

## Movement patterns in horses with multiple limb lameness

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Independent Project • 30 credits Swedish University of Agricultural Sciences, SLU Faculty of Veterinary Medicine and Animal Science Veterinary Medicine Programme

Uppsala 2024

#### Movement patterns in horses with multiple limb lameness

Rörelsemönster hos flerbenshalta hästar

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Credits:	30 credits				
Level:	Second cycle, A2E				
Course title:	Independent Project in Veterinary Medicine				
Course code:	EX1003				
Programme/education:	Veterinary Medicine Programme				
Course coordinating dept.:	Department of Clinical Sciences				
Place of publication:	Uppsala				

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2024

Keywords:

Year of publication:

multiple limb lameness, gait analysis, motion asymmetry, lameness assessment

#### Swedish University of Agricultural Sciences

Faculty of Veterinary Medicine and Animal Science Veterinary Medicine Programme

#### Abstract

Disorders from the locomotion apparatus is the most common disease category in horses and a common cause for euthanasia. A frequent symptom of these disorders is lameness and to be able to treat such disorders the correct diagnosis needs to be made. Detecting lameness presents its difficulties due to limitations in the human visual ability, bias and the complexities of orthopaedic diseases and biomechanics. Lameness can emerge in one or multiple limbs and the mechanisms to decrease the loading of the affected limb often results in the misperception of lameness in other limbs, via a so-called compensatory lameness. Sometimes after a successful diagnostic analgesia of the primary lameness another lameness appears, revealing existing problems in multiple limbs. While the movement pattern of horses with unilateral and compensatory lameness is investigated in several studies, the movement pattern of horses with multiple limb lameness is not. The aim of this study was to investigate horses with multiple limb lameness and whether specific movement patterns can be indicative of multi limb lameness.

This thesis included 193 horses that were recorded with the camera-based optical motion capture system (Q-horse by Qualisys) during routine lameness examinations by the veterinarians at four equine hospitals in Europe. Horses were included if they had an initial head or pelvic asymmetry above thresholds (HDmax and HDmin >|15|mm or PDmax and PDmin >|7|mm) that was reduced by at least 70% by diagnostic analgesia. Furthermore, they were divided into different groups depending on if they were considered lame on one or several limbs, or if they had a deviating movement pattern making them suspected to be multi limb lame.

The result of the thesis shows no clear indicators of multiple limb lameness but gives indications that a horse with an ipsilateral forelimb and hindlimb lameness either can appear as a forelimb lame horse with a head and wither asymmetry indicating different forelimbs or as a hindlimb lame horse with a head and wither asymmetry indicating the same forelimb. The result also shows that a bilateral forelimb horse in some cases also have a head and wither asymmetry indicating different forelimbs as well as a bilateral hindlimb lame horse in some cases have a head and wither asymmetry indicating the same forelimb. It is though uncertain whether it is the bilateral lameness causing the deviating head and wither asymmetry pattern or another (third) lameness.

This thesis is limited by the small study population and the difficulties to ensure that the horses only have the expected lameness, especially since the movement pattern of multiple limb lame horses is largely unknown. The possibility that the horses are lame in other limbs as well is a cause to possible errors in the study, since it can result in a misinterpretation of the movement pattern and erroneously comparing between the movement pattern of horses with lameness on one and multiple limbs.

Keywords: multiple limb lameness, gait analysis, motion asymmetry, lameness assessment

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### Abbreviations

RF	Right forelimb
RH	Right hind limb
LF	Left forelimb
LH	Left hindlimb
HDmax	Head maximum difference
HDmin	Head minimum difference
WDmax	Withers maximum difference
WDmin	Withers minimum difference
PDmax	Pelvis maximum difference
PDmin	Pelvis minimum difference
SLU	Swedish University of Agricultural Sciences

#### 1. Introduction

#### 1.1 General introduction

Disorders from the locomotion apparatus, where lameness is a common symptom, is the most common disease category in horses (Penell et al. 2005; Agria Djurförsäkring 2021) and a common cause for euthanasia (Egenvall et al. 2006). The disorder causing lameness needs to not only be correctly diagnosed, but the lameness needs to be detected in the first place. Lameness detection is difficult for both owners (Dyson & Greve 2016) and for veterinarians, especially when inexperienced and/or when the lameness is mild (Keegan et al. 2010).

The difficulties in detection of lameness are not only caused by limitations in the human ability to detect asymmetries (Parkes et al. 2009) but also because of limitations in the sensitivity of speed changes in the visual system (Haarmeier & Thier 2006; Holcombe 2009) and because of bias (Arkell et al. 2006). Additionally, the complexity of orthopaedic diseases and biomechanics further complicates to interpret the movements correctly.

A horse can be lame in one or multiple limbs, where the most obvious lameness is considered the primary lameness (Stashak & Baxter 2020). A lameness in one limb can create a false lameness in another limb due to adaptations in the movement pattern to decrease the loading of the affected limb. This lameness adaptation is called compensatory lameness and is considered false since it disappear when the primary lameness is eliminated (Uhlir et al. 1997; Maliye et al. 2015). A compensatory lameness can present as more severe than the primary lameness, making it more complicated to detect the true lameness (Kramer et al. 2004).

When a horse has a multiple limb lameness, the lameness in the other limbs than the considered primary lame limb is often referred to as a secondary lameness (Ross 2011a). A multiple limb lameness can result in a symmetric movement when the horse is trotting in a straight line. In such cases lungeing can be a helpful manner to determine which leg is lame according to the author (Stashak & Baxter 2020), but one needs to take into account the asymmetries emerging naturally because of the circular movement (Rhodin et al. 2013, 2016). The movements of sound and lame horses are investigated in several studies (May & Wyn-Jones 1987; Buchner et al. 1996a; b; Kramer et al. 2004; Rhodin et al. 2018) and the complementary mechanisms are well described (Uhlir et al. 1997; Kelmer et al. 2005; Maliye et al. 2015). Though common that horses have orthopaedic disorders in several limbs, few studies describe the movement pattern of horses with multiple limb lameness. More research is needed to better understand the complexities of the locomotion apparatus and to become better at finding and diagnosing orthopaedic disorders to improve the welfare of our horses.

#### 1.2 The aim of the study and our hypothesis

The aim of this study was to investigate the movement pattern of horses with multiple limb lameness to develop a better knowledge and understanding of the locomotion apparatus and the mechanisms of lameness. Our hypothesis was that it is possible to detect a difference in the initial movement pattern of a horse with multiple limb lameness compared to a horse with unilateral limb lameness when using an objective motion analysis system.

#### 2. Literature review

#### 2.1 The limb cycle, strides, and gaits

As the horse moves forward each of the four limbs will go through a limb cycle, including a stance phase, when the hoof has contact with the ground (Barrey 1999). When all four limbs have gone through a limb cycle the horse has taken one stride forward. Strides can be combined in different coordinated and automatic patterns, called gaits, to create the most energy efficient motion in different speeds. In some gaits the strides also include a third phase called suspension phase. During this phase all four limbs will be in swing phase, meaning no hoof has ground contact. Depending on hoof timing and the median plane of the horse, gaits can be divided into symmetric such as walk and trot or asymmetric such as gallop and canter. Different gaits also have varying beats depending on the number of steps (hoof strikes) during each stride (Ross 2011b).

