



Research report

Fischer (F-344) rats have different morphology, sensorimotor and locomotor abilities compared to Lewis, Long–Evans, Sprague–Dawley and Wistar rats

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Received 13 December 2002; received in revised form 17 February 2003; accepted 17 February 2003

Abstract

Locomotor and/or sensory behaviour is commonly evaluated in laboratory rats in the field of neuroscience. Many strains of rats, however, have been propagated through intensive breeding programs. With any breeding program, traits are selected purposefully or inadvertently. We set out to investigate whether differences in morphology, sensory or motor behaviours exist using five age-matched strains of laboratory rats. Personal observations of morphological differences between different strains of rats led us to hypothesize that Fischer rats were dissimilar to the other strains in each of the parameters investigated. Evaluation of morphology involved measuring long-bone lengths and body weights of each strain. Motor skills were evaluated by measuring paw preferences while rearing, abduction of the distal portion of hindlimbs during locomotion, footfalls through a horizontal ladder during locomotion, and ground reaction forces generated during trotting. Sensory ability was assessed by von Frey testing. Fischer rats had shorter long-bone lengths, weighed less, and had significantly abducted distal portion of their hindlimbs during locomotion compared to the other strains. Lewis and Sprague–Dawley rats were less sensitive to mechanical pedal stimulation compared to Fischer rats. While rearing, all strains of rats tended to use individual forelimbs 25% of the time for each right and left limbs, and both forelimbs together 50% of the time. There were no significant differences in the number of footfalls during the ladder task. Ground reaction force determination revealed that Fischer and Sprague–Dawley rats bore more weight on their hindlimbs compared to forelimbs during locomotion, Long–Evans and Lewis rats bore more weight on their forelimbs compared to their hindlimbs, while Wistar rats distributed weight evenly between forelimbs and hindlimbs during trotting. We conclude that morphologic, sensory and motor differences exist between the five strains of laboratory rats examined and several of these differences are most pronounced in the Fischer strain.

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Keywords: Locomotion; Kinematics; Ground reaction forces; von Frey

1. Introduction

Rats have been used for experimental purposes since at least the mid 1800's [16]. Since these early experiments, and the domestication of the wild Norway rat (*Rattus norvegicus*), many strains of rats have been developed through intensive breeding programs throughout the world. Breeding and husbandry programs for the laboratory rat in North

America were primarily brought about by the Wistar Institute of Philadelphia, USA [16]. Breeding programs have been developed based on the needs of various researchers and institutes.

Many strains of rats have been propagated using inbreeding or out-breeding strategies. A particular strain of animal is considered inbred if it was created following more than 20 generations of sibling or parent–offspring matings (inbred animals are considered homozygous but homozygosity may not be truly present until after 40 sibling matings; [8,9]). Outbred strains, however, are strains that are maintained to have a considerable amount of genetic heterozygosity [12]. Confirmation of genetic heterozygosity within outbred strains can be verified using a variety of genetic screening tests [12].

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Regardless, some of the more popular strains of rats originally developed in the early 1900's are still in use today (for historical highlights of breeding practices see [16]).

Animal husbandry practices generally select and enhance particular traits of a given species of animal. There is no better example of this than the result of breeding practices of domesticated dogs. Purebred dogs have been bred for selected characteristics for hundreds of years [37]. Consequently, highly breed-specific morphological and sensorimotor characteristics have been developed. For example, dogs can range in size from 1 to 100 kg [34], and particular breeds appear to be more sensitive to painful stimuli than others [10]. Although the laboratory rat has been bred for a shorter period of time [16], these animals can propagate quickly and are intensely bred by many established institutes and animal suppliers. Like many domesticated species of animals, laboratory rats have developed many strain specific characteristics, although they may not be as obvious as in domestic dogs. Because the laboratory rat is a popular animal used for studying sensory and motor behaviours, and particular anatomical characteristics may contribute to these behaviours, we have set out to determine whether differences in morphology and sensory and/or motor behaviours exist between various strains of rats. We hypothesized that differences exist with regard to specific morphological, sensory, and motor abilities of different strains of rats. Specifically, based on grossly observing morphological differences between different strains of rats, we hypothesized that the Fischer strain is least similar amongst the strains of rats examined. By using morphologic, endpoint, kinematic and kinetic measurements, we have described the morphologic, sensory, and motor differences that exist between five strains of rats. We found that Fischer rats tend to be most dissimilar from the other strains we examined. These differences have implications for investigations involving sensorimotor behavioural manifestations of neurologic disease such as spinal cord injury. We discuss our findings in light of their significance for those studying sensory and motor behaviour in the neurosciences.

2. Materials and methods

2.1. Subjects

Eleven female rats of each of five different age-matched strains were obtained from Charles River Laboratories, Canada (Quebec, Canada). Animals were approximately 4 months of age and considered to be mature. The strains used included: Fischer (CDF(F-344)/CrIBR); Lewis (LEW/CrIBR); Long-Evans (CrI:(LE)BR); Sprague-Dawley (CrI:CD (SD)IGS BR) and Wistar (CrI:(WI)BR). Fischer and Lewis are inbred albino strains; Long-Evans (pigmented), Sprague-Dawley (albino) and Wistar (albino) rats are outbred strains. The strains, in the present study, were chosen because of their relatively common use in the neu-

rosciences, particularly in spinal cord injury (e.g. F-344 [32,33], Sprague-Dawley [2,31], Lewis (commonly used for studying immune-mediated and inflammatory phenomenon in the CNS [23]), Wistar [6,14], and Long-Evans [36]). Animals were housed in pairs with 12 h light/12 h dark cycle in the laboratory animal care facility at the Department of Veterinary Biomedical Sciences, Western College of Veterinary Medicine at the University of Saskatchewan. All animals were examined daily and deemed healthy by a licensed veterinarian. All animals were cared for according to the standards set out by the Canadian Council on Animal Care.

2.2. Training

All animals were trained to travel along the length of a 182 cm × 20 cm runway for a food reward. Animals were also trained to run the length of the runway with a horizontal ladder placed equidistant between its ends. Training was successful when the animal was able to complete these tasks consistently.

2.3. Morphometric analysis

Morphometric evaluation was performed by measuring selected long-bone lengths. The long bones measured were the: (1) femur, (2) tibia, (3) humerus, (4) radius, and (5) ulna. Animals were anaesthetised with sodium pentobarbital to permit restraint during radiography. All animals were radiographed in dorsal recumbancy with limbs outstretched using a Picker Transix 800A model radiography unit. Radiographs were taken with a setting of 100 mA, 50 kVp, and 1 mA s. All images were obtained using Kodak film and were processed routinely using an automated radiograph developer. Bone lengths were measured from the resultant radiographic images. Briefly, each bone's length was determined by measuring the straight-line distance between the most proximal and distal points of each bone bilaterally (Fig. 1A and B). All measurements were made to the nearest tenth of a centimeter. Corresponding right and left long-bone lengths were averaged together to yield the average long-bone length for each animal. In addition to bone length, body weights of each animal for each strain were recorded and analysed for differences between strains of rats. Differences in long-bone lengths between strains of animals was determined using one-way analysis of variance (ANOVA; SigmaStat, Chicago, IL) with $P < 0.05$ considered significant. Pair-wise multiple comparisons were made using the Bonferroni adjusted t -test (SigmaStat) and significance was considered if $P < 0.05$. Body weight data did not meet the criteria for parametric data. Consequently, differences in body weight between strains of rats was determined using Kruskal-Wallis ANOVA on ranked data followed by post hoc analysis using Dunn's test ($P < 0.05$ considered significant; SigmaStat).

