *F*, respectively) strongly depended on the form of stepping. Although during swing the flexion and then extension of these joints was observed (*phases F* and *E1*, as defined by Philippson 1905), the amplitudes of
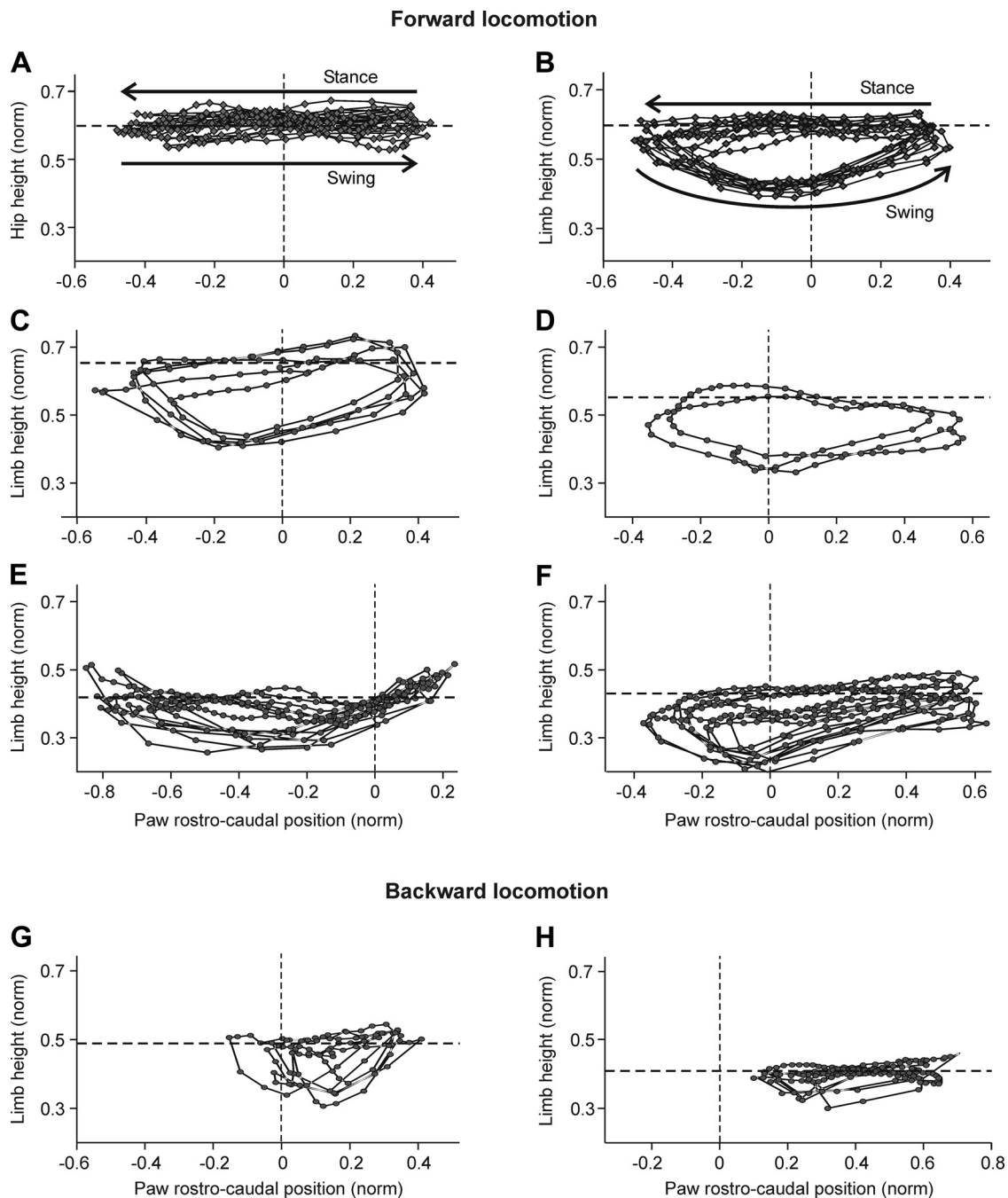
Fig. 5. Examples of forward (FW) and backward (BW) locomotion performed with different body configurations. *A* and *B*: hip height (*A*) and limb height (*B*) at different paw rostro-caudal positions during an episode of FW locomotion on the treadmill. Arrows indicate direction of the limb movement during stance and swing. *C–F*: limb height at different paw rostro-caudal positions during episodes of FW locomotion on the treadmill (*C* and *F*), in the tunnel (*D*) and on the air-ball (*E*). *G* and *H*: limb height at different paw rostro-caudal positions during 2 episodes of BW locomotion in the tunnel. The time interval between sequential points is 10 ms. Hip height, limb height, and paw rostro-caudal position were normalized to the maximal limb length (the position of the hip projection to the surface is taken as zero; marked by the vertical dashed line). The negative and positive values indicate caudal and rostral paw positions, respectively. The horizontal dashed line indicates the mean value of hip height during 1 locomotor episode. Note that during FW locomotion with high hip height (*C*) the rostro-caudal excursions of the limb were almost symmetric in relation to the hip; with lower hip height (*B* and *D*), they were slightly displaced posterior (*B*) or anterior (*D*). Finally, during FW locomotion with the lowest hip height (*E* and *F*), the posterior (*E*) and anterior (*F*) displacement of rostro-caudal paw excursions was substantial. In *A, B, C, D, E, F, G*, and *H*, number of cycles *n* = 9, 9, 5, 2, 8, 9, 6, and 6, respectively. *Episode C* is from the same animal as *episode B*. *Episode G* is from the same animal as *episode F*. *Episodes D*, *E*, and *H* are from 3 other animals.