Walk is a symmetric, four-beat gait characterized by all four hoofs being placed independently of each other (Ross 2011b). During walk there is always at least one leg in stance phase and thus no suspension phase.

Trot is a symmetric, two-beat gait characterized by two limbs moving simultaneously and striking the ground at the same time (Ross 2011b). In trot the diagonal pair of limbs move simultaneously (LF and RH respectively RF and LH). A suspension phase occurs between the stance phase of the diagonal pair of limbs. While the horse moves forward the trunk will have a vertical movement as the horse pushes off with and lands on the limbs (Buchner et al. 1996a). The movement can be measured by tracking the head, withers, and tuber sacrale. During trot these tracking points have a sinusoidal pattern (see Figure 1). Shortly after push-off of one diagonal pair of limbs the tracking points will reach their maximal height above ground. Thereafter they will move downward, reaching their minimal height above ground during midstance and then they will move upward once again. For the other diagonal pair of limbs, a symmetric oscillation occurs in a similar manner, which means that the tracking points will reach maximum and minimum positions twice during one stride. When a horse is sound the maximum and minimum positions twice during one stride are equal, resulting in a symmetric movement pattern (Kramer et

al. 2004). The vertical velocity as well as the vertical acceleration of the body has the same sinusoidal pattern as the vertical movement (Buchner et al. 1996a), where the maximal acceleration occurs close to the point when the body reach minimum height above ground.

The movement of the pelvis can also be measured at tuber coxae and the vertical movement of each one is sinusoidal as well (May & Wyn-Jones 1987; Buchner et al. 1993) although slightly asymmetrical. The displacement amplitude of tuber coxae on one side is greater during the stance phase of the contralateral limb than of the same side limb (Buchner et al. 1996a; Kramer et al. 2004). This indicates that the pelvis is not only moving vertically but also rotating along the longitudinal axis. In a study Faber et al. (2000) showed that the vertebra rotates toward the limb at stance.



Figure 1. Illustration of the vertical movement of the head, withers and/or pelvis that follows a sinusoidal pattern at trot.

#### 2.2 Lameness

#### 2.2.1 Definition of lameness

There is not a unanimous definition of lameness, but different wordings exist. Altogether lameness is considered a gait abnormality as a symptom from an underlaying cause such as a structural or functional disorder, pain in the limbs, or in the axial skeleton (Ross 2011a; Stashak & Baxter 2020). This underlaying cause is suggested to be inflammation or pain, as well as a mechanical defect. Furthermore, new technologies and objective measurements of locomotion has resulted in findings of asymmetric motion in horses considered sound by owners, trainers and clinicians which challenges and stretches these definitions (Van Weeren et al. 2017; Persson-Sjodin et al. 2019; Kallerud et al. 2021; Zetterberg et al. 2023).

#### 2.2.2 Classification of lameness

Lameness can be classified as primary or secondary (Ross 2011a:2). According to the author the most obvious lameness when the horse is observed in walk or trot is normally considered the primary lameness. Secondary lameness is evident in a limb other than the primary either during initial examination or after manipulation or diagnostic analgesia. Stashak and Baxer (2020) suggest that a secondary lameness often emerges as a result of uneven distribution from the primary lameness, causing an overloading of the other limbs.

Moreover, hindlimb lameness can be classified after when it emerges in the stride cycle as impact or push-off lameness (Bell et al. 2016). A hindlimb lameness is considered an impact lameness when the minimum position of the pelvis is higher due to a lower peak of the vertical ground reaction force during the first half of the stance. A push-off lameness is considered when the maximum position of the pelvis is lower due to a lower vertical ground reaction impulse in the second half of the stance because of a transition from vertical to horizontal ground reaction impulse. There can also be a mix of push-off and impact lameness (Phutthachalee et al. 2021).

#### 2.3 Mechanisms of lameness

Studies have shown that the most consistent factors to detect lameness are the vertical motion, velocity and acceleration of the head (during forelimb lameness) and pelvis (during hind limb lameness) (May & Wyn-Jones 1987; Buchner et al. 1996a). Lameness will also affect factors such as joint extension during stance, joint flexion during swing, hoof height during swing, limb protraction and step length (Buchner et al. 1996b; Kramer et al. 2000).

#### 2.3.1 Load redistribution

When a lame horse is trotting the maximum acceleration and vertical velocity of the body (tracked at the head, withers, and pelvis) will decrease during stance phase of the lame limb and increase on the sound contralateral limb (Buchner et al. 1996a). According to Newton's second law of motion the acceleration is related to the force, which indicates that the decrease in acceleration results in a reduction of

force on the lame limb. Two studies (Weishaupt et al. 2004, 2006) have investigated the load redistribution and movement adaptions during forelimb and hindlimb lameness. They found that the peak vertical ground reaction force decreased in the limb with induced lameness. The researchers described the mechanisms which lead to reduction of force on the affected limb as follows:

- Altering cadence: for both forelimb and hindlimb lameness the stride duration decreases while the step frequency increases.
- Asymmetric transition: At transition from the lame to the sound diagonal the transition time between the contralateral limbs and the suspension duration is shortened. This makes it possible for the sound diagonal to support the body earlier.
- Redistribution of load within the diagonal: When a horse is lame the load shifts along the longitudinal axis. When a horse is lame in a hindlimb the load is being shifted to the forelimb during the lame diagonal stance and to the hindlimb if the horse is lame on a forelimb.
- Prolongation of stance duration: The stance duration of the affected and the contralateral limb is prolonged for both forelimb and hindlimb lameness. The prolongation of stance duration is mainly counteracting the compensatory increased load of the contralateral limb.

#### 2.3.2 Vertical displacement

When a horse is lame their vertical movement pattern will change and become asymmetrical between the stride halves. Studies have shown that these changes in vertical motion of the head and withers (during forelimb lameness) and pelvis (during hind limb lameness) is some of the most consistent factors for lameness detection (May & Wyn-Jones 1987; Buchner et al. 1996a).

When a lame horse is trotting the downward movement of the body as well as the upward movement after push off will be less during the stance phase on the lame limb (Buchner et al. 1996a; Kramer et al. 2004; Rhodin et al. 2018). This results in a higher minimum position and a lower maximum position relative to the ground for the head, withers, and pelvis (see Figure 2). It is also shown that while the displacement amplitude decreases on the lame limb, it increases on the sound contralateral limb for both forelimb and hindlimb lameness (Buchner et al. 1996a).

