

Prevalence of objectively measured movement asymmetry in yearling Standardbred trotters

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Abstract

Background Objective measurement of movement asymmetry is gaining in popularity, especially as an adjunct to traditional lameness evaluation. Earlier research has highlighted the need for more knowledge regarding the clinical significance of measured movement asymmetry, and the influence of biological variation. Evaluating the locomotor system of the Standardbred trotter can be challenging, and studies using objective technology on this breed are few. The aim of this study was to quantify the prevalence and magnitude of objectively measured movement asymmetry in young, presumed sound Standardbred trotters, by performing objective movement analysis during in-hand trot and while driven on a track. Our hypothesis was that asymmetry scores would be higher when evaluating horses in-hand versus while driving on a track.

Results Of 103 horses included in the study, 77 were measured both in-hand and on the track, 24 were measured only in-hand, and two were measured only on the track. Previously set symmetry thresholds for the measurement system were used, during both in-hand and track trials. The majority of horses (91, 88.3%) did not have any trials below threshold. Front and/or hind limb parameters were above the symmetry thresholds during in-hand trials for 94 (93.1%) horses, and during track trials for 74 (93.7%) horses. For the total 180 in-hand and track trials, 166 (92.2%) trials were above threshold. Asymmetry magnitude ranged from mild to severe, with the majority of horses showing mild asymmetry. A minority of horses (19.7%) switched side of asymmetry for one or more parameters between in-hand and track trials. Trial standard deviations were overall high, mainly due to horse behavior, and this variability should be considered when interpreting the results. There was no significant effect on asymmetry of measuring horses in-hand versus driven.

Conclusions A high proportion of presumed sound Standardbred yearlings showed movement asymmetries above previously determined thresholds. The biological and clinical relevance of the study findings may be aided by examining how this asymmetry evolves over time and with training. This is important in order to ensure the welfare of the Standardbred trotter.

Background

Despite being the most commonly encountered health problem in athletic horses, lameness evaluation regularly poses diagnostic challenges to novice and seasoned equine practitioners alike. Traditionally, lameness evaluation relies on visually recognizing movement asymmetry during walking and trotting in-hand. However, subjective lameness evaluation may be unreliable, as even experienced equine veterinarians often disagree in determining from which leg the lameness originates (1,2). Veterinarians may also show bias when evaluating response to diagnostic analgesia during the lameness work-up (3).

Subjective assessment of movement irregularities can be particularly challenging in certain types of horses, such as the Standardbred trotter. Reasons for this, as suggested by experienced veterinarians and evaluators of Standardbreds (4), include that lameness seen at trot in-hand may not correlate to

lameness during training, and that the observed degree of lameness may vary with trotting speed. With these challenges of subjective lameness evaluation in mind, developing and refining more reliable, objective methods for equine lameness evaluation continues.

Objective measurement of movement asymmetry is possible with wireless technology using inertial measurement unit (IMU) sensors. This technology is rapidly gaining popularity amongst equine practitioners due to its ease of use under field conditions. When comparing a sensor-based system to traditional subjective lameness evaluation during in-hand trot, the IMU system detected induced lameness of a lesser magnitude than experienced veterinarians (5). Threshold criteria for movement asymmetry exist for a commercially available IMU system, and relate closely to between-trial repeatability (6). However, difficulties remain regarding interpretation of the clinical relevance of IMU measurements, demonstrated by the fact that a large proportion of sport horses in regular training and considered free from lameness by their owners measured above these asymmetry thresholds (7). Thus, knowledge is lacking regarding the biological variation and clinical significance of movement asymmetry in presumed sound riding horses. Being able to detect a wide range of asymmetries invites the question of what should be considered 'normal' asymmetry. This observation most likely extends to racehorses such as the Standardbred trotter as well, perhaps even more so due to the previously described challenges in subjective lameness evaluation of this breed.

The aim of the current study was to describe the prevalence and magnitude of motion asymmetry in young, untrained and presumed sound Standardbred trotters beginning training, evaluated both in-hand and during driven exercise. Our hypothesis was that asymmetry scores would be higher when evaluating horses in-hand versus driven, as trotting-up in-hand would allow the animals to move more freely versus when exercised within the constraints of a harness and sulky. In addition, we wanted to investigate potential associations between movement asymmetry and trainer, gender, height, track type, measuring in-hand asymmetry prior to or after track trials and presence of osteochondral lesions.

Results

Study population

A total of 114 horses were recruited to the study, with a median of five horses per trainer (range 1–29 horses). Average age in months at the time of measurement was 17.8 ± 1.5 , 17.5 (mean \pm standard deviation, median). Four horses had been broken to harness within the past 3–6 months, the remaining horses had been harnessed less than three months before the measurement date.

Of the 114 horses measured, eleven were excluded, due to lameness at time of measurement. One horse was excluded due to consistent pacing during measurements, leaving 103 horses included in the study. A flowchart illustrating the number of horses and trials in the study is presented in Figure 1. Included horses consisted of 56 males (55 stallions, one gelding) and 47 females. Median height at the withers was 153 cm (range 139 - 165 cm), median height at the pelvis was 157 cm (range 145–166 cm) and the median individual height difference between the withers and pelvis was 4 cm (range 1–9 cm).

Data on height was missing for one horse. Of the horses that had been radiographed for developmental osteochondral (OC) lesions (68 horses, 66.0%), 22 of 68 (32.4 %) horses had radiographic evidence of such changes while 46 of 68 (67.6 %) horses did not. For the remaining 35 (34.0 %) horses the results of radiographic screening were either unknown or radiographs had not been taken.