these motions were different in different episodes. In particular, the *F* flexion was the largest during posterior stepping, smaller during middle stepping, and the smallest during anterior stepping. In contrast, the largest *E1* extension at the knee was observed during anterior stepping, almost twice smaller during middle stepping, and very small during posterior stepping (Fig. 7*E*). The ankle *E1* extension was similar in all episodes, although the tendency to contribute more to the limb extension during anterior stepping compared with posterior stepping was observed for this joint as well (Fig. 7*F*).

Forward locomotion

Fig. 6. Correlation between hip height and the rostro-caudal paw excursions in relation to the trunk in episodes of forward (FW) and backward (BW) locomotion performed on different setups: hip height plotted vs. the midstance (middle of the rostro-caudal excursion of the paw) position in relation to the hip in individual cycles of FW locomotion performed in the tunnel (*A*), on the air-ball (*B*), and on the treadmill (*C*) as well as in individual cycles of BW locomotion performed in the tunnel (*D*) and on the air-ball (*E*). Note that during FW locomotion at low hip height, the midstance positions were posterior (vertical dashed line) in steps performed on the air-ball (*B*), anterior during FW treadmill locomotion (*C*), and either anterior or posterior during FW locomotion in the tunnel (*A*). During BW locomotion in the tunnel (*D*) and on the air-ball (*E*), at any hip heights, the midstance position was displaced anterior. In *A, B, C, D, E*, respectively: number of animals $N = 4, 4, 4, 3, 3$, number of cycles $n = 73, 80, 79, 38, 56$, coefficient of determination $R^2 = 0.42, 0.64, 0.33, 0.29, 0.40$, *P* value for linear regression slope $P = 4 \times 10^{-3}$ (left subpopulation in *A*; because of bimodal distribution of the data cloud, the cloud was first fitted with a 2-segment linear approximation to maximize the coefficient of determination R^2, the *x*-axis of fit maximum separated the cloud into left and right subpopulations, and then the linear regressions were calculated separately for these subpopulations: 5×10^{-7} (right subpopulation in *A*), 7×10^{-19}, 3×10^{-8}, 5×10^{-4}, 2×10^{-7}.