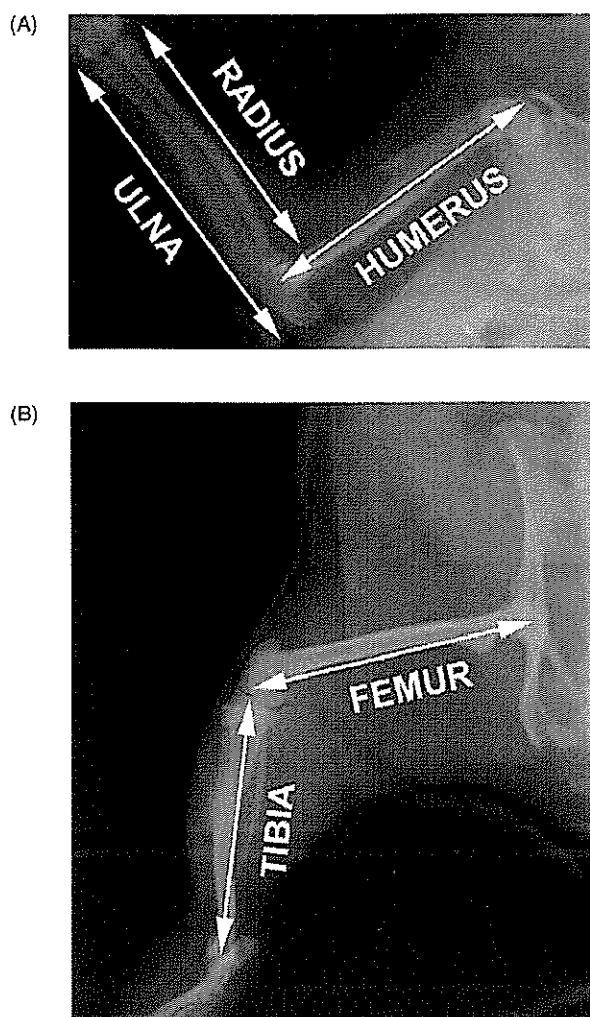


Fig. 1. (A and B) The length of each long bone was measured from the most proximal to the most distal aspect of each bone (arrows) from radiographic images.

2.4. Endpoint measurements

2.4.1. Ladder crossing

The number of times a rat's paw or limb falls between the rungs of a horizontally placed ladder during locomotion has been used to evaluate motor abilities of brain [30] and spinal cord injured rats [31]. Briefly, a ladder was constructed such that 1/4" dowels separated 1" apart (centre of one dowel to centre of next dowel) were used as rungs. The ladder was placed equidistant from the ends of a clear plexiglass runway. Animals were videotaped while crossing either 25 or 10 rungs of the ladder. Ten runs were collected for each rat. A "run" was considered successful if the rat moved across the ladder without hesitation. Videotape was analysed field by field (60 fields per second) and the total number of footfalls was recorded and expressed as percentage footfall for each rat. Differences between groups were

determined using Kruskal–Wallis ANOVA for ranked data (SigmaStat).

2.4.2. Forelimb support while rearing

Rats naturally use their forelimbs for balance and stabilisation while rearing against a wall. This naturally occurring behaviour has been exploited to describe the behavioural ability of rats with experimentally induced stroke [29], brain injury [29], dopamine depletion [29], and spinal cord injuries [1,17,29].

Each rat was placed in a clear plexiglass cylinder (46 cm high \times 40 cm diameter), set on a clear piece of glass, and videotaped from an angled mirror placed below the glass table top for 5 min. Videotapes were retrospectively reviewed field by field (60 fields per second) and the total number of times the left, right or both limbs were used for support while rearing was recorded. The criteria used to determine whether right, left or both limbs were being used was similar to that used in other studies [17]. Briefly, individual limbs (right or left) were scored when the rat would place the individual limb on the wall of the cylinder. Animals were scored as using both forelimbs if (1) both limbs simultaneously hit the wall of the cylinder or (2) for each time the animal walked along the side of the cylinder using alternating left and right limbs.

The percentage of time each rat used its right, left or both forelimbs was calculated from the total absolute values. Statistical differences were determined between strains of animals using ANOVA for absolute data and with Kruskal–Wallis ANOVA for ranked data for percentage data (SigmaStat).

2.4.3. von Frey testing

Withdrawal latencies to mechanical stimuli are commonly used in experiments involving the assessment of pain [19] or sensory function. We chose to evaluate withdrawal thresholds using von Frey filaments to determine whether differences exist between strains of rats.

Briefly, rats were placed in a clear plexiglass box placed over an opaque plexiglass perforated surface (30 cm \times 30 cm platform, 1.5 mm diameter perforations 5 mm apart) mounted on a frame such that there was easy access to the bottom of the opaque plexiglass. It has been shown that a decrease in variation occurs when using a hard, opaque surface for rats to stand on compared the more traditional use of wire mesh [22]. Animals were placed in the box, with the top covered, until exploratory behaviour subsided. Calibrated von Frey filaments (Semmes–Weinstein Monofilaments, Stoelting Co., Wood Dale, IL) were used as previously described [22] to evaluate the force required to elicit hindlimb withdrawal (mechanical threshold). Threshold was recorded as that force necessary to elicit a withdrawal three to four or four times. von Frey filaments were calibrated to the nearest hundredth of a gram of force, using a Mettler balance, prior to use as force elicited for each filament can change with environmental and physical factors (e.g.