A classic expression about forelimb lameness detection is "*nod on sound*" yet this is not fully correct. The head and withers will reach a less descendent position during stance on the lame forelimb in trot compared to the sound limb, where the head displacement amplitude is more pronounced than the withers. (Buchner et al. 1996a; Rhodin et al. 2018). The minimum position of the head and withers relative to the ground will be lower during stance on the sound limb compared to the stance on the lame limb. This causes the illusion of the horse nodding on sound.

When a horse is lame on a hindlimb there will be a greater vertical motion of the tuber coxae on the lame side during stance phase of the contralateral limb, while the vertical motion will decrease during stance phase of the lame limb. This results in an increased asymmetric movement on the lame side, which indicates an increased rotation of the pelvis during lameness. In contrast the asymmetric movement of tuber coxae will decrease on the sound side (Buchner et al. 1996a). The higher position of the pelvis after push-off from the sound limb (and the higher position of the tuber coxae on the lame side after push-off from the contralateral limb) is described sometime in literature as "*hip hike*" (Kramer et al. 2004; Stashak & Baxter 2020). Likewise the lower maximum position of the pelvis after push-off from the lame limb is sometimes described in literature as "*hip dip*" (Stashak & Baxter 2020)

The asymmetries in displacement amplitude are found to be more obvious in the pelvis during hindlimb lameness than in the withers during forelimb lameness (Buchner et al. 1996a). It is suggested that due to such observations, the changes in head motion can help unload the forelimbs through creating an upward energy and cause a caudal shift of the body mass. The caudal part of the body does not have the same lever arm as the cranial part (the head). Therefore, the horse might use the rotation in the pelvis to create more upward energy after push-off from the sound limb in order to reduce the energy needed to lift the trunk during push-off from the lame limb.



Figure 2. Illustration of when the maximum position is lower, and the minimum position is higher for the lame diagonal compared to the sound diagonal creating an asymmetric pattern.

#### 2.3.3 Compensatory lameness

When a horse adapts its movement to decrease the loading of the primary lame limb, the movement pattern can mimic a lameness in another limb, which is called a compensatory lameness. It is not caused by pain and disappears when the underlying cause is eliminated (i.e. when the induced lameness is reversed) or after diagnostic analgesia (Uhlir et al. 1997; Maliye et al. 2015). Generally a compensatory lameness can be visible in the contralateral hindlimb in trot during a primary forelimb lameness and in the ipsilateral forelimb during a primary hindlimb lameness (May & Wyn-Jones 1987; Uhlir et al. 1997; Kramer et al. 2004; Rhodin et al. 2018). However, it has been shown that compensatory lameness emerging from a primary forelimb lameness can emerge in the ipsilateral hindlimb as well (Kelmer et al. 2005; Rhodin et al. 2013).

When the lame hindlimb is in stance phase, the diagonal forelimb will be as well. To unload the hindlimb and shift weight to the cranial part of the body the horse will lower the head during impact of the forelimb. The head will then be lower during stance phase of the diagonal forelimb compared to the ipsilateral forelimb, mimicking a ipsilateral forelimb lameness (Buchner et al. 1996a; Weishaupt et al. 2004).

The compensatory lameness in a hindlimb as a result of a primary forelimb lameness seems to emerge at a higher degree of severity of the primary lameness compared to the opposite case (Uhlir et al. 1997). In a study (Kelmer et al. 2005) where lameness was induced to assess the compensatory mechanism they found that doubling the primary hindlimb lameness increased the compensatory head movement by 50% while doubling the primary forelimb lameness only increased the compensatory pelvis movement by 5%. In the same study they found that mild to moderate hindlimb lameness induces significant compensatory head movement, while the same degree of forelimb lameness only induced a small or non-visible compensatory pelvis movement.

Compensatory movements makes the lameness assessment even more complicated and studies have indicated that veterinarians find it challenging to interpret them correctly (Stenius 2022). The compensatory lameness in a front limb can be equivalent to the primary hindlimb lameness in the matter of displacement amplitude, making it hard to differentiate which one is the primary lameness (Rhodin et al. 2013). A study of Hammarberg et al. (2016) found that the interagreement between veterinarians was especially low for hindlimb lameness and suggest it could be due to the veterinarians interpreting the compensatory forelimb lameness as the true lameness. In a recent study (Persson-Sjodin et al. 2023) the vertical displacement amplitude of 317 clinically lame horses was analysed. They found that the movement of the withers and head can help to locate the primary lameness. The result showed that 80-81% of horses with primary forelimb lameness had asymmetries in head and withers displacement amplitude that indicated the same forelimb, while 69-72% of horses with primary hindlimb lameness had asymmetries that indicated different forelimbs (the head asymmetry indicated the ipsilateral forelimb, and the wither asymmetry indicated the diagonal forelimb).

#### 2.3.4 Lunging asymmetry

Evaluating the horse during lungeing is part of the movement analysis; but the circular movement will naturally affect the movement pattern, making it more asymmetric even in sound horses. The asymmetries are not always consistent between turns and horses, and can sometimes be seen in one direction but not in the other (Rhodin et al. 2013, 2016). For the hindlimbs, it is common with a less downward movement of the pelvis during stance phase on the inner hindlimb, giving the illusion of an inner limb impact lameness (Rhodin et al. 2013, 2016); similarly, it is common with a less upward movement of the outside hindlimb, giving the illusion of an outside limb push-off lameness (Rhodin et al. 2016). The inside hindlimb may be more affected by the tilt due to circular movement than the outside limb (Rhodin et al. 2013).

For the forelimbs, the movement is more inconsistent than in the pelvis. In one study the illusion of an outside forelimb lameness was more common (57 of 94 horses in left rein and 54 of 94 horses in right rein) than an inside lameness (37 of 94 on left rein and 40 of 94 on right rein) (Rhodin et al. 2016).

#### 2.3.5 Normal variation and laterality

In theory the sound horse moves symmetrically in trot while the lame moves asymmetrically. However, several studies have shown that horses seemingly sound have an asymmetrical motion pattern when their movement is analysed with objective assessment systems. In a study (Zetterberg et al. 2023) the movement of 54 foals (21 Swedish warmblood and 23 Standardbred) in age between 4 and 13 weeks were analysed. The result showed that 83% of the Standardbred foals and 45% of the Swedish warmblood foals had parameters that exceeded the established threshold for asymmetry when trotting on a straight line. In another study (Kallerud et al. 2021) 114 Standardbred yearlings, considered fit by their trainers, were evaluated. 93% had parameters above asymmetry threshold during in hand trials and 94% during track trials. The majority showed mild asymmetries and a minority switched side of the asymmetry for one or more parameters between in hand and track trials. Values for vertical motion parameters above threshold for asymmetry in horses considered sound where also found in another study (Rhodin et al. 2017) where 72.5% of 222 warmblood type riding horses where evaluated when trotting in straight line.

