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Repeatability of gait analysis measurements in Thoroughbreds in training.

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Background: With the view of implementing gait symmetry measurements in Thoroughbreds in training for early detection of injuries, repeatability of inertial measurement unit (IMU) gait parameters needs to be established. **Objectives:** To assess the variation of head and pelvis movement symmetry in Thoroughbreds in training. **Study Design:** Daily and weekly repeat gait assessments were conducted successfully in fourteen Thoroughbreds equipped with IMUs on poll, sacrum and right (RTC) and left (LTC) tuber coxae. **Methods:** Gait was assessed in trot, in-hand, on a level concrete surface. Difference between vertical displacement minima and maxima and range of motion (ROM) were obtained. Ranges containing 50% (median), 75%, 90% and 95% of absolute daily and weekly differences were calculated and intraclass correlation coefficients (ICC) calculated for daily and weekly repeats. **Results:** Median absolute daily differences ranged from 4 mm to 7 mm and median weekly differences from 4 mm to 8 mm. 90% of daily differences were between 9 mm and 16 mm and

90% of weekly differences between 11 mm and 19 mm. ICC values were found on average across sensors and gait parameters as 0.73 (ranging from 0.40-0.92 across parameters) for daily repeats and as 0.65 (0.27-0.91) for weekly repeats. **Main limitations:** Horses were of varying training and movement asymmetry levels and no veterinary lameness examination was conducted. **Conclusions:** Daily and weekly repeat gait assessments in this group of Thoroughbreds in training show lower ICC values than previously reported from within-day repeats in horses during lameness examinations.We recommend conducting repeatability studies for specific groups of horses when planning long term studies aiming at identifying horses at risk of injury. .

Ethical Animal Research: All procedures were performed according to Singapore Turf Club (STC) ethics guidelines and with approval of the Royal Veterinary College's Ethics and Welfare Committee (URN 2013 1238). Informed consent was given by the trainers of the horses.

Source of funding: Horse Betting Levy Board (HBLB)

Competing Interests: None.

Introduction

Technological advances have provided quantitative ways of evaluating gait asymmetry with inertial measurement units (IMUs) [1,2]. Asymmetry of head and pelvic movement have been linked to changes in the mechanics of movement, i.e. changes in force production between contralateral limbs [3,4]. Retrospective analysis of force plate measurements has revealed changes in loading pattern before the occurrence of injuries to the superficial digital flexor tendon [5]. While force plate data suggest low between trial variance [5], the first step for using IMU gait assessment for early detection of injury is to quantify the amount of variability in gait data between days and weeks. Repeatability of IMU based measurements has been assessed previously for measurements in quick succession [6] and IMUs have been used successfully to quantify changes in movement asymmetry after diagnostic analgesia [7–9]. However, in the envisaged long-term scenario, it is important to estimate the combined effect of biological (day-to-day) and methodological variation, the latter related to re-instrumenting horses on different days; this variation has not been estimated for Thoroughbreds in race training.

The aim of this study was to investigate the repeatability of head and pelvic movement parameters between days and weeks (the combined effect of biological and methodological variation) in a population of racing Thoroughbred horses in training for flat racing. Emphasis was put on a realistic setting, i.e. assessment of the horses in their usual location at their training yards. We hypothesised that daily and weekly repeat measurements exceed variability values established during repeat assessments at quick succession [6]. We were also interested in comparing intraclass correlation coefficients (ICC) to ICC values from published assessments performed in quick succession in horses undergoing clinical lameness examinations [6].

Material and Methods

All procedures were performed according to Singapore Turf Club (STC) ethics guidelines and with approval of the Royal Veterinary College's Ethics and Welfare Committee (URN 2013 1238). Informed consent was given by the trainers of the horses.

Horses

Fifteen Thoroughbred horses (12 geldings, 2 colts and 1 filly, body mass: mean 503 kg (standard deviation: 33 kg, range: 438-550 kg)) deemed fit for training by their trainers, were recruited to the study from three different training yards (5 horses from each yard) located at the facilities of STC. Five horses were chosen randomly out of the pool of horses in training at each yard. Horse age varied between 2 years and 6 years (2 years: N=4; 3 years: N=2; 4 years: N=3; 5 years: N=5; 6 years: N=1). Some of the horses had not had any race starts (N=6), while others had more than 20 starts (N=3).