Movement asymmetry

Of the 103 horses included in the study, 77 horses had valid measurements from both in-hand and track trials, and for these, two trials (one in-hand and one track trial). Two trials were assessed. per horse, one in-hand and one driven on a track. For the remaining 26 horses, only one trial was available for assessment; Of 24 horses hadwith in-hand trials only, whereas two horses had track trials only, resulting in a total of 180 trials included in the study. Of the horses with in-hand trials only, technical problems with data collection during track trials precluded data evaluation from twelve12 of the horses. were measured both in-hand and driven, but due to technical issues at the time of data collection the track trials of these horses could not be evaluated. The remaining twelve yearlings were only just broken to harness and were therefore measured only in-hand. Two horses had valid track trials only due to technical issues with in-hand measurements. A total of 180 trials were included in the study. The horses were measured in-hand either before ($n = 44$) or after ($n = 45$) driven exercise. For in-hand measurements, 37 ± 13.9 strides (mean \pm standard deviation) were evaluated per trial, whereas 302 ± 276.2 strides were evaluated for driven measurements. In-hand, 20 horses had trials where stride selection was below 25 stridessteps per trial for front and/or hind limbs., however, For these horses a minimum of 18 strides were evaluated. for all horses. Speed at track trials was 5.0 ± 0.6 m/s (18.1 ± 2.3 km/h); speed data was missing for five horses. Horses were driven either on straight ($n = 30$) or oval ($n = 49$) tracks. Of the horses on oval tracks, 30 were driven clockwise around the track and 19 anticlockwise. Tracks were either not banked or the horses were driven on a non-banked part of the track.

Descriptive statistics

Symmetry thresholds in regards to asymmetry mean values were based on those defined by the manufacturer of the IMU system used, as detailed in the Methods section. Of the 103 horses measured, 91 (88.3%) horses did not have any only had trials abovebelow these defined symmetry thresholds. Values for one or more front or hind limb parameters were above thresholds for 94 horses (93.1%) during the 101 in-hand trials. For the 79 track trials, 74 horses (93.7%) had one or more front or hind limb parameter values that were above the thresholds. None of the 77 horses with both in-hand and track trials were below thresholds for all parameters for both trials. For the combined 180 in-hand and track trials, 166 (92.2%) were above thresholds. For all horses but one, standard deviations (SD) were equal to or higher than the trial means. One horse had a trial where all four parameter values had standard deviations (SD) less than, or equal to, their respective asymmetry mean values. The asymmetry parameter means of the horse were all above the symmetry thresholds. An overview of the values for the horses exceeding the thresholds for front limb parameters (head minimum (HDmin) and/or maximum (HDmax) difference) and/or hind limb parameters (pelvis minimum (PDmin) and/or maximum (PDmax)

difference) for in-hand and track trials, as well as their respective standard deviations, is detailed in Table 1.

TABLE 1

During in-hand measurements, contralateral fore limb and hind limb asymmetry was recorded for 22 horses, and ipsilateral asymmetry in 18 horses. For track measurements, 12 horses had contralateral fore and hind limb asymmetry and 14 horses had ipsilateral asymmetry.

The distribution of asymmetry categories for each limb for all horses ($n = 103$), in-hand trials ($n = 101$) and track trials ($n = 79$) are presented in Figure 2. Distribution of asymmetry categories for combined front or hind limbs was also examined (Supplementary item 1). Of 71 horses measured both in-hand and driven, and that were above threshold during in-hand trials, 14 (19.7%) horses switched side of the asymmetry for at least one front or hind limb parameter between in-hand and driven trials (Figure 3). The remaining 57 horses showed asymmetry of the same limb during both in-hand and track trials. An overview of the proportion of these horses who increased or decreased in asymmetry from in-hand to track trials is detailed in Table 2.

TABLE 2

The intra-class correlation coefficient (ICC) calculated showed that horses at the same training yard did not resemble each other more closely in terms of asymmetry than horses across the individual training yards (ICC score = 0.014).

Effects of gender, height, OC status, trial mode, type of in-hand trial, track type and direction of travel

For the HDmin and PDmin models the residuals deviated from normality and a square root transformation was implemented that rendered reasonably normally distributed residuals. Being female decreased HDmax significantly ($p = 0.02$) by 1.9 mm (SE 0.79). In the PDmax model there was an overall significant effect of height at the withers ($p = 0.03$) with a reduction of 0.12 mm (SE 0.05) in PDmax per 1 cm increase in height at the withers. There was no significant effect of trial mode (in-hand or track trial, in-hand trial pre or post track trial), height difference between withers and pelvis or OC status.

Discussion

Data from the current study demonstrates that a high proportion of Standardbred yearlings in regular training are asymmetric at the trot both when evaluated in-hand and when driven at the track. No significant differences in asymmetry parameters were found between the trial modes, therefore, our original hypothesis that horses would trot more symmetrically when exercised within the constraints of a harness and sulky was refuted. Although there was no difference in trials mode at the group level, our descriptive data show that evaluating young Standardbreds both in-hand and on the track is useful, as there were individual differences in asymmetry magnitude and sometimes side of asymmetry. The large standard deviations demonstrate substantial within-trial variability, which should be considered a

potential source of uncertainty for both visual and objective assessment of movement asymmetry in this population of young horses. This is the main limitation of our study. In our cohort of horses, one of the biggest challenges was getting acceptable trot-ups in-hand from green and excitable yearlings. Even though this affects the collected data, it also reflects the clinical reality faced by veterinarians every day. The study design of a cross-sectional, observational study also dictates a certain relinquishment of control of the measurement circumstances in favor of real-life conditions. Measuring the yearlings just as they were initiating driven exercise was deemed important in order to get data from the horses prior to any substantial training had been done. The results from this study can serve as reference values for expected asymmetry in yearling Standardbred trotters initiating their training.