To try to understand the causes of these asymmetries, 65 young warmblood riding horses, assessed as sound by owners, were evaluated together with a rider questionnaire about perceived sidedness (Leclercq et al. 2023). A subgroup of 40 horses also participated in a preference test. The horses were recorded when eating from a bucket on the ground and the protracted forelimb was analysed. The study did not show a significant correlation between limb preferences and vertical asymmetry; 22% of the horses showed limb preferences while the vertical asymmetries presenting was two times higher. They could neither find a significant correlation between asymmetries and questionnaire responses. Therefore, the authors discuss the possibility that the rider's sidedness could be influenced by their own laterality or physical limitation instead of the horses'.

In a study by Persson-Sjodin et al. (2019) they investigated whether the movement asymmetry in riding horses considered sound by owners was affected by antiinflammatory treatment with meloxicam. They had 66 horses included in the study; all were in full training, had not been treated for lameness within two months from the data collection, and showed asymmetries greater than 6mm for the head movement or 3mm for the pelvic movement. The horses were given treatment with meloxicam and placebo for four days respectively with 14-16 days between the different treatments. They were furthermore evaluated during trot at hand on straight line and during lungeing as well as on hard and soft surface before treatment and on day 4 after treatment. No significant effect of the treatment with meloxicam was found, which could indicate that the asymmetries may emerge in the absence of pain due to biological variation, but it could also be due to pain by a cause that is nonresponsive to treatment with meloxicam.

#### 2.4 Lameness examination

A lameness examination includes several parts to get a comprehensive picture of the problem to be able to correctly diagnose the disorder and institute the correct treatment (Stashak & Baxter 2020). The examination includes taking information about signalment, use and an anamnesis. The anamnesis is the medical history, including information such as symptoms, when they occurred and if they have gotten better or worse, if they change through the day or after specific activities and information about earlier treatments and results. Furthermore, it includes examination of the body from a distance where the veterinarian observes the horse from all directions and take notice to conformation, body condition, posture, and symmetry (muscle mass, swelling) as well as palpation of skeleton, joints, and muscles.

The assessment of the gait is an important part of the examination and trot is a good gait for analysing the motion (Buchner et al. 1996a; Stashak & Baxter 2020). The horse is assessed moving on a straight line as well as in a circular motion during lungeing, both on hard and soft surfaces. The veterinarian observes the horse running and makes a subjective assessment of the movement patterns (Stashak &

Baxter 2020). Furthermore, the veterinarian will make use of a subjective scoring method to describe the lameness. A common lameness scale is created by the AAEP (American Association of Equine Practitioners) (AAEP n.d.). Since the AAEP scoring scale is a mix of both walk and trot, Mike Ross presented another 5-score method based only on the movement pattern in trot (Ross 2011b).

To be able to locate the lameness the veterinarian can perform manipulation tests and use local anaesthetics by perineural infiltration, ring blocks or intrasynovial injections (Stashak & Baxter 2020). When lameness is detected, diagnostic imaging such as radiography or ultrasound is often an important tool to make the correct diagnosis.

#### 2.4.1 Objective gait assessment

There are two perspectives of biomechanics, kinetics, and kinematics, and for both there are different objective measuring systems available. Kinetics is the study of the forces generated by musculoskeletal work (Serra Bragança et al. 2018). The stationary force platforms are considered the 'gold standard' for kinetic lameness assessment that for example measures the ground reaction force. Another technique involves a treadmill with integrated force plates and pressure plates.

Kinematics is the study of movement of body segments, and they can furthermore be described in relation to the surroundings or in relation to each other with concepts such as displacement, velocity, acceleration, or angles (Serra Bragança et al. 2018). The optical motion capture technique is considered the golden standard for kinematic lameness assessment, but inertial sensor-based systems can also be used.

Some optical motion capture techniques are based on the use of several cameras positioned around a calibrated measuring volume that track the position of reflective markers mounted on different segments of the horse's body (Roepstorff et al. 2021). The recorded tracking positions are thereafter used to calculate different vertical movement measurements. Some systems are based on markerless techniques that through computer vision and deep learning track different body segments (Lawin et al. 2023). Inertial sensor-based systems use accelerometers and a gyroscope mounted on the horse's body (at the head, pelvis and on one or multiple limbs) and record data about vertical acceleration and angular velocity. The data is furthermore used to determinate stride timing of stance and swing phase of the limbs and analyse the vertical movement (Keegan et al. 2011).

#### 3. Materials and methods

#### 3.1 Study protocol

This study is an observational study including a retrospective and a prospective part. Objective movement data were collected during routine lameness examinations and were recorded with a camera-based optical motion capture system (Qualisys).

The prospective part took place at the University Equine Clinic, located at the Swedish University of Agricultural Sciences in Uppsala between September and October 2023. Horses with initial head or pelvic asymmetry above thresholds (HDmax and HDmin >|15|mm or PDmax and PDmin >|7|mm) for one single limb, which was reduced by 150% after diagnostic analgesia, were collected for further analysis.

The retrospective part involved horses that presented for lameness examination at the University Equine Clinic at the Swedish University of Agricultural Sciences in Uppsala, Sweden; Evidensia Specialist Equine Hospital in Helsingborg, Sweden; the University Equine Clinic at Utrecht University, the Netherlands, the Equine Clinic in Lüsche, Germany between October 2015 and November 2020. Horses with an initial head or pelvic asymmetry above thresholds (see above) for one single limb, which was reduced by at least 70% after diagnostic analgesia, were collected for further analysis.

Furthermore, all the horses included in the study were divided into groups depending on whether they were considered unilaterally lame or if they were considered, or suspected, to be multi limb lame. The groups with unilaterally lame horses were used as a comparison to the horses in the multi limb lame groups.

#### 3.2 Data collection and motion recordings

For motion recordings a camera-based optical motion capture system (Oqus 7+ or Oqus 400, Qualisys AB, Gothenburg, Sweden) was used. During the recording, the horses were wearing reflective markers, each at the size of 25mm, mounted to the body with double-sided tape. One reflective marker was placed on the highest point

of the head between the ears (not used on all horses), three markers in the midline of the forehead, three at the withers (one in sagittal plane on the highest point and two laterally of the middle one), three at the pelvis (one in sagittal plane between the two *tuber sacrale* and one on the left and right *tuber coxae* respectively) and one on the right carpus.

The markers positions were consistent for every recording for each horse. The position of the reflective markers was recorded with infrared cameras mounted in the roof angle around the calibrated area making it possible for the cameras to register a three-dimensional position. The data was collected at 100-200Hz when the horses were trotting in a straight line on a hard asphalted surface. They were recorded before any diagnostic analgesia was administered to set a baseline, and then they were recorded once again after every diagnostic analgesia.