Instrumentation

Three MTx[a] inertial sensor units and one modified MTi-G[a] inertial sensor (IMU) were placed in a Velcro pouch and attached to the midline of the sacrum (MTi-G[a]) and to the left and right tuber coxae (LTC and RTC) with double sided tape as well as on the poll on the head band of the collar via Velcro attachments as previously described [10]. IMUs were connected by wires to an Xbus[a] transmitter in a customised pouch attached around the girth with a surcingle. Raw IMU data was sampled at 100 Hz per individual sensor channel and transmitted via Bluetooth from the Xbus[a] unit to a laptop computer running MTManager[a] software. Data collection was manually started and stopped via MTmanager[a] software.

Experimental Protocol

Horses were assessed at their trainer's yard and trotted in a straight line on a level, hard surface for at least 25 strides once a day for 5 consecutive days, then once a week for 5 consecutive weeks. Data were recorded into a laptop computer and subsequently analysed using customised software written in MATLAB[b]. All horses were in training and some did compete through the data collection period. Data collection was consistently performed after morning exercise approximately between 10 AM and 3 PM.

Data Analysis

Vertical sensor displacement in millimetres over time was obtained from each sensor [11] and was segmented into individual strides based on pelvic roll and vertical velocity of the pelvis [12]. Median values across strides were recorded for the following parameters: HD_{min}, PD_{min}, LD_{min}, RD_{min} (difference between the two displacement minima reached during left and right forelimb or hind limb stance for **h**ead, mid **p**elvis, **l**eft and **r**ight tuber coxae), HD_{max}, PD_{max}, LD_{max},

RD_{max} (difference between the two displacement maxima reached after left and right forelimb or hind limb stance for **h**ead, mid **p**elvis, **l**eft and **r**ight tuber coxae) [13], and range of motion (ROM: difference between overall minimum and maximum) for all four sensors. In addition hip hike difference (HHD, difference between upward movement amplitude of LTC and RTC during contralateral stance) and range of motion difference (RD, difference between overall movement amplitude of LTC and RTC) were calculated from LTC and RTC displacements [14]. This resulted in median values of 14 gait parameters for each assessment of each horse.

Statistical analysis

Statistical analysis was performed in MATLAB[b] (v2015a) and SPSS[c] (v22).

For estimating the amount of variation in movement asymmetry between days and weeks, absolute differences between the corresponding gait parameters obtained on consecutive days (daily differences) and consecutive weeks (weekly differences) were calculated. For example the absolute difference in HD_{min} (values for the other parameters with equivalent equations) between values of consecutive days (HD_{min}(day1) and HD_{min}(day2)) was calculated as:

$$\Delta HD_{min}(day1,day2) = |HD_{min}(day1)-HD_{min}(day2)| \quad (1)$$

Absolute differences, rather than the difference between absolute values, were used in this instance to calculate a difference that informs about the magnitude of the difference independent of the direction of the asymmetry since the latter depends on the order of gait assessments. Consequently, an absolute difference of 10mm (|10mm|) would be recorded for a horse showing +5mm asymmetry on day1 and -5mm on day2. The same absolute difference of 10mm (|-10mm|) would be recorded for a horse showing -5mm asymmetry on day1 and +5mm asymmetry on day2. Box plots were created for absolute differences (daily and weekly) for each of the 14 gait parameters and ranges (from zero) were calculated containing 50% (i.e. the median) as well as 75%, 90% and 95% of the daily and weekly absolute differences (MATLAB[b]).