Backward locomotion
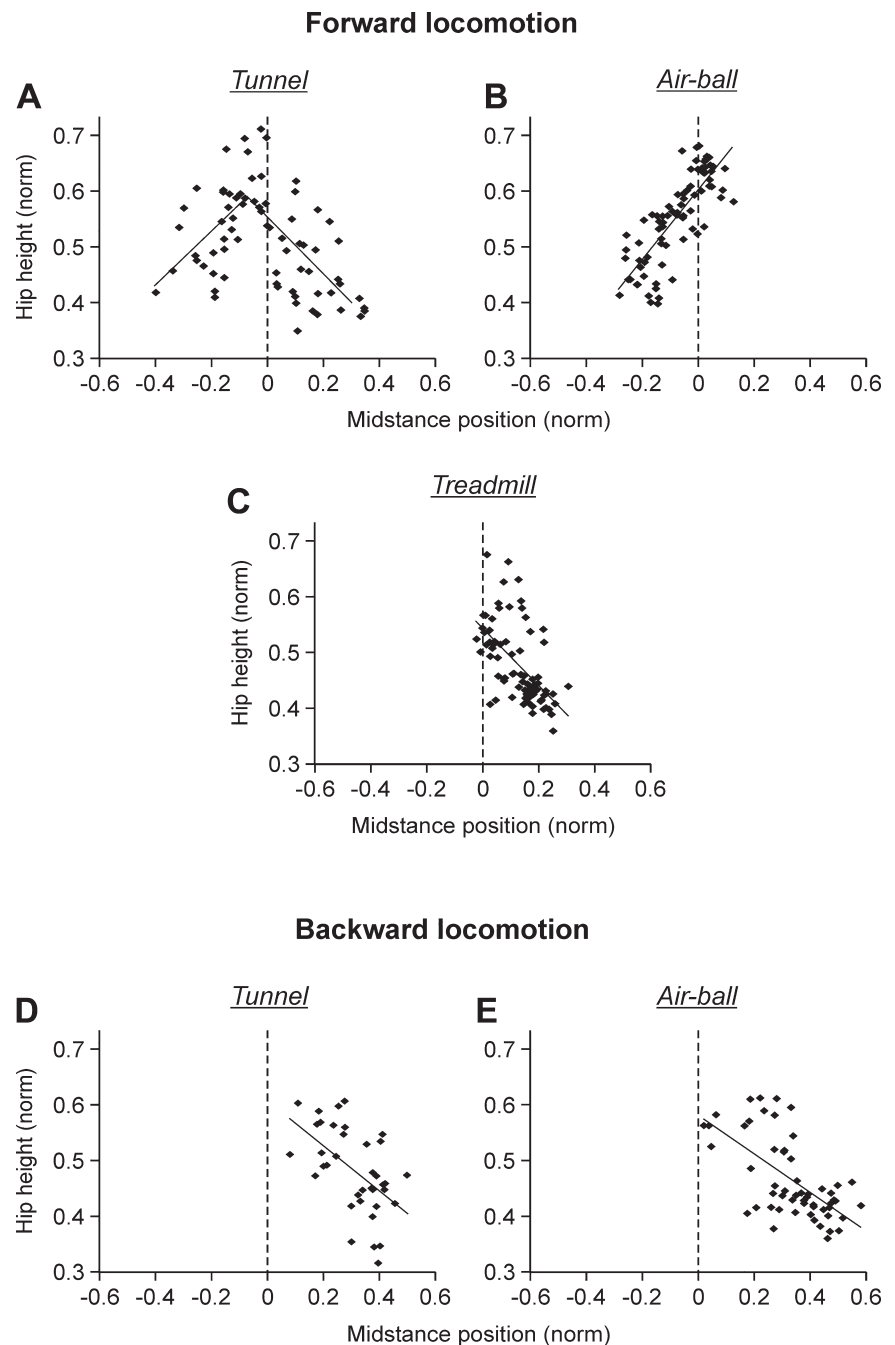
During stance (*phases E2, E3*), motions at the knee and ankle in each episode shared some similarities but were quantitatively and qualitatively different in different episodes (Fig. 7, *E* and *F*). Thus, during posterior stepping, at both the knee and ankle joints, a small *E2* flexion was followed by a large *E3* extension. In contrast, during anterior stepping the ankle *E2* flexion prevailed over the *E3* extension, whereas at the knee only the large *E2* flexion was observed and the *E3* extension was negligibly small. Finally, during middle stepping, a small *E2* flexion and a small *E3* extension were observed at both the knee and the ankle. These observations indicate that the motions at the knee and ankle during FW locomotion are rather complex and strongly depend on the limb orientation.