#### 3.3 Data processing

After the motion recordings, the recorded three-dimensional positions of the reflective markers were extracted from Qualisys Track Manager, which is the motion capture software. The position data was then exported to MATLAB, which filters the data and runs an algorithm that recognises the movement pattern when the horse is trotting. It then segments the motion data into steps based on peaks in the vertical position of the pelvis (Roepstorff et al. 2021). Furthermore, vertical movement parameters were calculated for the different tracking points (see Figure 3):

- Maximum difference (max. diff): the difference between the maximum positions. Positive values indicate the stance phase of the right limb and negative values indicates the stance phase of the left limb. Max. diff was calculated for the head (PDmax), the withers (WDmax) and the pelvis (PDmax).
- Minimum difference (min. diff): the difference between the minimum positions. Positive value indicate stance phase for right limb and negative value indicates stance phase of left limb. Min. diff was calculated for the head (PDmin), the withers (WDmin) and the pelvis (PDmin).

The variables used are positive for right sided asymmetry and negative for left sided asymmetry. To facilitate statistical analysis the asymmetry parameters were multiplied by -1 for all horses with left-sided lameness.



Figure 3. Illustration of how Min. diff and Max. diff is calculated.

#### 3.4 Data analysis

As seen in Figure 4, the horses were separated into groups as follows:

- Group 1: Horses with HDmax/HDmin/PDmax/PDmin initially above thresholds, decreasing with 150% after diagnostic analgesia. The horses in this group are considered bilaterally lame in the fore- or hindlimbs. This group was furthermore divided into subgroups, 1a and 1b, depending on if the horses were fulfilling the criteria for forelimb or hindlimb lameness. Horses were excluded if HDmax/HDmin and PDmax/PDmin respectively indicated different limbs.
- Group 2: Horses with hindlimb lameness (e.g., PDmax/PDmin initially above threshold, decreasing >70% after diagnostic analgesia) where head and withers asymmetry indicates lameness in the same forelimb. The horses in this group are suspected to be multi limb lame. The horses in this group were divided into subgroups, 2a and 2b, depending on whether they fulfilled the inclusion criteria for PDmax or PDmin. It was not possible to create a group with all horses with PDmax and PDmin on the same limb since most of the horses fulfilling the inclusion criteria for PDmax had PDmin on the contralateral limb, hence two subgroups were created.
- Group 3: Horses with forelimb lameness (e.g., HDmax/HDmin initially above threshold, decreasing with >70% after diagnostic analgesia) with head and withers asymmetry indicating a lameness in different forelimbs. These

horses were suspected to be multi limb lame. The horses with the HDmin and HDmax indicating the same limb were included in subgroup 3a, while the rest were excluded.

Group 4: Horses with HDmax/HDmin/PDmax/PDmin initially above thresholds, decreasing with 70-100% after diagnostic analgesia. These horses were considered unilateral lame. The horses in this group were divided into subgroup, 4a and 4b, depending on whether the horses were fulfilling the criteria for forelimb (HDmax/HDmin) or hindlimb (PDmax/ PDmin) lameness. The horses with HDmax/HDmin and PDmax/PDmin respectively indicating different limbs were excluded. To create a reference group of unilaterally lame horses with the expected head and wither pattern to use as a comparison against both the forelimb lame horses with a head and wither asymmetry pattern indicating different forelimbs (group 3a), as well as the hindlimb lame horses with head and wither asymmetry indicating the same forelimb (subgroup 2a and 2b), the subgroups 4c, 4d and 4e were created. Subgroup 4c includes horses fulfilling the inclusion criteria for HDmax and HDmin, where HDmax and HDmin indicates the same forelimb, and with a head and wither asymmetry pattern indicating the same forelimb. Subgroup 4d includes horses fulfilling the criteria for PDmax and subgroup 4e includes horses fulfilling the criteria for PDmin, where all horses have a head and wither indicating different forelimbs.

Horses can be included in several groups, but never in two groups that are compared to each other. The horses in group 1a can be included in group 3a as well as horses in group 1b can be included in group 2a or 2b. Horses from group 4a can also be included in group 4c as well as horses in group 4b can be included in group 4d or 4e.

No horses from the prospective part of the study were included since none fulfilled the inclusion criteria.



Figure 4. Illustration of the group division and number of horses in each group and subgroup. The same horse can be included in more than one group.

#### 4. Results

#### 4.1 Meta data

Of the 1963 horses included in the study, 85 were geldings, 79 were mares, 7 were stallions and 22 of unknown gender. The ages of the horses at the time of the visit were in the range of 2 to 32 years with a mean age of 10.8 years. For 17 horses the age was not recorded. The horse population is comprised of 113 Warmblood type horses, 15 ponies, 8 Quarter horses, 5 Icelandic horses, 25 of other breeds, and 27 of unknown breed.

The mean of strides used for the analysis were for Group 1: 20, Group 2: 23, Group 3: 21, and for Group 4: 22. The variation of the amplitude of asymmetry is shown in Figure 5 for each subgroup, the mean asymmetry is in the range 22.6-39.5 for hindlimb lameness and 10.6-16.8 for hindlimb lameness.



Figure 5. Boxplot of the amplitude of asymmetry for the variables (HDmax/HDmin/PDmax or PDmin) for which the horse fulfils the inclusion criteria.

#### 4.2 Group 1

#### 4.2.1 Bilateral forelimb lameness (Group 1a)

#### Head and wither asymmetry

In both the groups with bilaterally and unilaterally forelimb lame horses, some of the horses had a head asymmetry as well as a wither asymmetry indicating lameness in the lame forelimb, while some horses had a head asymmetry indicating lameness in the lame forelimb and a wither asymmetry indicating lameness in the contralateral forelimb.

Table 1. The table is showing the head and wither asymmetry pattern and the pelvic asymmetry pattern for horses with bilateral forelimb lameness (group 1a) and for unilaterally forelimb lame horses (group 4a). This table only takes notice to the polarity of the asymmetry (e.g. what limb) and not the magnitude. The pelvic asymmetry is ipsilateral or diagonal to the primarily lame forelimb. The table shows the frequency before and after diagnostic analgesia.

HDmin+	WDmin			Before			After		
	Forelimb			Ipsilateral	Diagonal	Both	Ipsilateral	Diagonal	Both
		#	%	%	%	%	%	%	%
Group 1a	Same	10	62.5	0	70	30	10	50	40
Group 4a	Same	70	88.6	11	53	38	24	37	40
Group 1a	Different	6	37,5	50	0	50	100	0	0
Group 4a	Different	9	11.4	44	33	22	55	33	11

There was a slightly higher percentage among the horses with unilateral forelimb lameness that had a head and wither asymmetry pattern that indicates the same forelimb compared to the horses with bilateral forelimb lameness, as seen in Table 1. Among the horses with a bilateral forelimb lameness with a head and wither asymmetry pattern indicating the same forelimb, most of the horses had a diagonal hindlimb asymmetry to the lame forelimb before diagnostic analgesia and none have only an ipsilateral hindlimb asymmetry. Among the unilaterally forelimb lame horses with a head and wither asymmetry pattern indicating the same forelimb, there was a lower percentage of horses with a diagonal hindlimb asymmetry before diagnostic analgesia and a few horses with an ipsilateral hindlimb asymmetry. Among both the bilaterally and unilaterally forelimb lame horses there was a higher occurrence of an ipsilateral hindlimb asymmetry to the primary lameness after diagnostic analgesia compared to before.