In the present study, we used the predetermined thresholds of the measuring system to describe the distribution of asymmetry magnitude. As has been pointed out by others (8), the valuable use of thresholds may not lie in making a dichotomous assessment of whether a horse is 'diseased' or not, as this can only be decided by a complete clinical evaluation; rather, the thresholds might aid in removing clinical bias. It could be argued that it would be better not to apply thresholds to describe the findings in our study to avoid 'mislabeling' or misinterpreting the health status of these horses. However, thresholds also allow for easier comparison of the changes in asymmetry between in-hand and track measurements. Therefore, using the thresholds and categories of the measuring system employed was perceived as the most appropriate approach for this study.

The findings in our study are similar to those of Rhodin et al (7), where 72.5% of 222 'owner-sound' Warmblood riding horses had at least one asymmetry parameter above the same symmetry thresholds as used in our study. Although the magnitude of mean asymmetries of the riding horses matched well with the Standardbreds in our study, the trials in the cited study all had standard deviations below their respective trial means. Apart from 'owner-sound' Warmblood riding horses (7,9,10), polo ponies in training (11) as well as Thoroughbred racehorses with 'naturally occurring' gait asymmetry (8) have been assessed. A shared finding in these studies is that the majority of horses in regular exercise with an assumption of 'soundness' show substantial asymmetries during in-hand straight line trot.

A reasonable question to ask is what the underlying cause of this high proportion of asymmetry could be. In general, horses experiencing unvarying orthopedic pain usually show consistent movement asymmetry of the same limb(s) due to offloading of the affected structures through changes in loading and force production (12). In such cases, pain causing movement asymmetry can be confirmed through a lameness examination using diagnostic analgesia, or in some cases systemic analgesic and/or anti-inflammatory medication, to eliminate or substantially reduce the asymmetry. This leads us to the next question; are we measuring widespread hitherto undetected subclinical, pain-mediated disease that could potentially have a negative effect on the welfare of these horses, or are we looking at the occurrence of biological variation within different breeds and disciplines?

The yearlings in the current study did not undergo further orthopedic examination; therefore, we cannot draw any conclusions as to if or to what extent pain caused the measured asymmetry. The data collected

and presented is aimed at describing the prevalence and magnitude of movement asymmetry in a population of young Standardbreds, and not its underlying causes. Horses were included in this study on a presumption of soundness, as judged primarily by the trainer of the horse. It is debatable whether 'soundness' as assessed by non-veterinary professionals is an appropriate criterion for selecting non-lame horses. This was highlighted in a study of 57 'owner-sound' riding horses, where 37 horses were classified as lame after thorough veterinary examination (13). Keeping in mind that 'sound' horses are not necessarily expected to be perfectly symmetric, our cohort nevertheless show mean asymmetries close to those from horses with induced lameness (14), as well as horses with clinical lameness that responded to diagnostic analgesia (15,16). In our study, all yearlings that fulfilled the inclusion criteria at the respective training yards were measured, avoiding any intentional selection bias, for example by the trainer selecting horses that were suspected to have a locomotor issue.

The young horses in our cohort had recently been introduced to harness and light training pulling a driver and sulky. This adjustment may influence the locomotion of these horses at the time of measurement. This does not seem to represent a systematic effect, as horse asymmetry either increases or decreases when driving on the track. We should also consider the effect of a driver's aids more directly influencing the horse. A minority of horses completely switched side of their asymmetry between in-hand and track trials. Especially puzzling are five horses with a large change in PDmax (Figure 3), in which a right-sided asymmetry changes over to a left-sided asymmetry, but there is otherwise no discernable common denominator for these horses, such as track type. In the mixed model used for the results of the present study, track type (straight or oval track) did not affect group-level asymmetry means. Further research is needed to see if this is a consistent finding.

Movement asymmetry of Standardbred trotters has been described in earlier research. Pioneering use of a high-speed cinematographic technique in the early 1980s (17–21) documented asymmetries such as left-right differences in the diagonal trot suspension phase as well as variations in diagonal dissociation of the limbs (19) in trotters exercised on a track. Varying step length and diagonal dissociation was described in Standardbred colts measured multiple times on a treadmill at a speed of 4 m/s (21). Locomotor asymmetries were most pronounced at the last measurement, when the colts were 18 months of age, and in the group that had been trained (21). In our yearling study the mean age was 18 months, with a mean track trial speed of 5 m/s. Without disregarding the differences in measuring techniques and parameters, it is worth noting the occurrence of measurable locomotor asymmetries in 18-month old Standardbred trotters at relatively low speeds across the two studies. The authors of the above study (21) hypothesized that asymmetries in the locomotion patterns of younger Standardbreds was a further manifestation of congenital laterality or sidedness. If so, should movement asymmetry increase, decrease or stabilize with age and training?

In 16 Swedish Standardbred trotter yearlings that were followed over 2.5 years, vertical displacement asymmetry increased during intensified training periods (22). While front limb asymmetry decreased after these periods, hind limb asymmetry remained elevated (22). In a group of French Standardbred horses, younger horses were more asymmetrical across different parameters than older horses, and this may

conceivably be caused by immaturity of gait and lack of co-ordination (23). Stages of learning in the development of gait optimization have been described in children (24). However, without the use of control groups we cannot readily discern the effect of age from the effect of training in horses. In young horses, the effect of growth on locomotion patterns must also be considered. In our study, the parameter PD_{max} was significantly influenced by height at the withers, but only to a small degree. The difference between height at the withers and height at the pelvis, calculated as a potential measure of intensity of growth or growth spurts, was not significant.