Figure 7, *G–I*, present three episodes of BW locomotion. Although anterior stepping was observed in all three episodes, in different episodes mice maintained different hip heights (the lowest in *G*, higher in *H*, and the highest in *I*) and the joint trajectories differed substantially. In particular, with an increase in hip height the amplitude of the paw lifting above the surface increased and the rostro-caudal paw position decreased (compare red trajectories in relation to the hip in Fig. 7, *G, H*, and *I*, respectively).

Figure 7, *J–L*, present averaged profiles of the hip, knee, and ankle angles during the episodes shown in Fig. 7, *G–I*. In general, in each of these episodes flexion of all three joints at swing onset contributed to lifting the paw above the surface, extension of the hip during the second half of swing transferred
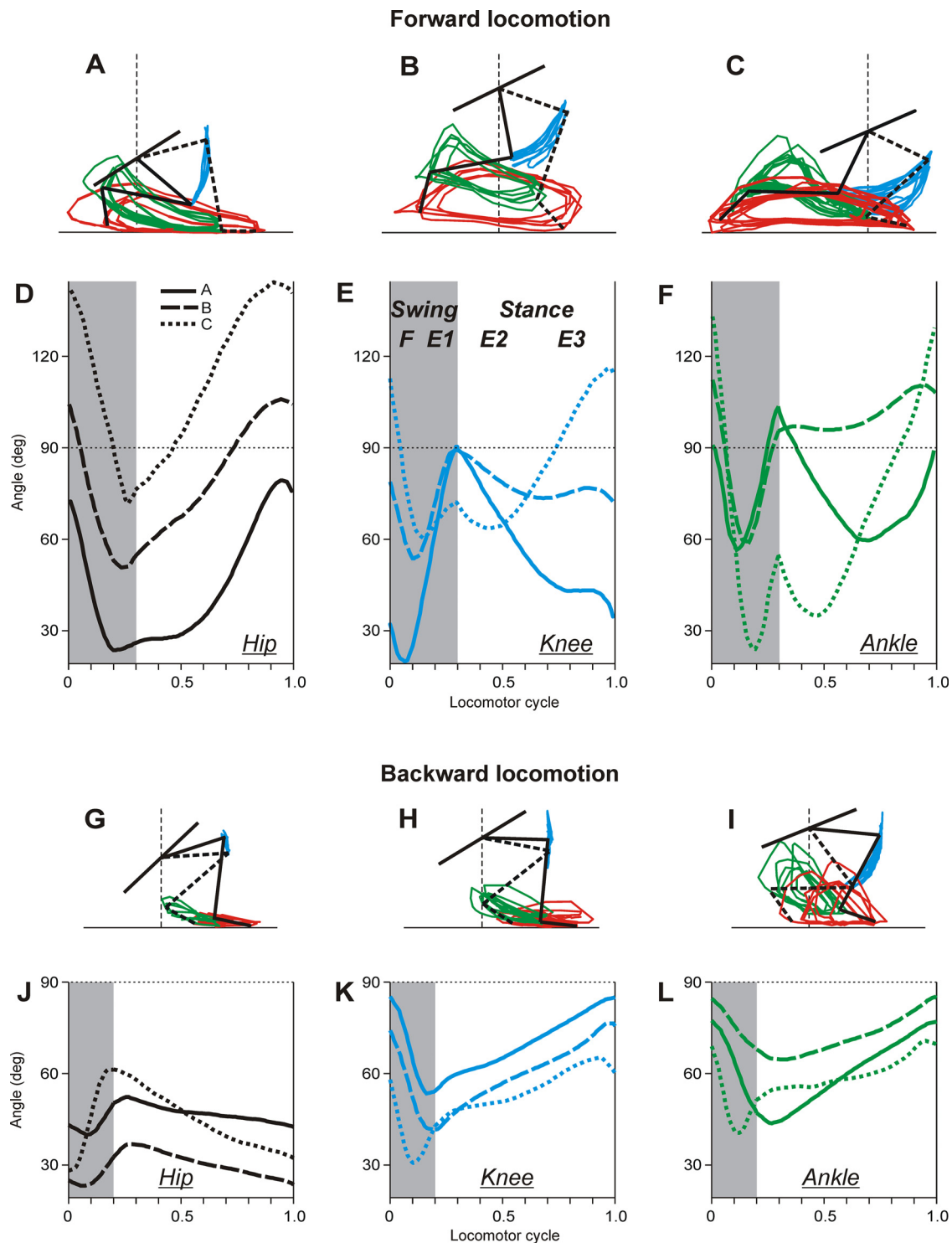
Forward locomotion