Among both bilaterally and unilaterally forelimb lame horses with a head and wither asymmetry pattern indicating different forelimbs, there was a higher percentage of horses with an ipsilateral hindlimb asymmetry to the lame forelimb compared to the horses with a head and wither asymmetry pattern indicating the same forelimb. Among the bilaterally forelimb lame horses with a head and wither asymmetry pattern indicating different forelimbs, none had a diagonal hindlimb asymmetry to the lame forelimb before diagnostic analgesia while all of them had ipsilateral hindlimb asymmetry to the primary forelimb lameness afterward. The variation of frequency of an ipsilateral or diagonal hindlimb asymmetry was greater among horses with unilateral forelimb lameness.

#### Pelvic asymmetry in forelimb lame horses

As seen in Table 2, the frequency of diagonal hindlimb lameness with a pelvic asymmetry above thresholds before diagnostic analgesia was similar in the group with bilaterally and unilaterally forelimb lame horses, as well as the frequency of horses with ipsilateral hindlimb lameness to the lame forelimb. All bilaterally and unilaterally lame horses with a diagonal hindlimb pelvic asymmetry to the primary lame forelimb had a head and wither asymmetry pattern indicating the same forelimb, while all horses among the bilaterally forelimb had a head and wither asymmetry below as with an ipsilateral pelvic asymmetry to the primary lame forelimb had a head and wither and wither asymmetry pattern indicating different forelimbs. In the group of horses with unilateral forelimb lameness and an ipsilateral pelvic asymmetry both kinds of head and wither asymmetry pattern were evident.

Table 2. The table is showing the frequency of horses with pelvic asymmetry above threshold before diagnostic analgesia for at least one parameter (PDmax/PDmin) and if they have asymmetry in the ipsilateral or diagonal hindlimb to the lame forelimb, as well as their head and wither asymmetry pattern. The table show the result for bilateral forelimb lame horses (group 1a) and unilateral forelimb lame horses (group 4a).

	Diagonal	Head and wither	Ipsilateral	Head and wither
		pattern		pattern
	% (#)		% (#)	
Group 1a	43.8 (7)	Same forelimb	12.5 (2)	Different forelimbs
Group 4a	40.5 (32)	Same forelimb	11.4 (9)	89% same/
				11% different forelimb

Most of the bilaterally and unilaterally forelimb lame horses had a pelvic asymmetry for both PDmax and PDmin for the same hindlimb, whereas the majority had a pelvic asymmetry for the diagonal hindlimb to the lame forelimb (see Table 3).

Table 3. The table shows the pattern of the pelvic movement for bilaterally forelimb lame horses
(group 1a) and unilaterally forelimb lame horses (group 4a) if the values of PDmax and PDmin
indicate asymmetry in the same or different hindlimbs. The table also show if the pelvic asymmetry
is ipsilateral or diagonal to the lame forelimb.

	PDmax+		Hindlimb	
	PDmin		Ipsilateral	Diagonal
	Hindlimb	% (#)	% (#)	% (#)
Group 1a	Same	68.8 (11)	18.8 (3)	50.0 (8)
Group 4a	Same	65.8 (52)	15.2 (12)	50.6 (40)
Group 1a	Different	31.2 (5)		
Group 4a	Different	34.2 (27)		

#### 4.2.2 Bilateral hindlimb lameness (Group 1b)

#### Head and wither asymmetry

As seen in Table 4, there was a higher frequency of bilaterally hindlimb lame horses with a head and wither asymmetry indicating different forelimbs compared to horses with a unilateral hindlimb lameness.

Table 4. The table show the frequency of the head and wither asymmetry pattern among the horses with bilateral hindlimb lameness (1b) and unilateral hindlimb lameness (4b), as well as if the head movement indicates an asymmetry in the ipsilateral or diagonal forelimb to the lame hindlimb. This table only take notice to the polarity of the asymmetry (e.g., what limb) and not the magnitude.

	HDmin+ WDmin		Forelimb	
			Ipsilateral	Diagonal
	Forelimb	% (#)	% (#)	% (#)
Group 1b	Same	25.0 (3)	8.3 (1)	16.7 (2)
Group 4b	Same	42.6 (26)	3.3 (2)	39.3 (24)
Group 1b	Different	75.0 (9)		
Group 4b	Different	57.4 (35)		

#### Forelimb asymmetry in hindlimb lame horses

As seen in Table 5. The table shows the frequency of horses with head asymmetry above threshold before diagnostic analgesia for at least one parameter (HDmax/HDmin) and if the head asymmetry is ipsilateral or diagonal to the lame was a higher frequency of horses with bilateral hindlimb lameness with ipsilateral forelimb lameness compared to horses with diagonal forelimb lameness. It was also observed a higher frequency of bilaterally hindlimb lame horses with an ipsilateral forelimb lameness compared to the frequency among the horses with unilateral hindlimb lameness.

Table 5. The table shows the frequency of horses with head asymmetry above threshold before diagnostic analgesia for at least one parameter (HDmax/HDmin) and if the head asymmetry is ipsilateral or diagonal to the lame hindlimb. Notice of the head and wither asymmetry is only taken to the polarity of the asymmetry (e.g., what limb) and not the magnitude. The table shows the result for horses with bilateral hindlimb lameness (group 1b) and unilateral hindlimb lameness (4b).

	Diagonal	Head and wither	Ipsilateral	Head and wither
		pattern		pattern
	% (#)		% (#)	
Group 1b	0.0 (0)	-	41.7 (5)	Different forelimbs
Group 4b	9.8 (6)	Same forelimb	27.9 (15)	24.6% different/
				3.3% same forelimb

As seen in Table 6, most of the horses had a head asymmetry ipsilateral to the lame hindlimb in both groups. The variation of HDmax and HDmin indicating the same or different forelimbs was similar between the groups.

Table 6. The table shows the pattern of the head asymmetry among horses with bilateral hindlimb lameness (group 1b) and with unilateral hindlimb lameness (group 4b), if the values of HDmax and HDmin indicate asymmetry in the same or different forelimbs. The table also shows if the head asymmetry is ipsilateral or diagonal to the lame hindlimb. Notice is only taken to the polarity of the asymmetry (e.g., what limb) and not the magnitude.

	HDmax		Forelimb	
	+ HDmin		Ipsilateral	Diagonal
	Hindlimb	% (#)	% (#)	% (#)
Group 1b	Same	83.3 (10)	66.7 (8)	16.6 (2)
Group 4b	Same	78.7 (48)	49.2 (30)	29.5 (18)
Group 1b	Different	16.7 (2)		
Group 4b	Different	21.3 (13)		

#### 4.3 Group 2

# 4.3.1 Hindlimb lame horses (PDmax) with head and wither asymmetry pattern indicating the same forelimb (Group 2a)

#### Hindlimb lameness

Among the horses with hindlimb lameness and a head and wither asymmetry pattern indicating the same forelimb, the diagnostic analgesia resulted in a change of the lame side in 75% of the horses; the asymmetry value changed from being positive (right-sided) before diagnostic analgesia to becoming negative (left-sided) afterward. For one (25%) of these horses the negative value was close to zero and for one (25%) the initial lameness was reduced by 150% after diagnostic analgesia.