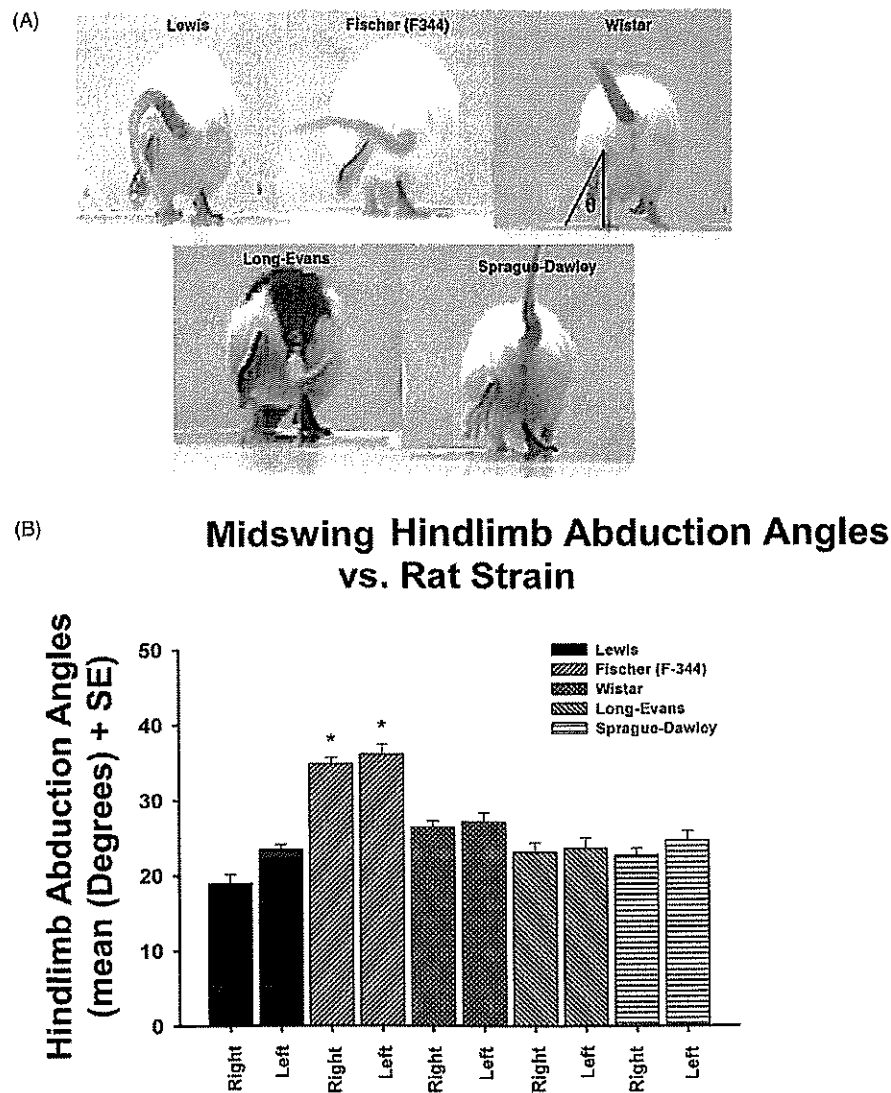


Fig. 2. (A) Distal hindlimb abduction angles were measured at midswing while rats locomoted unrestrained across a runway for food reward. (B) Fischer rats had significantly larger distal hindlimb abduction angles compared with any other strain ($n = 11$ per strain, $*P < 0.001$).

amount of use, temperature, and humidity). Differences in mechanical threshold between strains were determined using one-way ANOVA ($P < 0.05$ was considered significant; SigmaStat).

2.5. Kinematic measurements

Each rat was marked with a black, non-toxic marker along the skin covering the length of their fifth metatarsal bone to the point of the calcaneus (point of the hock) for each hindlimb. Animals were videotaped from a caudal position as they trotted along a runway for a food reward (Fig. 2A). Animals had to successfully complete at least 10 runs. A run was deemed successful if the animal moved without hesitation and was trotting. No galloping or bounding gaits were measured kinematically. Videotapes of each animal

were reviewed at 60 fields per second and two different fields/limb/run at midswing were captured and digitized using Northern Eclipse 6.0 software (Empix Imaging Inc., Mississauga, ON). Angle measurements from the edge of the fifth metatarsal to a line drawn from the point of the calcaneus perpendicular to the ground were determined electronically (Northern Eclipse 6.0) for each captured field for a given rat (Fig. 2A). These angles were averaged to yield an individual abduction angle for a given limb for each animal. These individual abduction angles were kept separate for left and right limbs for each animal of a given strain. These individual abduction angles were used to determine the mean group abduction angles for each of the five strains of rats. Differences between groups were determined using single factor ANOVA (SigmaStat). Post hoc analysis was performed using Bonferroni adjusted t -test (SigmaStat).

2.6. Kinetic measurements

As previously mentioned, rats were trained to cross the runway for food reward. Three force platforms used to describe the locomotor abilities of spinal cord injured rats [36] were used to determine ground reaction forces. These force plates are capable of measuring vertical, fore-aft and mediolateral forces. Each of the three force platforms was 10.5 cm × 11 cm. The platforms were arranged in series and embedded in the centre of the runway. Each animal's weight was recorded prior to collecting ground reaction force data. Rats triggered the data acquisition system (RC Electronics, San Raphael, CA) and an LED timer by breaking an infrared beam of light placed across the runway and located behind the force platforms. Data was amplified and analogue-to-digital converted (RC Electronics) at 1000 Hz. The LED timer was stopped when the rats broke a second infrared beam of light placed behind the last force platform. The distance between each beam of light was measured prior to collecting data and was used for calculating average velocity. Data was only collected from animals that were trotting. Trotting involves the alternating pattern of diagonal limbs being placed on the ground simultaneously. Animals were videotaped using a Super VHS video recorder (Panasonic AG-456U S-VHS, Secaucus, NJ) from a lateral position. This permitted retrospective evaluation of the limb pairs that "hit" each plate as well as permitting the determination of the time elapsed between breaking the first and second infrared beams of light (Fig. 3).

Ground reaction force data was imported as ASCII data to a personal computer. Data was divided into individual runs for each rat, then filtered in the forward and reverse direction using a modified low pass recursive filter (SPSS, Chicago, IL). Force data was expressed in proportion of body weight and normalized over time as a proportion of stride using custom written software (SPSS) for each rat. Limb pairs were



Fig. 3. Rats broke an infrared beam of light to trigger the data acquisition system and LED timer just prior to crossing the force platforms. All rats used a symmetrical trotting gait where diagonal limb pairs alternate.

kept separate and averaged together each day for each rat (minimum of five runs per trial). Only runs where the animal was travelling between 50 and 100 cm/s were used to determine the mean ground reaction forces for an individual. This averaged ground reaction force data served as representative individual ground reaction force data for each animal. All individual ground reaction force data was averaged together for a particular strain to provide the mean strain ground reaction force data. Standard error of each strain's group data was calculated using custom written software (Microsoft Visual Basic 6.3, Microsoft Corp.). Animals had to appear to be travelling at a relatively constant velocity both at data collection time and upon inspection of videotape.

Variables of the ground reaction force data examined statistically included: (1) symmetry between the peak vertical forces for the fore- and hindlimbs bilaterally (calculated as: peak vertical force forelimb/peak vertical force hindlimb); (2) ratio of peak braking to peak propulsive forces for fore- and hindlimbs bilaterally (calculated as: |peak braking force/peak propulsive force|); (3) symmetry peak vertical forces between diagonal limb pairs (calculated as: (peak vertical force left fore, right hind)/(peak vertical force left hind, right fore)); and (4) net peak fore-aft force for one complete stride taken from summed ground reaction forces (calculated as: (peak braking + peak propulsive force for left fore-right hind) + (peak braking + peak propulsive force for right fore-left hind)). For vertical force symmetry ratios a value greater than 1 indicates higher peak vertical force on forelimbs compared to hindlimbs and values less than 1 indicate higher peak vertical force in the hindlimbs compared to the forelimbs. Values greater than 1 for braking to propulsive peak force ratios indicate higher braking forces, values less than 1 indicate higher propulsive forces are being generated. For symmetry of peak vertical forces for diagonal limb pairs values greater than 1 indicate higher peak forces generated on left fore-right hindlimb pair, values less than 1 indicate higher peak forces generated on right fore-left hindlimb pair, values equal to zero indicate perfect symmetry between diagonal limb pairs. Finally, for net peak fore-aft force for one complete stride taken from summed ground reaction forces, values above zero indicate that the animals were accelerating, and less than zero indicate that animals were decelerating. Ideally, the amount of force generated in the propulsive direction should equal the amount of force generated in the braking direction if constant velocity is maintained while travelling over plates. All variables were calculated or determined using custom written software (Microsoft Visual Basic 6.3). Statistical differences were determined using either one-way ANOVA or Kruskal–Wallis ANOVA for ranked data depending on whether data met the criteria for equal variances and normal distribution (SigmaStat). Post hoc analysis was performed using Bonferroni adjusted *t*-test for parametric data and Dunn's method for determining differences for nonparametric data (SigmaStat).