Daily and weekly repeat values of gait parameters (non-absoluted, i.e. directional values in case of asymmetry parameters) were tested for normality using the Kolmogorov Smirnov test with Lilliefors significance correction at a significance level of $p<0.05$. Intraclass correlation coefficients (two-way random, with 95% confidence intervals) for daily and weekly values for each parameter and anatomical landmark were calculated (SPSS[c]) and categorized in accordance with Cicchetti [15]. Directional movement asymmetry parameters were used in this instance reflecting the fact that changes in asymmetry direction may occur between days (or weeks). This approach is also consistent with the published study with repeat measurements conducted in quick succession [6].

Results

Median values of gait parameters were calculated from a total of 5232 strides from 70 daily and 67 weekly gait assessments across 14 of the 15 horses (mean

38 strides/horse, maximum 68 strides, minimum 11 strides) (Table S1). For one horse trot ups on the 4th and 5th weeks had to be excluded and for one horse the 5th weekly trotup had to be excluded due to the feisty temperament of the horses. One horse was found to be lame (by the stable veterinarian) and was hence excluded from the study. Average values of stride to stride variability (quantified by the difference between 25th or 75th percentile and median over all strides of an assessment) across all daily and across all weekly assessments varied from about +/-4mm to about +/-9 to 10mm for the 14 gait parameters (Table S2).

Absolute differences between repeat assessments

Boxplots for absolute differences between daily and weekly values (Figure 1) illustrate the spread of values quantified for the 14 gait parameters. Ranges containing 50%, 75%, 90% and 95% of the absolute differences are presented in Table 1 and Table 2 for daily and weekly assessments.

Absolute daily differences for asymmetry variables qualitatively appear to be smaller for the sacrum (PD_{min} and PD_{max}: 50% within 4mm; 90% within 9-11mm) than for the poll (HD_{min} and HD_{max}: 50% within 5-7mm; 90% within 14-16mm). Values for asymmetry parameters derived from differences between LTC and RTC amplitudes are found in between the sacral and head values: HHD (50% within 6mm; 90% within 12mm) and RD (50% within 4 mm; 90% within 12mm). Absolute weekly differences for asymmetry parameters qualitatively appear smaller for the sacrum (50% within 4-5mm; 90% within 12-13mm) and for the parameters derived from differences between LTC and RTC amplitudes (50% within 5mm; 90% within 11-12mm) than for the poll (50% within 5-7mm; 90% within 18-19mm).

Intraclass correlation coefficients.

Kolmogorov Smirnov tests showed that, with the exception of PD_{min} ($p=0.047$), LD_{min} ($p=0.005$), LD_{max} ($p=0.028$) and PROM ($p=0.0323$), daily repeat values of the remaining gait parameters followed a normal distribution (remaining $p \geq 0.265$). Weekly repeat values of all gait parameters except HD_{min}, LD_{min} and RD_{max}($p=0.016$, and $p=0.005$, $p=0.016$) followed a normal distribution (all remaining $p \geq 0.0672$).

ICCs for daily and weekly repeat values (and their confidence intervals) are presented in Table 3. Daily ICC values are varying between 0.40 for PROM and 0.92 for LROM averaging to a value of 0.73 across all gait parameters. All daily ICC values (except for PROM which was categorized as fair) were categorized as either good (6 parameters) or excellent (7 parameters).

Weekly ICC values range from 0.27 for RD_{min} and 0.91 for RTC ROM averaging to a value of 0.645 across all 14 gait parameters. Weekly ICC values were categorized as poor for RD_{min} and fair for HD_{max}, LD_{min}, LD_{max}, RD_{max}, and PD_{min}, while the remainder were categorized as good (PD_{max}, HHD) to excellent (HD_{min}, HROM, LROM, RROM, PD_{min}, PROM and RD).

Discussion

In this study we have investigated the repeatability of 14 gait parameters calculated from four anatomical landmarks on head and trunk of Thoroughbreds in race training quantified from in-hand assessments in trot. This is a first step towards establishing the potential benefits of long term monitoring of gait asymmetry parameters for early detection of impending injuries providing veterinarians with quantitative data. Gait asymmetry is associated with a change in force distribution between contralateral limbs [3,4] and force plate measurements have highlighted the potential of subtle changes to be useful for detecting impending injuries to the superficial digital flexor tendon [5]. That study however was conducted in a retrospective fashion and force plate records were analysed only after a clinical lesion had been identified. It remains to be shown whether changes in gait asymmetry can be used prospectively, in particular since the movement asymmetry measures used here (differences between displacement minima or maxima) are less detailed than the measurements from the force plate data in [5], where measurements at specific time points over the stance phase were taken and in particular rate of loading (slope of force time curve) was found to change in the injured horses.