As seen in Table 7, there was a lower percentage of horses with hindlimb lameness for PDmax (group 2a) and a head and wither asymmetry pattern indicating the same forelimb, with a right-sided pelvic asymmetry for both PDmax and PDmin (both parameters indicated the same hindlimb) as well as a higher percentage of horses with PDmax and PDmin indicating different hindlimbs, compared to the other groups of hindlimb lame horses.

Table 7. The table shows the frequency of the pelvic asymmetry pattern before diagnostic analgesia if the values of PDmax and PDmin indicates asymmetry on the same hindlimb or different limbs. The table only take notice to the polarity of the asymmetry (e.g., what limb) and not to the magnitude. The table shows the result for horses with hindlimb lameness for PDmax (group 2a) and PDmin (2b) as well as a head and wither asymmetry pattern indicating the same forelimb. The table also shows the result for horses with hindlimb lameness for PDmax (group 4d) and PDmin (group 4e) with a head and wither asymmetry pattern indicating different forelimbs.

Group	2a	4d	2b	<b>4</b> e
PDmax +PDmin	% (#)	% (#)	% (#)	% (#)
Same hindlimb (right-sided asymmetry)	25.0 (1)	78.8 (26)	100.0 (3)	87.1 (27)
Different hindlimbs	75.0 (3)	21.2 (7)	0.0 (0)	12.9 (4)

#### Forelimb asymmetry in hindlimb lame horses

As seen in Table 8 the percentage of horses with forelimb lameness with a head asymmetry above threshold was similar between the groups. There was a higher occurrence of horses with a hindlimb lameness and a head and wither pattern indicating the same forelimb that also had an ipsilateral forelimb lameness compared to the hindlimb lame horses with a head and wither asymmetry indicating different forelimbs. For one of the horses with hindlimb lameness and a head and wither asymmetry pattern indicating the same forelimb, that also had an initial ipsilateral forelimb lameness, the forelimb asymmetry was just slightly reduced after diagnostic analgesia of the primarily lame hindlimb. None of the horses with a hindlimb lameness and a head and wither asymmetry pattern indicating the same forelimb had a diagonal forelimb lameness, which was a slightly lower occurrence compared to the hindlimb lame horses with a head and wither asymmetry pattern indicating different forelimbs.

Table 8. The frequency of forelimb lameness with a head asymmetry above threshold for at least one parameter (HDmax/HDmin) among the horses with hindlimb lameness for PDmax and a head and wither asymmetry indicating the same forelimb (group 2a) as well as among horses with hindlimb lameness for PDmax and a head and wither asymmetry pattern indicating different forelimbs (4d). The table show the frequency before and after diagnostic analgesia.

HDmax/HDmin	Before		After:	
Group	2a	4d	2a	4d
Forelimb	% (#)	% (#)	% (#)	% (#)
Ipsilateral	50.0 (2)	36.4 (12)	50.0 (2)	30.3 (10)
Diagonal	0.0 (0)	6.0 (2)	0.0 (0)	9.1 (3)
None	50.0 (2)	57.6 (19)	50.0 (2)	60.6 (20)

# 4.3.2 Hindlimb lame horses (PDmin) with head and wither asymmetry pattern indicating the same forelimb (Group 2b)

#### Hindlimb lameness

Among the horses with hindlimb lameness (for PDmin) and a head and wither asymmetry pattern indicating the same forelimb (group 2b), the diagnostic analgesia resulted in a change of the lame side in 33% of the horses since the asymmetry value was reduced with 150% after diagnostic analgesia.

As seen in Table 7, there was a slightly higher percentage of horses with PDmax and PDmin indicating the same hindlimb and a lower percentage of horse with PDmax and PDmin indicating different hindlimbs, compared to the horses with a unilateral hindlimb lameness and a head and wither asymmetry indicating different forelimbs.

#### Forelimb asymmetry in hindlimb lame horses

There was a higher occurrence of horses with an initial forelimb lameness with a head asymmetry above thresholds for at least one parameter (HDmax or HDmin) among the horses with hindlimb lameness and a head and wither asymmetry pattern indicating the same forelimb (group 2b) compared to the horses with unilateral hindlimb lameness and a head and wither pattern indicating different forelimbs

(group 4e), as seen in Table 9. Especially there was a higher frequency of horses with an initial ipsilateral forelimb lameness among the horses with a hindlimb lameness and a head and wither pattern indicating the same forelimb. For one of the hindlimb lame horses with an ipsilateral forelimb lameness, the lameness was just slightly changed after diagnostic analgesia of the primary lame hindlimb.

Table 9. The frequency of forelimb lameness with head asymmetry above threshold for at least one parameter (HDmax/HDmin) among the horses with a hindlimb lameness for PDmin and a head and wither asymmetry pattern indicating the same forelimb (group 2b) and horses with unilateral hindlimb lameness for PDmin with a head and wither pattern indicating different forelimbs (group 4e). The table show the frequency before and after diagnostic analgesia.

HDmax/HDmin	Before		After:	
Group	2b	4e	2b	4e
Forelimb	% (#)	% (#)	% (#)	% (#)
Ipsilateral	66.7 (2)	25.8 (8)	33.3 (1)	25.8 (8)
Diagonal	0.0 (0)	3.2 (1)	0.0 (0)	13.4 (6)
None	33.3 (1)	71.0 (22)	66.7 (2)	54.8 (17)

- 4.1 Group 3 Forelimb lame horses with head and wither asymmetry pattern indicating different forelimbs.
- 4.1.1 Forelimb lame horses with head and wither asymmetry pattern indicating different forelimbs (Group 3a)

#### Forelimb lameness

Among the horses with forelimb lameness and a head and wither asymmetry pattern indicating different forelimbs, the diagnostic analgesia resulted in a change of lameness to the contralateral side in 35% of the horses; the asymmetry value (HDmax or HDmin) changed from being positive (right-sided) before diagnostic analgesia to become negative (left-sided) afterward. In 30% of the horses in this group, the initial lameness was reduced by 150% after diagnostic analgesia. For the horses with bilateral forelimb lameness, the ipsilateral hindlimb asymmetry to the primary forelimb lameness was reduced after diagnostic analgesia except for one horse where it increased.

#### Hindlimb asymmetry in forelimb lame horses

There was a lower percentage of horses with an initial pelvic asymmetry above threshold in the group with forelimb lame horses with a head and wither asymmetry pattern indicating different forelimbs compared to the group with forelimb lame horses with a head and wither asymmetry pattern indicating the same forelimb (see Table 10). The biggest difference was found among the horses with a diagonal pelvic asymmetry to the lame forelimb. There was a higher occurrence of horses with a diagonal pelvic asymmetry among the horses with a forelimb lameness and a head and wither asymmetry pattern indicating the same forelimb.