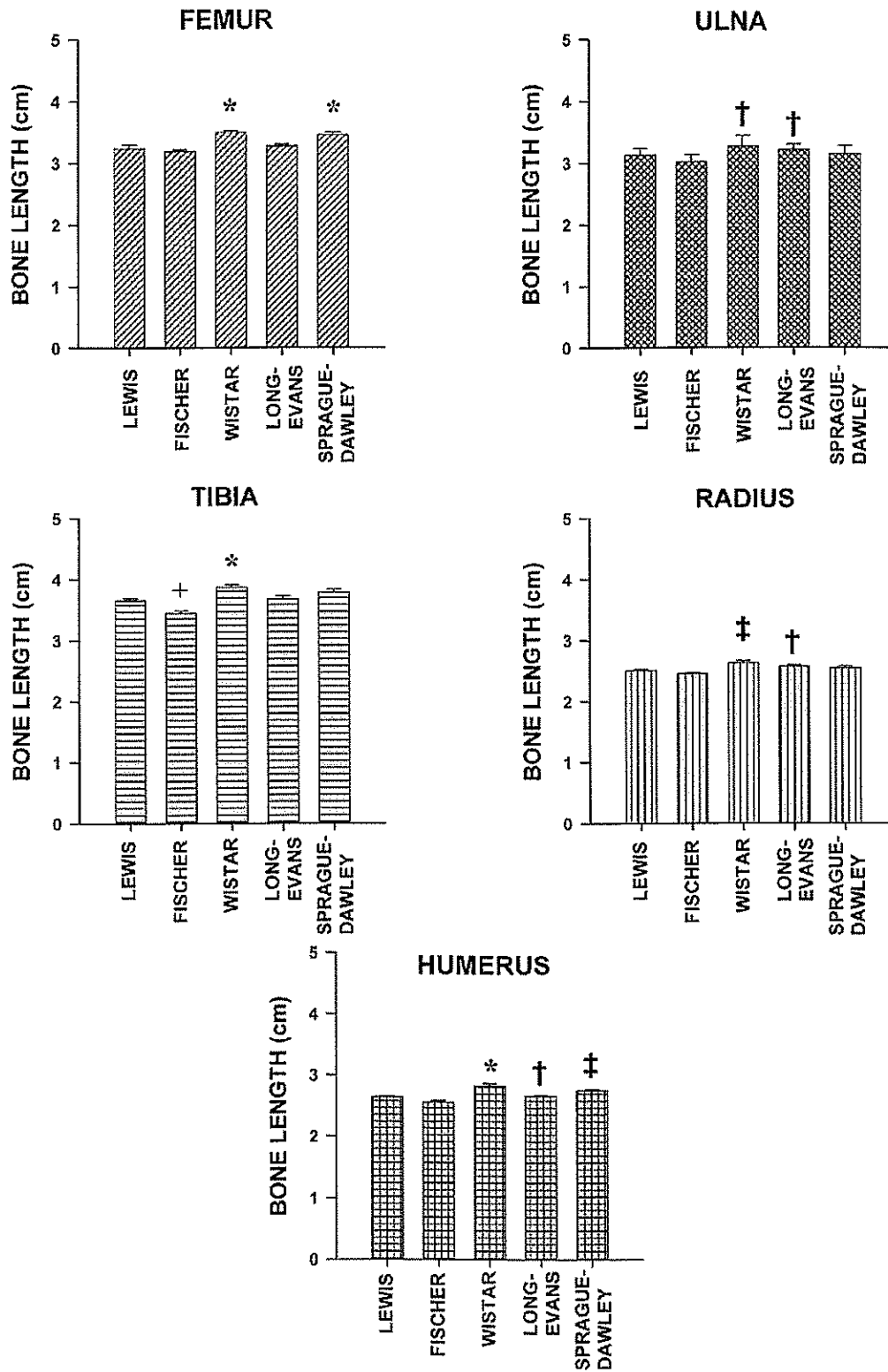


Fig. 4. Fischer rats had significantly shorter long-bone lengths, whereas Wistar had longer bone lengths compared to many of the other strains ((*) significantly different from Fischer, Lewis, and Long-Evans; (+) significantly different from Sprague-Dawley, Long-Evans and Lewis; (‡) significantly different from Fischer and Lewis; (†) significantly different from Fischer ($n = 11$ per strain)).

3. Results

3.1. Morphometric analysis

There were several differences noted between long-bone lengths of different strains of rats (Fig. 4). Wistar and Sprague–Dawley rats had longer femurs than Fischer ($P < 0.01$), Lewis ($P < 0.01$) and Long–Evans ($P < 0.01$) rats. Wistar rats also had longer tibiae when compared to Fischer ($P < 0.001$), Lewis ($P < 0.01$) and Long–Evans ($P < 0.01$); there were no statistical differences between the tibiae of Sprague–Dawley, Long–Evans, and Lewis rats. Fischer rats had significantly shorter tibiae than Sprague–Dawley ($P < 0.001$), Long–Evans ($P < 0.01$), and Lewis ($P = 0.01$) rats. Wistar rats had longer humeri than Fischer ($P < 0.001$), Lewis ($P < 0.001$), and Long–Evans ($P < 0.001$) rats. Sprague–Dawley rats' humeri were longer than Fischer ($P < 0.001$) and Lewis ($P < 0.05$) rats, and Long–Evans rats' humeri were longer than the Fischer strain ($P < 0.05$). Wistar and Long–Evans rats had longer radii than Fischer rats, but Wistar rats also had longer radii compared to Lewis rats. Last, Wistar and Long–Evans rats had significantly longer ulnae compared to Fischer rats.

Significant differences existed between strain body weight ($H = 32.65$, 4 df, $P < 0.001$). Post hoc analysis of body weights demonstrated that Fischer rats weighed significantly less than all other strains ($P < 0.05$, Fischer median = 187 g, Wistar median = 272 g, Long–Evans

median = 247 g, Sprague–Dawley median = 246, and Lewis median = 238 g).

3.2. Endpoint measurements

3.2.1. Ladder crossing

Fischer rats had a median footfall incidence value of 1% compared to 0.4% for Lewis and 0% for the remaining strains. No statistically significant differences were observed between strains of rats ($H = 6.79$, 4 df, $P = 0.148$) for number of footfalls between the rungs of the ladder.

3.2.2. Forelimb support while rearing

No significant differences were determined between strains of animals for the percentage of time they used their right ($H = 4.24$; 4 df; $P = 0.375$), left ($H = 4.28$; 4 df; $P = 0.37$) or both limbs together ($H = 4.52$; 4 df; $P = 0.34$) for support while rearing against a plexiglass cylinder (Fig. 5). All strains tended to use their right and left limbs approximately 25% of the time while using both limbs together approximately 50% of the time. Examination of the total absolute number of times that each strain used their left, right and both limbs for use while rearing indicated a significant difference between strains ($F_{(4,54)} = 10.7$, $P < 0.001$). Post hoc analysis indicated that Wistar rats total forelimb activity was greater than Lewis ($P < 0.001$), Fischer ($P < 0.001$), and Sprague–Dawley ($P = 0.002$), while Long–Evans rats placed their limbs significantly more