In this study, in addition to head and pelvic movement asymmetry, we are presenting tuber coxae movement asymmetry as well as ROM values for all sensor locations. We have included these parameters to give a more complete picture of head and trunk movement and specifically to allow for calculation of normalised asymmetry measures, such as the symmetry index [16] enabling other researchers to compare reported values to other groups of horses.

Repeatability between consecutive days

Median values for daily differences are smallest for sacral movement (PD_{min}, PD_{max}: 3-5mm) and head movement (HD_{min}, HD_{max}: 5-7mm). More practically relevant ranges – containing 90% of the daily absolute differences and hence leading to higher specificity in the context of the envisaged scenario of early detection of impending injuries – are considerably higher with values of 14-16mm for head movement and 9-15mm for sacral movement. This suggests that differences between repeat assessments of this magnitude should not be unexpected in this group of Thoroughbred racehorses in training.

ICCs of daily repeat measurements range from 0.62 to 0.84 (good to excellent) for head movement related parameters and (with the exception of PROM) from 0.61 to 0.92 (good to excellent) for trunk movement related parameters. Compared to a previous study with a different IMU based gait analysis system [6] where repeat assessments on the same day (within minutes of the initial assessment) resulted in ICC values ≥0.89 for head movement and ≥0.93 for pelvic movement, the day-to-day consistency reported here is lower.

Several factors may play a role here. We have reported a difference in the amount of movement asymmetry quantified between the two IMU systems [17] and are speculating that this may have to do with the different filtering approaches applied to the underlying acceleration data: a Fourier and polynomial approach [18] versus a highpass filter [19]. This may have an effect on stride to stride variability retained in the signal. Testing for this systematically is beyond the scope of the present manuscript.

It is important to emphasize that here we were dealing with Thoroughbred racehorses and the fact that data collection was not possible in all circumstances due to the temperament of some (e.g. younger, more inexperienced) horses highlights the difficulty of this task and may explain some of the high variability values found. Across 69 out of a total of 137 gait assessments, for which GPS based speed measurement was successful, an average (+/-SD) trotting speed of 3.32±0.44 m/s was found, indicating that 68% of assessments were found within +/-13.3% of the mean value, representing a considerable spread in speed. No effort was made to correct for any speed differences, since in practice, when dealing with this group of horses, control of speed may be difficult and our aim was to provide realistic values representative of the envisaged application. It is possible, that with a speed correction, for which additional data with more reliable speed measurement would be necessary, slightly smaller variability would have been found. A previous study has indicated that quantitative gait data of horses during in-hand, straight line trot is affected comparatively little by speed [20], however it may be interesting to further investigate this under the circumstances of the current study.

While in the original repeatability study [6] sensors were left in place between assessments, the study design here with measurements on consecutive days and weeks necessitated removal of the sensors between assessments. This situation is compatible with the envisaged long term monitoring of horses. However this renders it impossible to disentangle the effects of sensor placement and biological variability.

It appears likely that some of the horses, had they undergone a clinical lameness examination, would have been declared lame (see Table S1 for average and range of movement asymmetry data for daily and weekly repeats). The study design of the overarching study, aiming at investigating the predictive potential of gait assessment in Thoroughbreds in training over a continuous period of several months did not allow for any veterinary interventions other than when identified (by the staff, e.g. trainers or stable staff) during normal routine. Head and pelvic movement asymmetry values of some horses exceed the visual movement asymmetry threshold of 25% [21]. It is possible, that daily variation of movement asymmetry is different between lame and non-lame horses with considerable variation between days reported in lame horses [8].