Backward locomotion

Fig. 7. Motions at different hindlimb joints during forward (FW) and backward (BW) locomotion performed with different body configurations. *A–C*: side view of knee, ankle and metatarsophalangeal (MTP) trajectories (blue, green and red, respectively) in 3 episodes of FW locomotion performed with different body configurations. The hindlimb configurations at anterior (black dashed lines) and posterior (black solid lines) extreme position during stance as well as orientation of the pelvis maintained during the locomotor episode are shown. Note that in *A* and *C* hip height was low and the MTP trajectories were located anterior and posterior to the hip (indicated by thin dashed lines), respectively, whereas in *B* hip height was high and MTP trajectories were symmetric in relation to the hip. *D–F*: averaged profiles of the angle change at the hip (*D*), knee (*E*), and ankle (*F*) joints during the locomotor cycle in episodes shown in *A*, *B*, and *C*. *G–I*: side view of hip, knee, and MTP trajectories in 3 episodes of BW locomotion performed with different body configurations. Note that although in all 3 episodes MTP trajectories were rostral to the hip, in the episode with the highest hip height (*I*) the trajectory was closer to the hip projection than in *G* and *H*. *J–L*: averaged profiles of the angles at the hip (*J*), knee (*K*), and ankle (*L*) joints during the locomotor cycle in episodes shown in *G*, *H*, and *I*. In *D–F* and *J–L*, swing is indicated by the shaded area. Swing phase is normalized to 30% of the locomotor cycle in *D–F* and to 20% in *J–L*. Swing and stance phase as well as phases *F*, *E1*, *E2*, and *E3* (defined by Philippson 1905) are indicated in *E*. In *A*, *B*, *C*, *G*, *H*, and *I*, number of cycles n = 4, 5, 8, 4, 6 and 5, respectively. *Episodes A, G, H*, and *I* are treadmill locomotion; *episodes B* and *C* are air-ball locomotion. *Episode I* is from the same animal as *episode A*. *Episode H* is from the same animal as *episode G*. *Episodes B* and *C* are from 2 other animals.

the limb to the extreme caudal position, and the simultaneous flexion of the knee and ankle was aimed at keeping the paw above the surface. Finally, during stance flexion of the hip and simultaneous extension of the knee and ankle moved the paw from the extreme caudal to the extreme rostral position. However, the angle profiles at the hip, knee, and ankle in the episode with a high hip height were somewhat different from those in the episodes with a lower hip height. Hip flexion at swing onset was very small; thus the hip joint practically did not contribute to the paw lifting. Furthermore, extension of the knee and ankle started almost in the middle of swing, whereas in two other episodes it started at about the stance offset. Thus, extension of the knee and ankle at the end of swing contributed to limb lowering and landing.

An important characteristic of intralimb coordination is the sequence of angular motion reversals in the flexion-extension sequence during swing and stance at different joints during the locomotor cycle. To compare these sequences during FW and BW locomotion, we measured the time of reversals at a particular joint (extension to flexion for all cases except for flexion to extension for the hip joint during BW locomotion) relative to swing onset as well as the time of reversals in the opposite direction (flexion to extension for all cases except for extension to flexion for the hip during BW locomotion) (Fig. 8, *A* and *B*) relative to stance onset. Figure 8, *C–F*, show the mean values of these reversal times for the ankle, knee, and hip joints in five episodes of FW locomotion (Fig. 8, *C* and *E*) and eight episodes of BW locomotion (Fig. 8, *D* and *F*). Negative values indicate that the reversal time preceded the referent time; positive values indicate that it followed the referent time. The locomotor episodes chosen for this analysis were performed

with different body configurations. Despite this, during FW locomotion the reversals at the hip joint always preceded swing onset and stance onset (Fig. 8, *C* and *E*, respectively), whereas during BW locomotion they were always delayed (Fig. 8, *D* and *F*, respectively). In contrast, the reversals at the knee joint always preceded swing onset and stance onset during both FW and BW locomotion (Fig. 8, *C* and *E*, and Fig. 8, *D* and *F*, respectively). Finally, the time of the reversals at the ankle joint almost coincided with the knee reversals during FW locomotion (Fig. 8, *C* and *E*), whereas in different episodes of BW locomotion (most likely depending on the body configuration) could precede or follow swing onset (Fig. 8*D*) and stance onset (Fig. 8*F*).