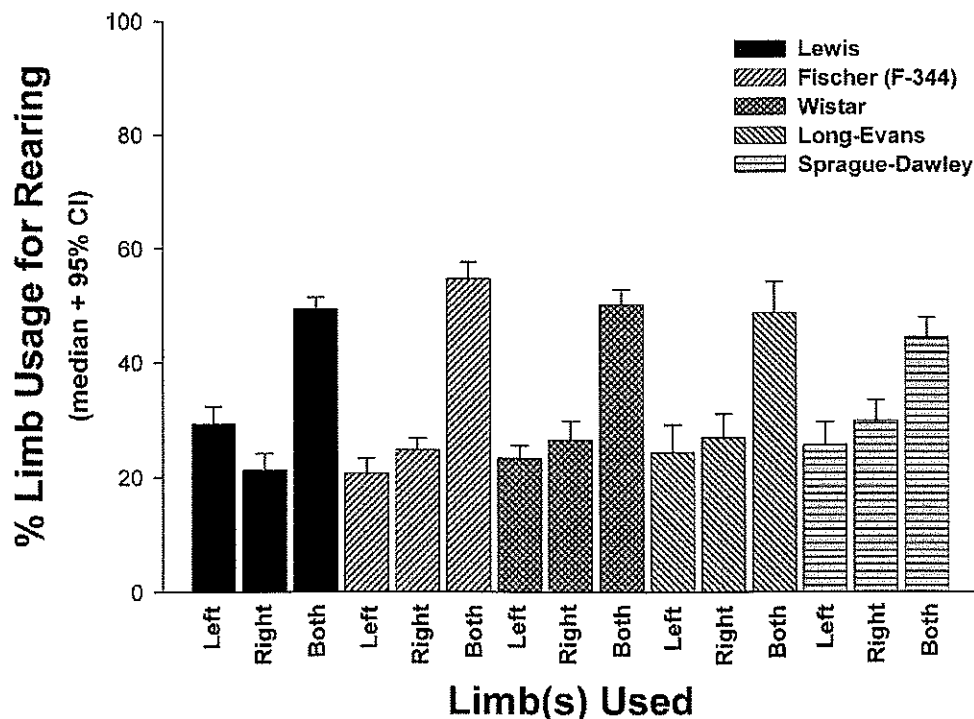


Fig. 5. No statistical difference was observed between the different strains for paw preference while rearing against a vertical cylinder. All strains used each of their forelimbs independently approximately 25% of the time while using both limbs together the remaining 50% of the time.

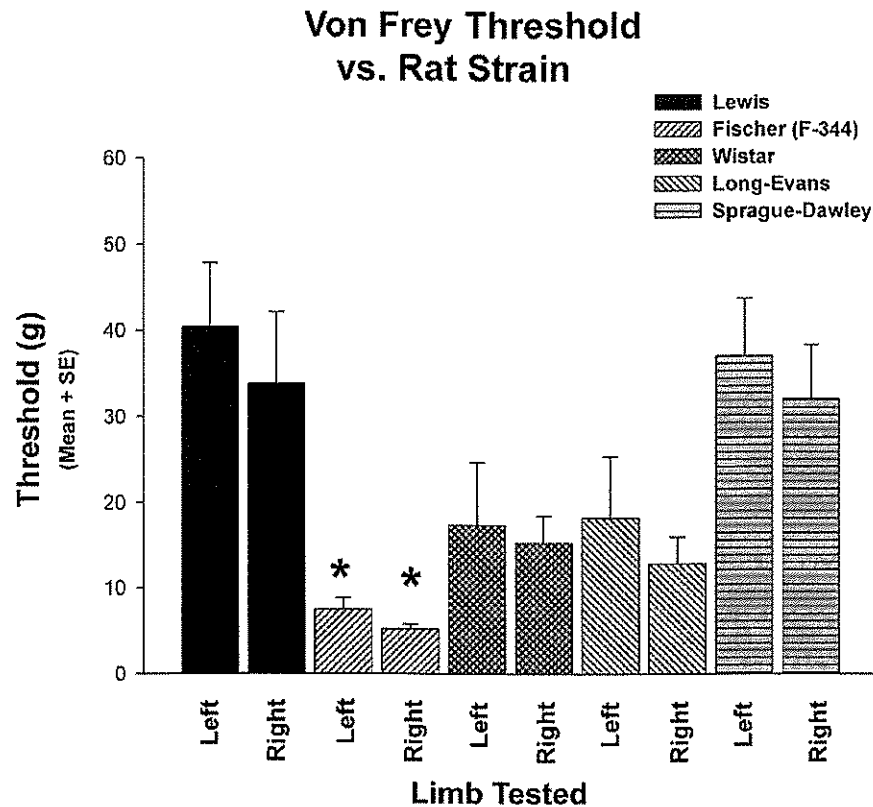


Fig. 6. Fischer rats had significantly lower thresholds for mechanical paw stimulation compared to Lewis and Sprague–Dawley rats. ($n = 6$ per strain; $*P < 0.05$).

often than Lewis ($P < 0.05$) and Fischer ($P < 0.05$) (strain (mean total foot placements \pm S.E.M.): Wistar (94 ± 5); Long–Evans (74 ± 10); Sprague–Dawley (54 ± 6); Fischer (42 ± 7); Lewis (38 ± 7)).

3.2.3. von Frey testing

There was a statistical difference observed for thresholds to mechanical sensitivities between strains of rats ($F_{(4,25)} = 4.74$, $P = 0.005$). Post hoc analysis revealed that this difference was due to Lewis and Sprague–Dawley rats having significantly higher threshold values ($P < 0.05$) compared to Fischer rats (Fig. 6).

3.3. Kinematic measurements

Bilateral distal hindlimb abduction angles were significantly larger for Fischer rats when compared to the other four strains ($P < 0.001$ bilaterally; Fig. 2A and B).

3.4. Kinetic measurements

Ground reaction force determination combined with limb overlap data indicated that all strains of rats moved over the force plates at a trot (Fig. 7). Trotting is considered a symmetrical gait where diagonal limb pairs hit the ground at approximately the same time (Fig. 7). These findings are

consistent with previous reports of ground reaction forces of female Long–Evans rats. Summing the ground reaction forces for right and left limbs provides the total ground reaction forces acting on the body. For trotting animals, this involves summing ground reaction forces for diagonal limb pairs. There were no asymmetries observed from examining the total ground reaction force versus time graphs (Fig. 7). Left and right limbs were used similarly and symmetrically for both forelimbs and hindlimbs within each particular strain (Fig. 8).

Statistically significant differences were detected for peak vertical forelimb to hindlimb ratios for left ($F_{(4,52)} = 13.8$; $P < 0.001$) and right ($F_{(4,52)} = 11.9$; $P < 0.001$) limbs (Fig. 8). Post hoc analysis revealed this difference was due to Lewis rats having statistically higher peak vertical force ratios (forelimb:hindlimb), bilaterally, compared to Sprague–Dawley ($P < 0.001$ left and right), Fischer ($P < 0.001$ left and right) and Wistar rats ($P < 0.024$ left, $P < 0.023$ right). Long–Evans rats had higher peak vertical force ratios compared to Sprague–Dawley ($P < 0.001$ left and right) and Fischer ($P < 0.001$ left and right). These statistical differences occurred because Lewis and Long–Evans rats generated higher peak vertical forces with their forelimbs compared to their hindlimbs, while Fischer and Sprague–Dawley rats generated smaller peak vertical forces with their forelimbs compared to their hindlimbs