Repeatability between consecutive weeks

Absolute differences between weeks were not considerably larger than absolute differences between days (compare values in Table 1 and Table 2 and see figure 1) as may have been expected based on the observation that movement asymmetry increases in horses in high speed training [22] and hence over longer time periods larger increases (or decreases) in movement asymmetry may be expected which would have resulted in larger weekly differences. However, the effects reported elsewhere were measured in Standardbred trotters over a training season, whereas the racehorses in this study were at varying stages of their racing career at a racetrack with all-year-round racing.

In order to evaluate the potential benefit of long term monitoring of gait with quantitative methods to detect individual horses at risk of injury it appears crucial to compare the variability values to what can be detected reliably 'by eye'. Depending on the overall movement amplitude, changes in asymmetry values of up to 16mm (90%, Table 1: daily variation) are in the region of the previously reported limits of the human eye of 25% for reliably spotting movement asymmetry [21]. A value of 17mm (just outside normal variation for the group of horses investigated here) would result in 28% asymmetry based on an assumed movement amplitude of 60mm in a trotting horse (e.g. vertical head movement in a sound horse, [23]). Quantitative assessment may hence not be more sensitive for detecting small changes between days than what can be achieved by eye. However, one distinct and essential advantage of quantitative measurement is that it is not prone to expectation bias shown to influence expert assessments [24] or to observer drift, a commonly acknowledged phenomenon in longitudinal observational research [25]. Hence quantitative assessment may the method of choice for population level studies into the development of training and racing related movement asymmetries, such as the published study conducted in Standardbred trotters [22] providing veterinarians with quantitative data for their decision making. It remains to be shown (ideally in a prospective manner) whether it may indeed be possible to detect injuries with the help of quantitative monitoring with inertial sensors. Retrospectively analysed force plate data indicates this may be possible [5].

ICC values show inconsistencies when comparing daily and weekly values (Table 3). Eleven of the 14 gait parameters show smaller weekly ICC values averaging to 0.645 while daily ICC values show a higher average of 0.732. Interestingly, the largest differences (i.e. the two parameters showing the largest differences between daily and weekly values, Table 3) are found for pelvic gait parameters calculated from differences between the minimum position of the tubera coxae (LD_{min} and RD_{min}). The minimum position of the pelvis (PD_{min}) is related to the amount of peak vertical force production during contralateral hind limb stance phases [4]. Symmetry of peak vertical force is also one of the kinetic parameters observed to change in horses with hind limb lameness [26]. We speculate that the drop in weekly ICC value may be the result of changes in gait parameters related to the intense training that racehorses undergo pushing the musculoskeletal system near its limit. This however needs further investigation in larger number of horses and with horses undergoing a clinical lameness examination.

Study limitations

While all horses were Thoroughbreds in training using identical training, racing and veterinary facilities of the STC, the horses were of varying ages and at varying stages of their racing career, some with many previous races, and some without any race starts. The amount of high speed training/racing has been shown to affect injury rates [27–29] and exercise level also affects bone remodelling, which is an important process in dealing with microdamage incurred during high intensity exercise [30,31]. A direct relationship between movement asymmetry and the introduction of high speed and incline exercise has been shown in Standardbred trotters [22]. Training and racing related factors are hence likely to influence the amount of gait asymmetry measured in our study horses (see Table S1).

It is essential to note that it was not possible to conduct gait assessments in a safe manner in all horses at all times – even without the need to attach sensors to the limbs – and this should be taken into account when planning studies with young and inexperienced Thoroughbreds. The stride to stride variability found in our study horses (Table S2, +/- 4-10mm) is of similar magnitude compared to the daily repeat values (median differences across asymmetry parameters) reported here and emphasizes the need to collect a sufficient number of strides to achieve a good estimate for average values.

Related to the study design of the overarching long term study, no veterinary lameness exams were conducted in conjunction with the data collection for this repeatability study. Movement asymmetry values of some horses exceed what can be observed reliably by eye (25%, [21] or approximately 15mm assuming an amplitude of 60mm) indicating that some horses would have been declared lame visually and presence and/or severity of lameness may affect day-to-day variability.