DISCUSSION

In the present study, we compared the kinematics of FW and BW locomotion performed by mice on three setups (a tunnel, a treadmill, and an air-ball). We found that the different environmental conditions provided by these setups did not affect the basic characteristics of locomotor movements. On each of the setups, mice were able to perform locomotion in a particular direction with a wide range of speeds, cycle durations, and step lengths, which strongly overlapped. Similar results have been obtained in previous studies comparing the kinematics of overground and treadmill FW locomotion in humans (Riley et al. 2007), horses (Barrey et al. 1993), cats (Miller et al. 1975), rats (Pereira et al. 2006), and mice (Herbin et al. 2007). However, we found some differences in the kinematics of locomotor movements performed on different setups, which are discussed below.

Fig. 8. Sequence of motion reversal times occurring at different hindlimb joints during forward (FW) and backward (BW) locomotion. *A* and *B*: examples of angle changes at the ankle, knee, and hip joints during FW (*A*) and BW (*B*) locomotion. Direction of the angle change corresponding to joint extension (E) and flexion (F) is indicated by arrow. *C* and *D*: mean values for time of motion reversal (indicated by red arrows in *A* and *B*) at the ankle, knee, and hip joints relative to swing onset in 8 episodes of FW (*C*) and 5 episodes of BW (*D*) locomotion. *E* and *F*: mean values for time of motion reversal (indicated by dark blue arrows in *A* and *B*) at the same joints relative to stance onset. Positive and negative values of time indicate, respectively, the time of motion reversal preceding and delayed in relation to the referent time (swing onset or stance onset). Data related to different episodes are shown by different colors. Sequences of the motion reversal times are shown at *bottom* in *C–F*, e.g., in *E* the reversal took place first at the knee, then at the ankle, and finally at the hip joint. FW locomotor episodes are the same as in Fig. 5, *B–F*, whereas BW locomotor episodes include those shown in Fig. 7, *G–L*. Data obtained in different episodes are shown in *B–E* by different colors.
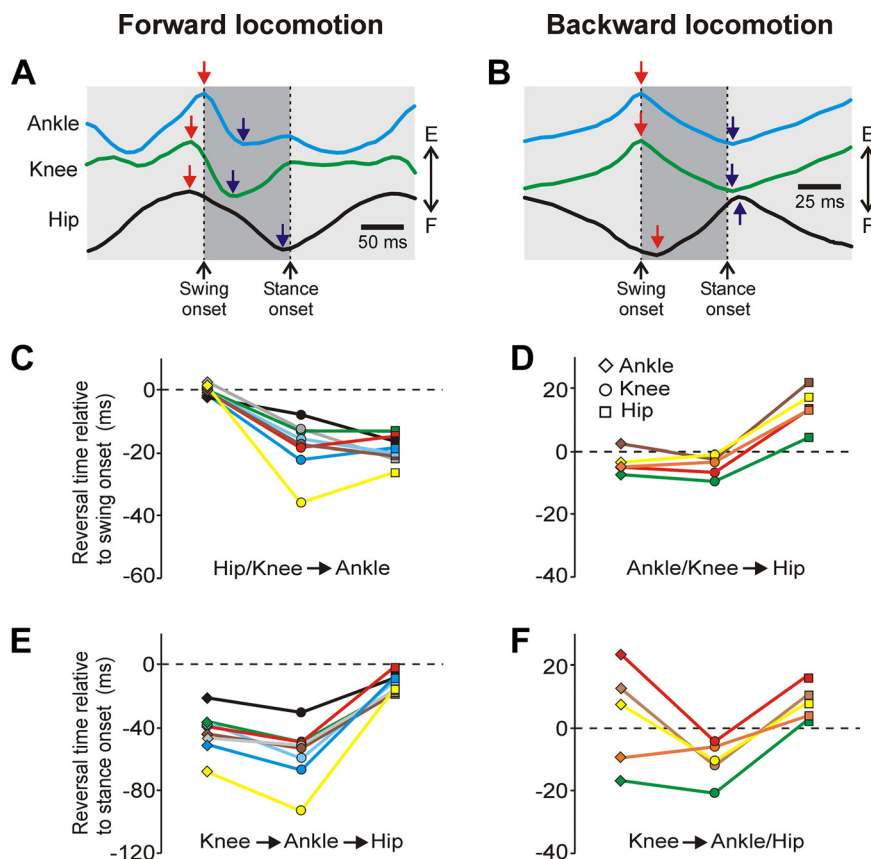
We found that in mice the height of the pelvis ("hip height") was maintained almost constant during individual FW and BW locomotor episodes but differed in different episodes. We found a correlation between hip height and the rostro-caudal range of the hindlimb movements in relation to the hip. With a high hip height, the extreme rostral and extreme caudal positions of the paw were symmetric in relation to the hip ("middle steps"), whereas a reduction in hip height was associated with anterior or posterior bias in the paw trajectory ("anterior" and "posterior" steps, respectively). We found that mice preferred to use specific forms of stepping during FW locomotion on a particular setup. Thus, on the air-ball they walked with middle or posterior steps, whereas they used middle or anterior steps on the treadmill. Possibly, mice prefer posterior stepping on unstable surfaces (such as the floating air-ball) because it increases the base of support and thus the postural stability. On the other hand, anterior stepping on a moving treadmill belt (posturally stable but behaviorally insecure environment) allows mice to switch quickly from FW to BW locomotion (see below) and to escape any sudden threat that may appear in front of them. Finally, in the tunnel (which represents a rather natural environment for mice, as they live in burrows) the entire repertoire of possible stepping was observed. It was previously reported that cats and mole rats use anterior and posterior stepping during downslope and upslope FW locomotion, respectively (Carlson-Kuhta et al. 1998; Eilam et al. 1995; Smith et al. 1998), and cats use posterior stepping during crouched FW walking (Trank et al. 1996).