Conclusion

Our cohort of Standardbred trotter yearlings show a high proportion of movement asymmetry, with considerable individual variation between measurements. Our proposed hypothesis that asymmetry would be lower when the horses were driven was not confirmed. Within-trial variability was high, influencing the reliability of the data. This variability gives a degree of uncertainty to the results and affects our ability to assess the significance of our findings. This does not mean that the measured asymmetries are unimportant, or the horses necessarily misclassified as asymmetric, as the opposite may just as well be true: uncertainty due to high variability might also hamper our recognition of clinically significant asymmetries. The relationship between movement asymmetry and orthopedic disorders is especially important, as this has implications for the welfare of the Standardbred trotter. Going forward, exploring the effects of age, training and speed on asymmetry in these horses may provide further answers regarding the role of biological variation versus clinically significant orthopedic issues.

Methods

Study design and cohort description

Fifteen trainers, known to have Standardbred yearlings in training, were contacted regarding study participation. Twelve agreed to participate. One additional trainer was recruited based on advertisement of the study. Training yard-level inclusion criteria were location (proximity to Oslo, Norway or Stockholm, Sweden), a licensed professional trainer in charge, and willingness to participate in the study over time. One additional stable in southern Sweden was included despite not fulfilling the proximity criteria due to the large amount of horses available at the yard.

Horse-level inclusion criteria were breed, age and training level; only Standardbred trotter yearlings that were broken to harness and within the first six months of driven exercise were recruited to the study. The horses were assessed by their trainer as fit to train, meaning that the trainer had not observed any lameness or other issues of the horse that would lead to reduced or paused training. Exclusion criteria were horses that paced instead of trotted during trials, or an observed subjective lameness of >2/5 degrees according to the American Association of Equine Practitioners (AAEP) scale (0–5) during in-hand measurement.

Clinical examination and measurements of movement asymmetry

The horses were examined at their training yards or local racetrack. All horses underwent a general physical examination with auscultation of heart and lungs, measurement of height at the withers and pelvis, palpation of the musculature and distal extremities as well as an evaluation of their conformation. Examination was performed by one of the authors (ASK, EHSH or MH). The trainers filled out a questionnaire pertaining to their horses' history of previous injury, veterinary treatment including dental examinations, hoof care/shoeing routine and radiographic screening for osteochondrosis (OC; radiographs included all four fetlock joints as well as hocks).

The horses were measured at the trot both in-hand and driven on a track with a sensor-based objective gait analysis system (Lameness Locator®, Equinosis, St. Louis, MO, USA). The horses were measured in-hand either before or after driven exercise. In-hand the horses were trotted in a straight line by their regular handler or one of the authors (ASK, EHSH, MR or EH). The ground surface was firm, consisting of either gravel, asphalt, packed dirt or hard packed snow/ice, and as even and level as circumstances allowed. Default software settings (Lameness Locator 2017 v1.2r) were used for stride selection for the asymmetry analyses; preferred stride selection was ≥ 25 steps. At the time of measurement, data from in-hand trials were subjectively deemed valid when the horse completed a trot-up with acceptable straightness and regularity. One in-hand trial per horse was used for analysis.

For track trials, the horses were exercised by their usual driver, with their regular tack and according to their planned schedule. All tracks were dirt tracks with a surface of packed dirt/sand, mixed with snow during the winter months. An electrode belt with a heart rate monitor (Polar Equine Belt, Polar Electro, Kempele, Finland) was placed around the trunk of the horse in front of the harness. Heart rate data was sent via Bluetooth to a device worn by the driver (Polar M450, Polar Electro, Kempele, Finland), which in addition registered speed, distance and route of the trial by means of an integrated GPS. Track trials were subjectively assessed as valid when the horse had completed a trial with acceptable straightness and regularity, and stride selection was ≥ 25 steps. If a horse had more than one valid track measurement, the first measurement was used for analysis. No standardized warm-up was performed as the yearlings were in an early phase of training and were only driven shorter distances at low speed.

The IMU sensors of the movement analysis system were mounted on four locations on the horse: the poll, at the top of the withers, on the pelvis (between the tubera sacrale) and at the dorsal aspect of the right front pastern. For attachment on the poll, a purpose-made neoprene head bumper was attached to the head collar or bridle, and on the right front pastern a purpose-made pastern wrap was used. Sensors on the withers and pelvis were fastened with extra strong double-sided adhesive tape (Teppeteip, Clas Ohlson, Insjön, Sweden) and standard-issue duct tape; care was taken to ensure sagittal midline positioning. During track trials, the withers and pelvis sensors were covered with additional adhesive tape (Snøgg Animal Polster, Norgesplaster AS, Vennesla, Norway) to prevent loosening. The pastern wrap was secured with elastic tape (Norbind, Norgesplaster AS, Vennesla, Norway) to prevent rotation during exercise. The IMU sensors consisted of a tri-axial accelerometer, gyroscope and magnetometer that

recorded the vertical acceleration of the head and torso and the angular velocity of the right front limb at 200 Hz with 8-bit digital resolution. Data transmission from the sensors was wireless via Bluetooth technology to a nearby computer tablet with the corresponding program software. For measurements on oval tracks, the IMU system tablet was placed in a small backpack worn by the driver to ensure continuous connection between the horse-mounted sensors and the receiving computer tablet.