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Manufacturers' details

[a] Xsens, Enschede, The Netherlands

[b] The Mathworks, Natick, MA, US

[c] SPSS, IBM, Armonk, NY, US

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Figure and table legends

Table 1: Range (from zero to given value in mm) containing 50%, 75%, 90% and 95% of the **daily absolute differences** in 14 movement symmetry and range of motion parameters derived from 4 head and trunk mounted inertial measurement units in 14 Thoroughbreds in training.

	50%	75%	90%	95%
HD $_{min}$	7	11	14	16
HD $_{max}$	5	10	16	20
HROM	6	9	12	18
LD $_{min}$	5	7	14	16
LD $_{max}$	5	8	15	21
LROM	6	8	13	15
RD $_{min}$	5	8	12	17
RD $_{max}$	5	8	12	17
RROM	5	8	15	19
PD $_{min}$	4	8	11	11
PD $_{max}$	4	6	9	11
HHD	6	10	12	15
RD	4	7	12	16
PROM	7	11	15	18

Acronyms: HD_{min}, LD_{min}, RD_{min}, PD_{min}: difference between displacement minima for head, left, right tuber coxae and mid pelvis, HD_{max}, LD_{max}, RD_{max}, PD_{max}: difference between displacement maxima for Bead, left, right tuber coxae and mid pelvis, ROM: range of motion (H: head, L: LTC, R: RTC, P: pelvis), HHD: hip hike difference, RD: range of motion difference, LTC: left tuber coxae, RTC: right tuber coxae.

Table 2: Range (from zero to given value in mm) containing 50%, 75%, 90% and 95% of the **weekly absolute differences** in 14 movement symmetry and range of motion parameters derived from 4 head and trunk mounted inertial measurement units in 14 Thoroughbreds in training.

	50%	75%	90%	95%
HD_{min}	7	11	19	26
HD_{max}	5	11	18	22
HROM	6	11	17	17
LD_{min}	6	9	17	33
LD_{max}	5	11	15	18
LROM	5	10	17	22
RD_{min}	5	9	12	30
RD_{max}	5	11	16	23
RROM	5	11	15	18
PD_{min}	4	9	12	13
PD_{max}	5	9	13	18
HHD	5	10	11	15
RD	5	9	12	16
PROM	8	14	15	27

Table 3: Intraclass correlation coefficients calculated across day-to-day (daily ICC) and week-to-week (weekly ICC) repeat measurements of gait parameters in 14 Thoroughbred racehorses in training by means of head and pelvis mounted inertial sensors during in-hand trot.

Gait parameters	Daily ICC	Weekly ICC	Daily - Weekly
HD_{min}	0.84 (0.69;0.94)	0.76 (0.55;0.91)	0.08
HD_{max}	0.62 (0.39;0.83)	0.40 (0.15;0.71)	0.22
HROM	0.77 (0.58;0.90)	0.75 (0.54;0.90)	0.02
LD_{min}	0.81 (0.65;0.92)	0.47 (0.22;0.76)	0.34
LD_{max}	0.73 (0.54;0.89)	0.58 (0.33;0.82)	0.15
LROM	0.92 (0.84;0.97)	0.82 (0.65;0.93)	0.10
RD_{min}	0.68 (0.47;0.86)	0.27 (0.05;0.61)	0.41
RD_{max}	0.61 (0.38;0.82)	0.47 (0.22;0.76)	0.14
RROM	0.88 (0.76;0.95)	0.91 (0.80;0.97)	-0.03
PD_{min}	0.81 (0.66;0.93)	0.76 (0.55;0.91)	0.05
PD_{max}	0.73 (0.54;0.89)	0.62 (0.38;0.85)	0.11
HHD	0.70 (0.49;0.87)	0.66 (0.43;0.87)	0.04
RD	0.75 (0.57;0.90)	0.77 (0.58;0.92)	-0.02
PROM	0.40 (0.17;0.69)	0.80 (0.62;0.93)	-0.4
Average	0.732	0.645	