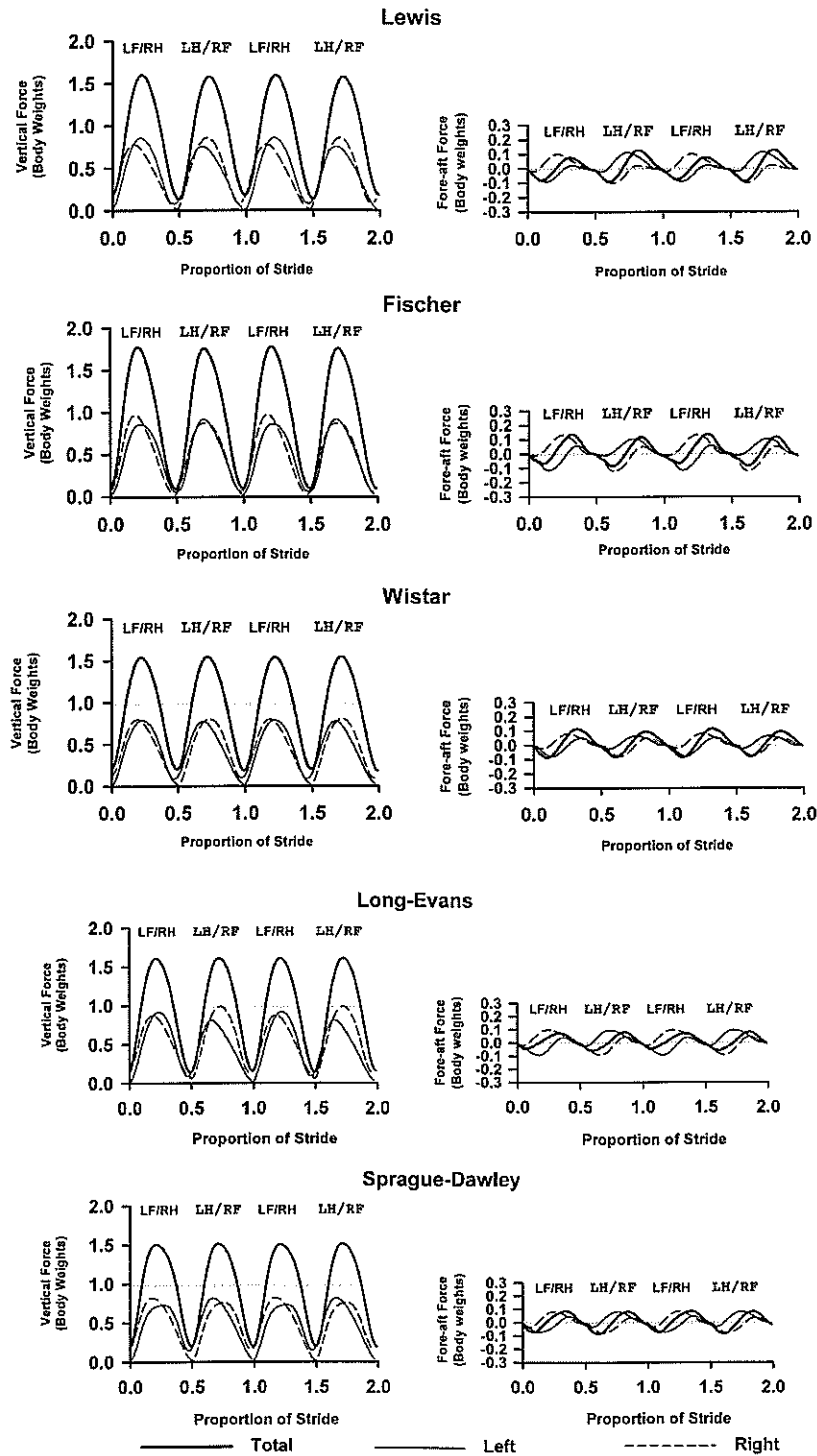


Fig. 7. Summed ground reaction forces revealed that all strains of rats locomoted symmetrically while at a trot. Peak summed vertical forces were symmetrical between diagonal limb pairs for all strains ($n = 11$ for Fischer, Lewis, Long-Evans, and Wistar rats; $n = 9$ for Sprague-Dawley rats ($P = 0.09$)).