We found that, in contrast to FW locomotion, mice performed BW locomotion with strong dorsiflexion of the spine and used anterior stepping on all of the setups. Such a body configuration during BW locomotion is also typical for cats (Buford et al. 1990) but is not observed in dogs (Vilensky and Cook 2000). Buford et al. (1990) argued that the specific body configuration during BW locomotion in cats was crucial to the animal's ability to propel itself backward (Perell et al. 1993). However, the lack of such a body configuration during BW locomotion in dogs suggests that it is not obligatory and may play a different role. For example, dorsiflexion of the spine and anterior stepping may be an ethological adaptation related to the aggressive context: shifting the weight bearing to the hindlimbs frees the forelimb for defensive actions and also facilitates (if necessary) a forward leap.

As in other animals and humans (Ashley-Ross and Lauder 1997; Buford et al. 1990; van Deursen et al. 1998; Thorstensson 1986), speed during BW locomotion in mice was slower than that during FW locomotion. This correlated with the lower cycle duration and shorter steps (reduced excursions of the paw) during BW compared with FW walking. Shorter cycle durations, with a concomitant reduction in step length, may reflect a perceived reduction in stability during BW locomotion. As in all studied animals and in humans (Buford et al. 1990; Clarke and Still 1999; Grasso et al. 1998; Halbertsma 1983; Hruska et al. 1979; Thorstensson 1986; Vilensky and Cook 2000), an increase in the FW or BW locomotor cycle duration in mice was caused by an increase in stance duration while swing duration remained essentially constant. We found that swing duration during BW locomotion in mice was almost half that observed during FW. One can suggest that a shorter swing duration helps to improve balance, as it results in a

higher proportion of the cycle when the body is supported by three or even four limbs.