Data processing

The program software mathematically converted the measured acceleration into a measurement of vertical displacement of the horse's body using a double integration process (6). The biological basis for this approach is the natural locomotion of the horse. During trot, horses alternate between loading and unloading each diagonal leg pair with a corresponding sinking and rising of the head and pelvis. During this repeated diagonal loading and unloading, unequal head movement is interpreted as reflecting forelimb asymmetry, while unequal pelvic movement reflects hind limb asymmetry. Software data output consisted of four parameter values for each trial measurement calculated from the mean difference in head minimum (HDmin) and head maximum (HDmax) positions between the right and left diagonal of each trotting stride, and the mean difference in pelvis minimum (PDmin) and pelvis maximum (PDmax) positions between the right and left diagonal of each trotting stride (6). In addition, the vector sum (VS) of the mean HDmax and HDmin values was calculated as $\sqrt{(\text{HDmax}^2 + \text{HDmin}^2)}$. Left front or hind limb attributed asymmetries were defined by negative values and right front- or hind limb asymmetries by positive values. Parameter output data were expressed in millimeters. A parameter value of 0 mm would indicate perfect symmetry between the left and right portion of the strides of a trial measurement. More detailed descriptions of the data processing can be found elsewhere (6,7).

Statistical methods

Descriptive data calculations

Criteria for movement asymmetry were based on recommendations for clinical use by the IMU system provider (25) and correspond to published confidence intervals for repeatability of measurements with the system (6). This applies to the use of symmetry thresholds and subsequent division into asymmetry categories in this study, and not in regard to the recommended limits for standard deviation. For front limb VS the symmetry threshold was 8.5 mm, for front limb values (HDmin, HDmax) ± 6 mm and for hind limbs (PDmin, PDmax) ± 3 mm; parameter values below these pre-determined thresholds were consequently defined as 'symmetric'. When measurements above these symmetry thresholds were observed, asymmetry categories defined as "mild", "mild-moderate", "moderate", "moderate-severe" and "severe" were used based on a set increase in millimeter asymmetry by adding the threshold value (8.5, 6 or 3 mm, for front limb VS, front limb asymmetry, and hind limb asymmetry, respectively) to the threshold for each parameter, corresponding to the classification presented in the IMU system output data (AIDE statement) with the exception of an additional 'severe' category. The resulting categories had thresholds for VS/front limb/hind limb values in mm as follows: Symmetric: 0-8.5/0-6/0-3, mild asymmetry: 8.5-

17/6-12/3-6, mild-moderate asymmetry: 17-25.5/12-18/6-9, moderate asymmetry: 25.5-34/18-24/9-12, moderate-severe asymmetry: 34-42.5/24-30/12-15, severe asymmetry: >42.5/>30/>15. For all horses, trial output data (VS, HDmin, HDmax, PDmin, PDmax) for each trial condition (in-hand and/or track), was systematized according to the described thresholds to assess the distribution of limb asymmetry. Other categories were made based on the trial standard deviation magnitude, with three categories based on distance from the trial mean: Trials with SD > 120% of asymmetry mean, trials with SD > 50% and < 120% of asymmetry mean and trials with SD < 50% of asymmetry mean. These categories correspond to the levels of evidence (weak, moderate, strong) presented in the IMU system output data (AIDE statement).

A combined score per horse was created for front and hind limbs where the horse was classified as front limb asymmetric if one front limb parameter (HDmin or HDmax) was above threshold, and hind limb asymmetric if one hind limb parameter (PDmin or PDmax) was above threshold. Horses could be included in both front and hind asymmetry categories. The combined severity category was based on the highest asymmetry score.

For the horses with both in-hand and track trials, those with in-hand trial parameter values above threshold were noted, and corresponding values for the track trials of these horses were compared. Changes in parameter values were registered as one of three alternatives: The horse showed asymmetry of the same limb and above threshold during both trial modes; the horse showed same limb parameter values below symmetry threshold for track trials; or the horse showed parameter value asymmetry above threshold during both trial modes but changed the side of asymmetry between trials (left to right or vice versa).

An intra-class correlation coefficient (ICC) was calculated to investigate if horses at the same training yard resembled each other more closely in terms of magnitude of movement asymmetry than horses across the individual training yards.

Model building

Movement asymmetry data was analyzed using open software (R, version 3.6.1, The R Foundation for Statistical Computing, Vienna, Austria). Mixed models were created using the lmer function in the lme4 package. Four models were created, where each outcome variable was the absolute values of one of the four asymmetry parameters HDmin, HDmax, PDmin and PDmax. In all models, fixed effects were mode (with the levels: in-hand before track, in-hand after track, driven on a straight track or driven on an oval track) gender (male or female), height at the withers, height difference between the withers and pelvis, OC status (OC at radiographic screening; yes, no or missing data). Trial speed and surface were not included in the model as these were considered similar for all horses. Horse nested within trainer was entered as a random effect (random intercept) in all models. Normality of residuals was checked using q-q plots and homoscedasticity by plotting the residuals against the fitted values. Evaluation of statistical significance was made using type II p-values generated by a Wald F test with Kenward-Roger approximated degrees of freedom. The level of significance was defined as < 0.05.

List Of Abbreviations

AAEP: American Association of Equine Practitioners

HDmin: Difference in head minimum position between the right and left portion of one trotting stride

HDmax: Difference in head maximum position between the right and left portion of one trotting stride

ICC: Intra-class correlation coefficient

IMU: Inertial measurement unit

LF: Left front limb

LH: Left hind limb

NA: Not applicable

OC: Osteochondral lesion

PDmin: Difference in pelvis minimum position between the right and left portion of one trotting stride

PDmax: Difference in pelvis maximum position between the right and left portion of one trotting stride

RF: Right front limb

RH: Right hind limb

SD: Standard deviation

SE: Standard error

VS: Vector sum of mean HDmax and HDmin values

Declarations

Ethics approval and consent to participate

The study and all procedures were approved by the ethics committee at the Faculty of Veterinary Medicine, Norwegian University of Life Sciences (approval number 14/04723-47) and were in accordance with national legislation regarding ethical animal research (FOR-2015-06-18-761). A signed consent form for participation in the study was obtained from trainers of all horses included in the study.