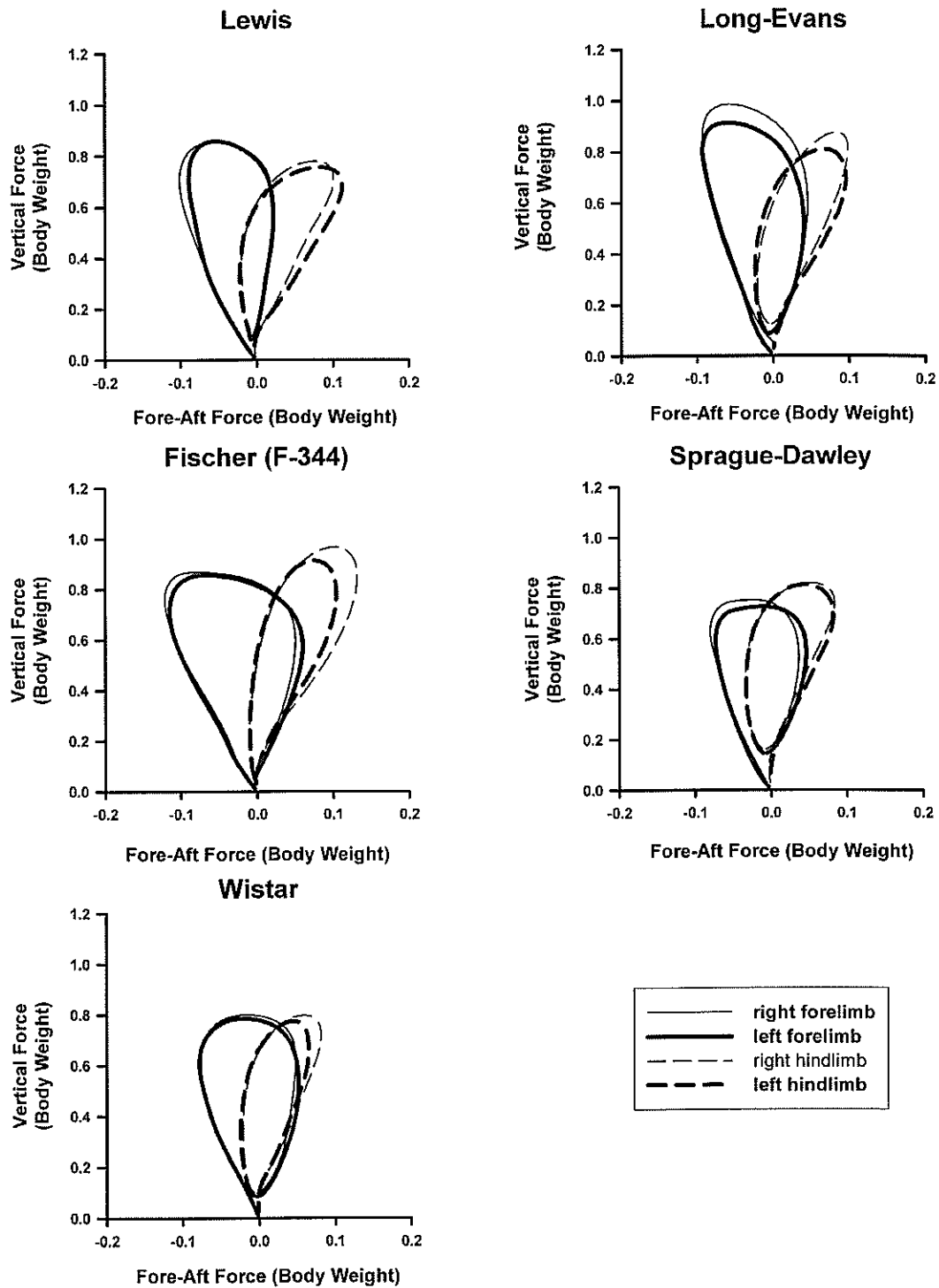


Fig. 8. Vector dynamograms for each strain illustrate the contribution of fore- and hindlimbs to braking and propulsion, and the asymmetry observed between relative weight bearing for fore- and hindlimbs during trotting. For all strains, forelimbs are used mostly for braking, while hindlimbs are used mostly for propulsion. Lewis and Long-Evans bore more weight on their forelimbs while Sprague-Dawley and Fischer rats bore more weight on their hindlimbs. Wistar rats distributed their weight evenly between the fore- and hindlimbs ($n = 11$ for Fischer, Lewis, Long-Evans, and Wistar rats; $n = 9$ for Sprague-Dawley rats).

(Fig. 8). Wistar (1.01 left ratio, 0.989 right ratio) rats generated similar peak vertical forces with their fore- and hindlimbs. Overall, Lewis rats use their forelimbs more than their hindlimbs for bearing body weight compared to Sprague–Dawley, Fischer and Wistar rats, and Long–Evans rats bear more weight on their forelimbs compared to their hindlimbs than Sprague–Dawley and Fischer rats. Wistar rats tend to bear approximately 50% of their body weight on their forelimbs and 50% on their hindlimbs.

Analysis of braking to propulsion ratios indicated that all rats have values greater than 1 for forelimbs and less than 1 for hindlimbs (Fig. 8). Statistical differences were detected for hindlimb peak braking to peak propulsive forces bilaterally ($H = 13.3$; 4 df; $P < 0.05$ (left hind) and $H = 12.8$; 4 df; $P < 0.05$ (right hind)). Fischer rats had smaller braking:propulsion force ratios, and generated smaller braking forces, compared to Wistar and Sprague–Dawley rats (Fig. 8). No significant differences were detected between strains of rats for forelimb peak braking to propulsive ratios.

Analysis of peak vertical forces generated by diagonal limb pairs revealed no statistical difference between strains of animal ($H = 1.22$; df = 4; $P = 0.875$). Median values for the strains of rats approached 1, indicating that nearly perfect symmetry was attained between diagonal limb pairs (Lewis = 0.992; Sprague–Dawley = 0.983; Wistar = 0.987; Fischer = 0.991; Long–Evans = 0.979).

Analysis of net peak fore-aft force for one complete stride revealed no statistical difference between strains ($F_{(4,52)} = 2.1$; $P = 0.09$). Statistical power was lacking, so there may actually be a difference, although we cannot detect one. Mean values for each strain indicated that some rats may have been accelerating more than others (values above zero indicate net propulsive force, below zero indicate net braking force) ((mean \pm S.E.M.): Fischer = 0.11 ± 0.02 ; Long–Evan = 0.07 ± 0.04 ; Wistar = 0.04 ± 0.02 ; Lewis = 0.03 ± 0.03 ; Sprague–Dawley = 0.01 ± 0.02).

4. Discussion

Several recent papers have examined strain differences in rats specifically pertaining to visual acuity, reaching, and spatial performance [11,24,35]. This, however, is one of the first studies which describes simultaneously the morphologic, sensorimotor and locomotor differences between different age-matched strains of rats. We found that Fischer rats are much different in their morphology, sensory and locomotor abilities compared to the other four strains examined. Fischer rats are smaller in physical stature, have increased abduction of their distal hindlimbs, and are more sensitive to mechanical stimulation of their hindlimbs than Lewis and Sprague–Dawley rats. Interestingly, the amount of time each limb is used individually or together for stabilization during rearing appears to be conserved across all strains of rats examined. That is, rats tend to use each individual forelimb approximately 25% of the time while using both fore-

limbs together the remaining 50% of the time. Wistar and Long–Evans rats, however, tended to place their paws more frequently throughout the 5 min testing period. Regardless of strain, rats are able to locomote across a horizontal ladder without slipping between ladder rungs. All strains prefer to use a trotting gait when travelling between 50 and 100 cm/s for food reward. Analysis of ground reaction forces during trotting reveal that Fischer and Sprague–Dawley rats bear more weight on their hindlimbs while Long–Evans and Lewis rats tend to bear more weight on their forelimbs. Wistar rats bear weight evenly between their forelimbs and hindlimbs during trotting. These morphologic, sensorimotor and locomotor differences have important implications for those evaluating sensorimotor behaviour in laboratory rats.

From simply observing each strain of rat in their cage, it is obvious that Fischer rats are smaller in physical stature compared to the other strains of rats, while Sprague–Dawley and Wistar rats appear to be larger rats. This observation is confirmed by our long-bone measurements and body weight measurements for each strain. Although body weight alone does not directly indicate the physical stature of an animal, in the present study we ensured that all rats were not obese by restricting their food consumption. The importance of the observed physical stature differences is relevant to those assessing behaviours requiring apparatuses that are required to “fit” the particular strain of rat. For example, the Montoya staircase test ([20]; examines skilled reaching) requires a box to be made in such a way that the rat cannot turn around in the staircase apparatus and must use each limb independently for grasping pellets. A recent study examined the effect of strain on performance using the Montoya staircase test [21]. This study found that there were indeed differences between the strains of rats examined. Although this study did not determine what factors affected strain performance for the task, they did find that weight alone was not a factor contributing to the differences observed. Physical stature was not evaluated in their study, and considering that only one-size staircase apparatus was used for all strains, it could be that the strain-dependent differences they found were due to mismatched scaling of staircase apparatus to strain size. This study did not examine the reaching abilities of Fischer rats, but one might suspect that this strain would perform differently than other strains given its small physical stature. In another more recent study it has been shown that Fischer rats displayed different abilities in skilled reaching compared to Long–Evans rats [35]. Although the cage the animals were placed in was relatively large, the slot through which the animals had to grasp the food pellet was narrow and was kept constant for both strains examined. The qualitative and quantitative differences could be due, in part, to a mismatched scaling effect of the apparatus to the strain size.

The results we obtained for footslips while locomoting across a horizontal ladder are similar to those reported for uninjured Wistar [18], Long–Evans [18], and Sprague–Dawley [31] rats. Uninjured rats typically fall between ladder rungs less than 2–3% of the time regardless of

the strain being used [18,31]. Although Metz and Whishaw described the locomotor abilities of Wistar and Long–Evans rats [18] while performing locomotion over a horizontally placed ladder, this study did not comment on whether there were significant differences between the two strains of rats. It is interesting, however, that in our study Fischer rats fell through with a median rate of 1% compared to the other strains that fell between ladder rungs 0–0.4% of the time. It could be that rung spacing should be scaled according to the physical stature of the strain being examined. This could be an important consideration if direct comparisons are to be made between results of studies using different varieties of rats especially because these differences might be enhanced following induction of some form of neurological injury or condition.