Although mice can perform FW locomotion with different interlimb coordinations (walk, trot, gallop, bound; Bellardita and Kiehn 2015), in our experiments they performed only FW walking. Observation of these symmetric gaits is consistent with the data of Bellardita and Kiehn (2015) for the same range of locomotor speeds (<60 cm/s), whereas the asymmetric gaits were observed by these authors for speeds higher than 75 cm/s, which were never seen in our study. Coordination of the fore- and hindlimbs during FW locomotion in our experiments corresponded to walk with a lateral sequence (Hildebrand 1980). It was suggested that this gait, used by the vast majority of mammals, provides better postural stability (Gray 1944) than the gait corresponding to walk with a diagonal sequence used by some mammals (Hildebrand 1980). During BW locomotion, mice walked with either a lateral or a diagonal sequence or trotted and sometimes even switched from one gait to another within the same locomotor episode. One can suggest that during BW locomotion in mice postural stability is provided by the short duration of the swing phase, so that an exact stepping sequence is not essential, and therefore intergirdle coordination is weakened. A rather variable stepping sequence during BW locomotion, which does not correspond to any standard gait pattern, has been described in salamanders (Ashley-Ross and Lauder 1997). In contrast, cats and mole rats have a stable gait during BW locomotion corresponding to walk with a diagonal sequence (Buford et al. 1990; Eilam and Shefer 1992).

We found that during FW locomotion in mice intralimb coordination strongly depended on the form of stepping (anterior, middle, or posterior). Although motions at the hip joint were qualitatively similar during different forms of stepping (flexion during swing and extension during stance), motions at the knee and ankle joints were substantially different. For example, during stance almost pure extension of these two joints was observed during posterior stepping, almost pure flexion during anterior stepping, and weak flexion-extension sequence during middle stepping. Motions at the knee and ankle joints similar to those observed in mice during FW anterior and posterior stepping were described in cats performing anterior and posterior stepping during FW downslope and upslope/crouched walking, respectively (Carlson-Kuhta et al. 1998; Smith et al. 1998; Trank et al. 1996). We found that during BW walking the intralimb coordination in mice was similar to that previously reported in cats (Buford et al. 1990): the hip joint profile was reversed, while at the knee and ankle joints flexion during swing and extension during stance were observed. In contrast, in humans and dogs the BW joint angle profiles represent reversed FW joint angle profiles (Grasso et al. 1998; Thorstensson 1986; Vilensky and Cook 2000).

The neuronal mechanisms generating FW and BW locomotion reside in the spinal cord (Courtine et al. 2009; Hsu et al. 2014; Shah et al. 2012). It was demonstrated that supraspinal command originating from the mesencephalic locomotor region specifically selects and activates the spinal networks generating FW stepping (Musienko et al. 2012). The origin of the supraspinal command activating the spinal networks that generate BW locomotion in mammals is unknown.

It has been demonstrated that displacement of epidural electrical stimulation (ES) in the rostro-caudal direction along

a lumbosacral enlargement in decerebrate cats caused, respectively, rostro-caudal displacement of the range of FW stepping movements in relation to the hip (Merkulyeva et al. 2018). It was suggested that ES increases the excitability level of the motoneurons located in the stimulated segment, and because motoneuronal pools for hip flexor and extensor muscles prevail, respectively, in the rostral and caudal segments of the lumbosacral enlargement (Vanderhorst and Holstege 1997), the rostro-caudal shift of the range of FW stepping movements was observed. One can suggest that in an intact animal specific supraspinal commands determine the ratio of the excitability level of hip flexor/extensor motoneuronal pools leading to a specific form of FW stepping. In contrast, it is most likely that anterior stepping during BW locomotion is not determined by supraspinal command but rather is hardwired in the spinal networks, as ES evokes only anterior BW stepping in decerebrate cats (Merkulyeva et al. 2018; Musienko et al. 2012).

To conclude, in the present study the kinematics of FW and BW locomotor movements performed by mice on different setups has been characterized for the first time. The data obtained represent a benchmark for modern studies based on manipulations with activity of specific populations of neurons to reveal their roles in the control of particular aspects of locomotor movements.

GRANTS

DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the authors.

AUTHOR CONTRIBUTIONS

T.G.D. and P.V.Z. conceived and designed research; M.G.V., T.G.D., and P.V.Z. performed experiments; M.G.V. and P.V.Z. analyzed data; M.G.V., T.G.D., and P.V.Z. interpreted results of experiments; M.G.V., T.G.D., and P.V.Z. prepared figures; M.G.V., T.G.D., and P.V.Z. drafted manuscript; M.G.V., T.G.D., and P.V.Z. edited and revised manuscript; M.G.V., T.G.D., and P.V.Z. approved final version of manuscript.

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