Consent for publication

Not applicable.

Availability of data and materials

The datasets used and analysed during the current study are available from the corresponding author upon reasonable request.

Competing interests

The authors declare that they have no competing interest.

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Authors' contributions

MR and EH designed the study and ASK, EHSH, MH, MR and EH collected the data. ASK, EPS and EH performed data analysis and statistics. ASK and CTF wrote the manuscript. All authors contributed to data interpretation and revising the manuscript, as well as read and approved the final manuscript.

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Tables

Table 1: Trials exceeding the symmetry threshold for one or more parameters.

Parameter, side of asymmetry	Number of trials	Asymmetry mean (mm)	Range of asymmetry mean (mm)	SD mean (mm)	Trials with SD > 120% of asymmetry mean	Trials with SD > 50% and < 120% of asymmetry mean	Trials with SD < 50% of asymmetry mean
IN-HAND							
HDmin, right	25	14.2	6.3 - 30.0	22.5	19	6	0
HDmin, left	23	-13.6	-6.1 - -27.9	20.6	11	11	1
HDmax, right	21	11.6	6.1 - 21.8	14.4	10	11	0
HDmax, left	17	-11.6	-6.9 - -16.3	18.1	11	6	0
VS, right	24	18.1	9.6 - 34.8	N/A	N/A	N/A	N/A
VS, left	26	-16.2	-9.6 - -28.8	N/A	N/A	N/A	N/A
PDmin, right	23	5.9	3.2 - 11.9	7.3	13	10	0
PDmin, left	25	-6.0	-3.3 - -10.4	9.0	14	11	0
PDmax, right	30	5.2	3.0 - 10.4	6.8	14	14	2
PDmax, left	27	-4.9	-3.1 - -11.4	6.6	12	15	0
TRACK							
HDmin, right	22	10.2	6.0 - 19.8	13.4	10	12	0
HDmin, left	12	-10.0	-6.2 - -17.9	14.6	7	5	0
HDmax, right	24	11.7	6.4 - 20.2	12.6	10	12	2
HDmax, left	17	-10.4	-6.1 - -15.6	15.7	12	5	0
VS, right	31	13.4	8.5 - 20.5	N/A	N/A	N/A	N/A
VS, left	18	-13.8	-9.1 - -21.8	N/A	N/A	N/A	N/A
PDmin, right	23	7.4	3.1 - 27.5	7.8	10	11	2
PDmin, left	18	-7.1	-3.1 - -11.2	6.4	5	11	2
PDmax, right	18	5.1	3.2 - 13.0	4.8	6	11	1
PDmax, left	23	-5.9	-3.1 - -11.2	6.3	9	14	0

Horses; n = 103, in-hand trials; n = 94, track trials; n = 74. Left limb side asymmetry = negative values, right limb side asymmetry = positive values. NA = not applicable. VS = Vector sum of mean values of HDmin and HDmax, HDmin = difference in head minimum positions between right and left portions of the stride, HDmax = difference in head maximum positions between right and left portions of the stride, PDmin = difference in pelvis minimum positions between right and left portions of the stride, PDmax = difference in head maximum positions between right and left portions of the stride.

Table 2: Increase or decrease in limb asymmetry from in-hand to track trials.

	IN-HAND TRIAL	TRACK TRIAL		
Parameter, side of asymmetry	Horses with parameter above threshold in- hand trial	Horses with parameter > threshold both trials, <i>increase</i> in asymmetry from in-hand to track trial	Horses with parameter > threshold both trials, <i>decrease</i> in asymmetry from in-hand to track trial	Horses with parameter <i>decreased</i> to < threshold from in- hand trial
HDmin, right	19	3	4	12
HDmin, left	15	3	4	8
HDmax, right	17	8	3	6
HDmax, left	11	1	2	8
VS, right	16	5	6	5
VS, left	16	5	5	6
PDmin, right	19	7	4	8
PDmin, left	18	6	4	8
PDmax, right	19	7	3	9
PDmax, left	18	7	2	9

Change in asymmetry of horses (n = 57) that exceeded the parameter threshold during in-hand trials and did not switch side of asymmetry between in-hand and track trial.

Definitions of VS, HDmin, HDmax, PDmin and PDmax as described for Table 1.