Our finding that rats use each individual limb for rearing approximately 25% for each right and left forelimbs, while using both forelimbs together approximately 50% of the time for rearing is consistent with the findings of others [17]. This implies that comparisons of results from laboratories using different strains of rats can be made without concern for rat strain contributing to differences observed between laboratories. It appears as though symmetry in limb usage during exploratory behaviour has been preserved amongst different strains of rats. Our results also provide evidence that the frequency of total limb usage over the 5 min test period varies between strains. Greater frequencies of paw placement occurred in Wistar > Long–Evans > Sprague–Dawley > Fischer > Lewis strains. These differences may, in part, be due to differences in anxiety between the different strains of rats examined. For example, it has been shown that Fischer (F-344) rats behave more anxiously than Wistar rats [4,26,27]. In addition, Fischer rats tend to rear less frequently than Wistar rats in an open field, and this difference is exacerbated with habituation [3]. These differences may influence a researcher's decision of what strain to use when designing an experiment that incorporates this particular paw preference task.

In the present study, von Frey threshold values were significantly lower for Fischer rats when compared to Lewis and Sprague–Dawley rats. This implies that Fischer rats are more sensitive to mechanical stimulation of their hindlimbs when compared to these two strains. Although not statistically significant, the trend indicated that Lewis and Sprague–Dawley were the least sensitive to von Frey stimulation, while Wistar and Long–Evans were intermediate in their response to stimulation. These results are in disagreement with a study which examined percentage of paw withdrawals to a particular force of mechanical stimulation using Long–Evans, Wistar, and Sprague–Dawley rats [19]. In this earlier study, it was found that Long–Evans responded less frequently to a given mechanical stimulus than Wistar and Sprague–Dawley, whereas Wistar rats withdrew their limb more frequently than Long–Evans rats. These results imply that Long–Evans rats are less sensitive to mechanical stimulation compared to Sprague–Dawley and

Wistar rats. The discrepancies between our study and that of Mills et al. [19] may have resulted from the particular criteria used to evaluate sensitivity to mechanical stimuli. That is, we determined the minimum force necessary to elicit consistent paw withdrawal. Mills et al. determined the percentage of paw withdrawals to a given amount of force. The discrepancies between these two studies may also reflect differences between similar strains obtained from different sources. Alternatively, the observed von Frey threshold differences may reflect, in part, factors other than the sensory ability of a particular strain. For example, anxiety, freezing behaviour or habituation may contribute to the differences observed in the present study and the discrepancies between our study and Mills et al.

Examination of von Frey thresholds concurrently with ground reaction forces during trotting provides a possible explanation of why Fischer rats are more sensitive to mechanical stimulation than the other strains. The ability to determine changes in an animal's physical environment is essential when considering the adjustments in balance and posture that must be made to meet the needs of travelling on different substrates. Ground reaction forces are similar between strains when comparing vertical force in body weights. However, one must keep in mind that Fischer rats weigh significantly less than the other strains examined. Consequently the forces they produce on the ground during locomotion are smaller. It is possible that Fischer rats require enhanced sensitivity to mechanical stimulation to determine changes in their physical environment. For example, if Fischer rats had equivalent tactile sensitivity to Sprague–Dawley rats, they may not detect changes in environmental substrates as readily and consequently as they might be unable to make any necessary locomotor adjustments.

An interesting subjective observation made prior to conducting the current investigation was that Fischer rats standing in their cages had larger distal hindlimb abduction angles. These rats do not stand as upright with their hindlimbs when compared to the other four strains. We postulated that this anatomical difference would be preserved during locomotion and, indeed, we found that Fischer rats locomote with their distal hindlimbs laterally displaced more than the other strains examined. These results could have serious implications for those evaluating recovery following spinal cord injury. A popular method used to evaluate locomotor recovery following spinal cord injury is the Basso, Beattie, Bresnahan locomotor rating scale (BBB scale; [2]). This scale was designed using Sprague–Dawley and Long–Evans rats. The BBB scale evaluates various components of the hindlimbs during the recovery phase of thoracic spinal cord injury in adult rats. One of the features examined is the degree of inward or outward rotation of the paw during the late phases of recovery [2]. In light of our present results, it is reasonable to consider that lateral deviation of the paw could be enhanced in spinal cord injured Fischer rats compared to other strains of rats. In addition,

lateral deviation of the distal hindlimb in Fischer rats may actually influence whether these rats will regain the ability to support body weight (another criteria for BBB scoring). Results of BBB scoring from studies using Fischer rats may possibly result in lower BBB scores compared to similar studies using other strains of rats. Lateral deviation of the hindlimb of Fischer rats may be one of the factors contributing to the disparity between results in recent studies examining the therapeutic potential of olfactory ensheathing glia [25,32] or the anti-inflammatory cytokine IL-10 [5,33].

Ground reaction force determination has been used to describe locomotion in a variety of species [7,13,28]. Recently, this method of evaluation has been successfully used to evaluate locomotor abilities of spinal cord injured rats [36]. Ground reaction force determination is a very sensitive method used to evaluate an animal's locomotor abilities. This form of evaluation could be used to determine the locomotor abilities of different strains of rats following various therapies for a variety of conditions. It is important to determine whether differences exist between different strains of healthy adult rats.

The present study provides evidence that the distribution of weight between fore- and hindlimbs differs between different strains and genders of rats during trotting. Sprague–Dawley and Fischer rats bore more weight on their hindlimbs compared to their forelimbs (implying a more caudally located body centre of mass) while Lewis and Long–Evans rats bore more weight on their forelimbs (implying a more cranially located body centre of mass) compared to their hindlimbs during trotting. Wistar rats bore approximately 50% of their weight on their forelimbs and 50% on their hindlimbs. This finding may have an impact on the ability of particular strains of rats to support their body following thoracic spinal cord injury. It has been shown that thoracic spinal cord injured rats bear more weight on their forelimbs compared to sham-operated controls indicating a cranial movement of their centre mass [36]. Strains which normally distribute more of their body weight on their forelimbs might compensate more quickly, and potentially locomote more effectively, following thoracic spinal cord injury, than those strains that normally bear more weight on their hindlimbs.

Recently, it has been shown that body weight is distributed towards the forelimbs during deceleration and to the hindlimbs during acceleration in trotting dogs [15]. Pitching moments (nose-up or nose-down) due to fore-aft acceleration are balanced by redistributing vertical impulse (product of vertical force and time) and this method of balance seems to be important for cursorial animals such as dogs [15]. In sprawling or crouched quadrupeds, such as rats, roll moments are equally important in maintaining balance [15]. It could be argued that the differences observed between the strains of rats with regard to peak vertical forelimb to hindlimb ratios is due to excessive acceleration or deceleration. In our study, however, the net peak fore-aft forces generated over one stride confirm that all strains

were accelerating similarly, and although statistical power was lacking, there were no differences detected between the groups of animals.

The findings of the present study are consistent with those of previous studies which examine strain differences in rats. The present report indicates that researchers evaluating sensorimotor abilities of rats should be cognisant of the potential role of strain differences. These differences could account for discrepancies between laboratories and within a laboratory where more than one strain is used. Of importance, Fischer rats appear to be most dissimilar in their morphology, sensory and motor performances when compared to Lewis, Long–Evans, Sprague–Dawley and Wistar rats.

Acknowledgements

The authors thank Dr. Valerie Verge and Tracy Wilson for providing the von Frey filaments and pedal withdrawal apparatus for this study. Funding was provided by a grant from Rick Hansen (Saskatchewan) Neurotrauma Fund to GDM and a WCVI Interprovincial Fellowship to AAW.

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