Figures

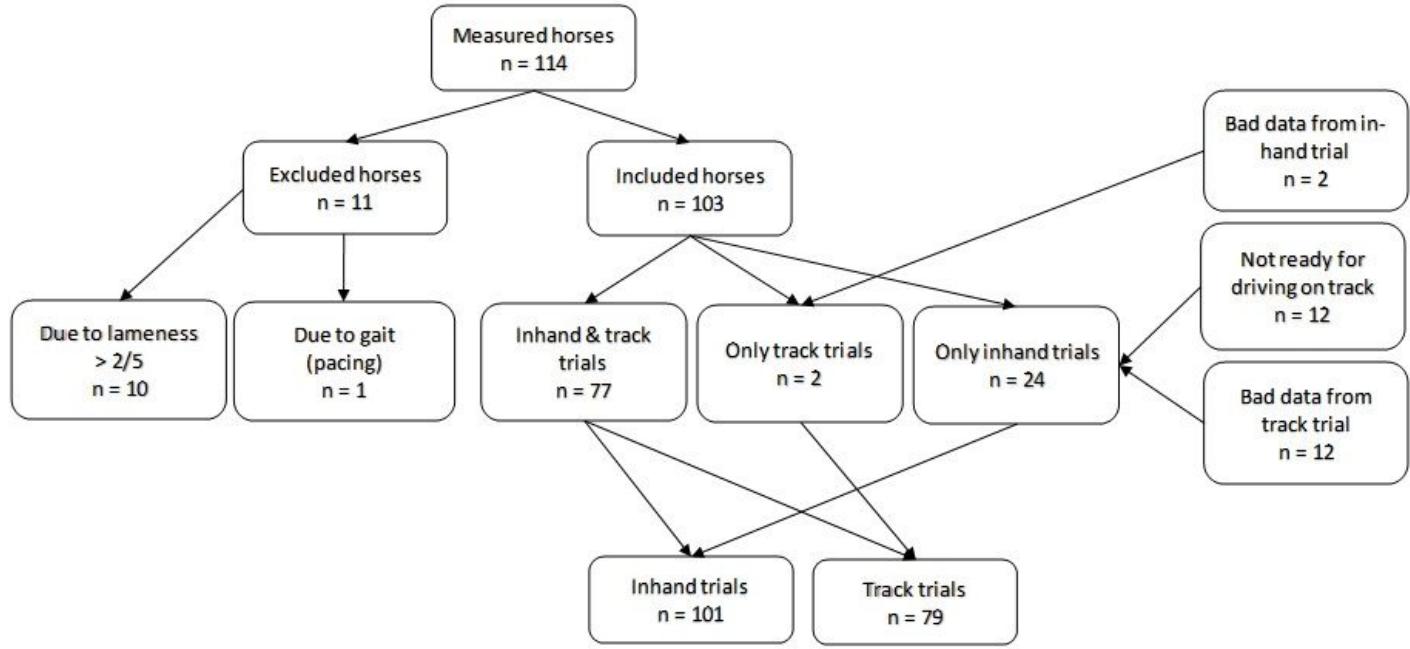


Figure 1

Flowchart of number of horses and trials in the study.

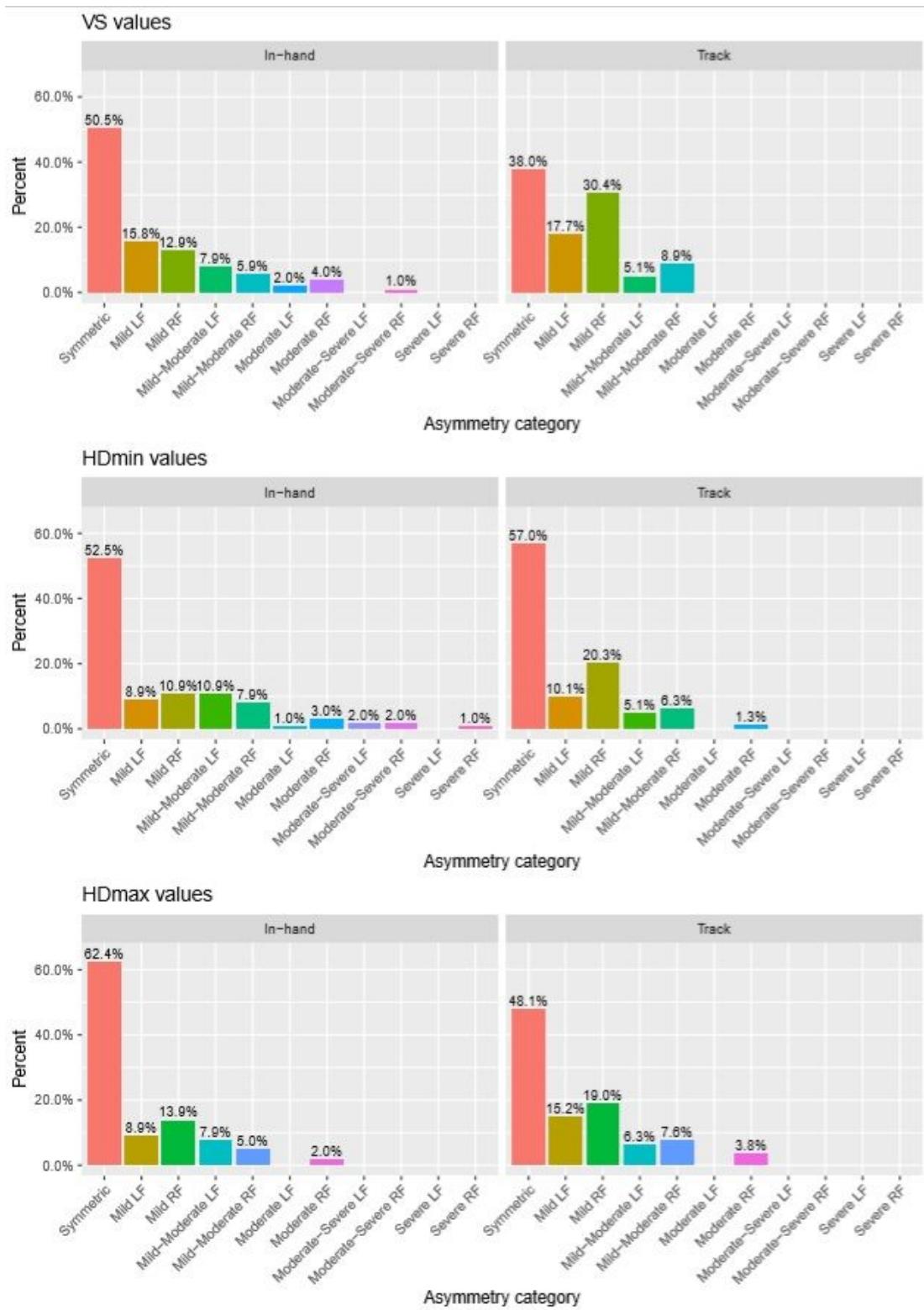


Figure 2

Distribution of limb asymmetry parameters for all horses. Horses; n = 103, in-hand trials; n = 101, track trials; n = 79. LF = Left front limb, LH = Left hind limb, RF = Right front limb, RH = Right hind limb.

Asymmetry in mm per category for VS/HDmin or HDmax/PDmin or PDmax: Symmetric: 0-8.5/0-6/0-3, mild: 8.5-17/6-12/3-6, mild-moderate: 17-25.5/12-18/6-9, moderate: 25.5-34/18-24/9-12, moderate-

severe: 34-42.5/24-30/12-15, severe: >42.5/>30/>15. Definitions of VS, HDmin, HDmax, PDmin and PDmax as described for Table 1.

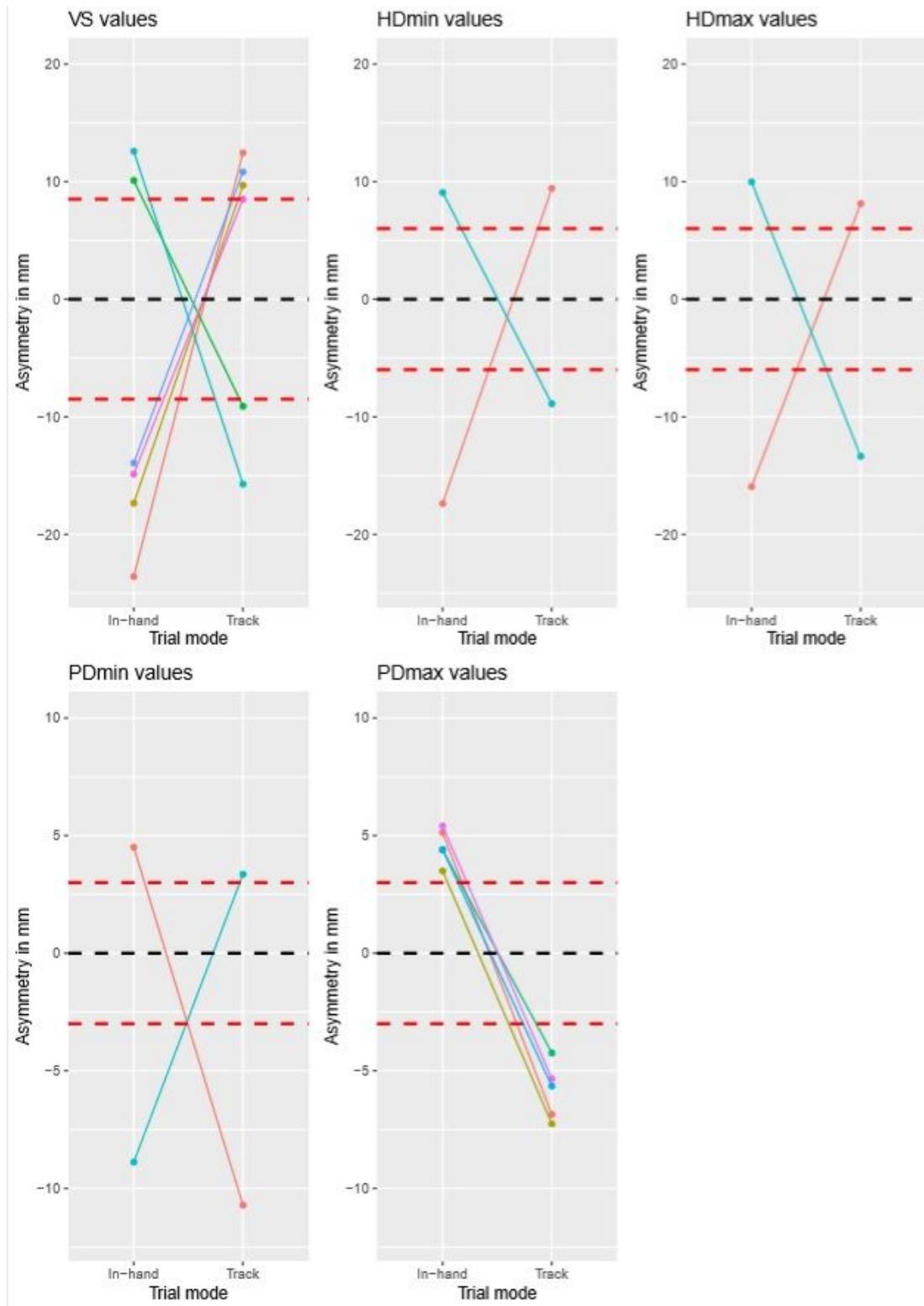


Figure 3

Horses ($n = 14$) that switched sides of limb asymmetry between trials. Each colour in the lineplot represents an individual horse. Left limb side asymmetry = negative values, right limb side asymmetry = positive values. Red stippled line denotes the symmetry threshold for the parameter (VS 8.5mm,

HDmin/HDmax 6 mm, PDmin/PDmax 3 mm). Definitions of VS, HDmin, HDmax, PDmin and PDmax as described for Table 1.