Among the horses with an ipsilateral hindlimb asymmetry to the primary lame forelimb and a head and wither asymmetry pattern indicating different forelimbs, the hindlimb asymmetry was reduced after diagnostic analgesia of the primary forelimb lameness for two horses, while for one horse the hindlimb lameness increased and for one horse it was not changed afterward. For one of the two horses with both ipsilateral and diagonal hindlimb lameness, the hindlimb asymmetry was not changed after diagnostic analgesia.

Table 10. The frequency of pelvic asymmetry above threshold for at least one parameter (PDmax/PDmin) in the group of horses with forelimb lameness and a head and wither asymmetry pattern indicating different forelimbs (group 3a) and the group of horses with unilaterally forelimb lameness and a head and wither asymmetry pattern indicating the same forelimb (group 4c). The table show the frequency before and after diagnostic analgesia.

	Before		After:	
Group	3a	4c	3a	4c
Hindlimb	% (#)	% (#)	% (#)	% (#)
Ipsilateral	15.0 (3)	11.4 (8)	25.0 (5)	18.6 (13)
Diagonal	0.0 (0)	45.7 (32)	5.0 (1)	40.0 (28)
Both	10.0 (2)	4.3 (3)	5.0 (1)	2.8 (2)
None	75.0 (15)	38.6 (27)	65.0 (13)	38.6 (27)

#### 5. Discussions

#### 5.1 Study design

#### 5.1.1 Inclusion criteria

One inclusion criterion was that the horses should have an initial head or pelvic asymmetry above thresholds which was chosen to be >|15|mm for the head parameters (HDmax/HDmin) and >|7|mm for the pelvic parameters (PDmax/PDmin). The threshold was chosen to include asymmetries of a greater amplitude, in the hope of increasing the chance of finding a specific movement pattern for multiple limb lameness and decreasing the risk that the compensatory mechanisms would become too subtle. The threshold was nevertheless chosen to be low enough to represent the severity of lameness in horses found in the clinic and that is relevant for the clinicians.

Furthermore, the horses were selected if the initial lameness was reduced by at least 70% after diagnostic analgesia. This threshold was chosen to increase the likelihood of selecting horses with a lameness that was reduced from the diagnostic analgesia (to correctly locate and classify the lameness for the study) and not by other factors. If the threshold is too low the risk increases that horses with a lameness reduced by other factors are being included, such as the lameness being reduced after trotting or due to natural fluctuations of the intensity of the lameness. This would increase the risk that the lameness is interpreted wrong (for example located to the wrong limb), which could result in a wrong classification and analysis of the movement pattern of the horse.

#### 5.1.2 Group division

#### Group 1 – bilateral lameness

The horses in this group are considered bilaterally lame in the forelimbs or in the hindlimbs due to a reduction of 150% for one parameter (HDmax, HDmin, PDmax or PDmin) after diagnostic analgesia, which means that the value for the parameter changes from positive (right side) to negative (left side) or vice versa. The inclusion

criteria are based only on the change of either the head or pelvic asymmetry. This makes it possible that the horses also have a lameness in the other pair of limbs that could affect the movement pattern. The threshold of 150% reduction of the primary lameness after diagnostic analgesia was chosen to have a notable change in the lame side and a relatively great lameness on the other limb to reduce the risk of having too subtle changes in the movement pattern.

# Group 2 – Hindlimb lameness with a head and wither asymmetry pattern indicating the same forelimb.

This group includes horses with a hindlimb lameness as well as a head and wither asymmetry pattern indicating lameness in the same forelimb. These horses are suspected to have a multiple limb lameness since their compensatory head and wither pattern does not follow the most common one to a primary hindlimb lameness according to the study of Persson-Sjodin (2023).

# *Group 3* – *Forelimb lameness with a head and wither asymmetry pattern indicating different forelimbs.*

This group includes horses with forelimb lameness and a head and wither asymmetry pattern indicating lameness in the opposite forelimbs. These horses are suspected to have multiple limb lameness since their head pattern does not follow the most common one according to a primary forelimb lameness according to the study of Persson-Sjodin (2023).

#### Group 4 – Unilateral lameness

This group includes horses with a lameness which is reduced with 70-100% after diagnostic analgesia. These horses are considered unilaterally lame since the asymmetry decreased after diagnostic analgesia and became close to symmetric; 100% was chosen to not include horses where the asymmetry changes to the contralateral limb (e.g., bilaterally lame horses) and 70% was chosen with the same reasons as mentioned above. Nonetheless, it is possible that these horses may have a bilateral lameness anyway. There is a possibility that the diagnostic analgesia has not fully anesthetized the structures causing the pain due to time or because the pain emerges from several sites that are not all included in the anesthetized area. This can lead to the remaining pain in the primarily lame limb being as great as the pain in the contralateral limb making the horse's movement look symmetrical due to a bilateral lameness, could result in the pain still affecting the movement of other limbs through generating adaptive mechanisms to decrease the loading of the painful limbs.

# 5.2 The movement pattern of multiple limb lame horses

#### 5.2.1 Group 1 - Bilateral lameness

#### Bilateral forelimb lameness (Group 1a)

The head and wither asymmetry pattern in this group was slightly different in percentage compared to the group with unilaterally lame horses and compared to the result in the study by Persson-Sjodin et al. (2023). The result of this thesis shows a higher percentage of horses with a head and wither asymmetry indicating different forelimbs among the horses with bilateral forelimb lameness compared to unilaterally forelimb lame horses. This might be due to that some of the bilaterally lame horses are trying to compensate for the lameness by moving the head and withers in different directions, but one also needs to consider if there could be a compensatory cranial movement to a real hindlimb lameness.

Initially the horses with a bilateral forelimb lameness with a head and wither movement pattern indicating asymmetry in different forelimbs have a higher frequency of ipsilateral hindlimb asymmetry to the lame forelimb and none of the horses show only a diagonal hindlimb asymmetry to the lame forelimb. This is the opposite pattern of hindlimb asymmetry that was found among the horses with a bilateral forelimb lameness with the head and wither pattern indicating asymmetry in the same forelimb. The compensatory movement pattern of forelimb lameness is found to mostly mimic a diagonal hindlimb lameness (Uhlir et al. 1997; Maliye et al. 2015) hence one can question why none of the horses with bilateral forelimb lameness and a head and wither asymmetry indicating different forelimbs have a diagonal hindlimb asymmetry. It could be due to that these horses have a true ipsilateral hindlimb lameness.

A hindlimb lameness can give a compensatory head asymmetry indicating the ipsilateral forelimb and a compensatory wither asymmetry indicating the diagonal forelimb (Persson-Sjodin et al. 2023). Therefore, it is a possibility that the head and wither asymmetry pattern is not indicative of bilateral forelimb lameness, but that it is due to an ipsilateral hindlimb lameness. This argument is supported by the fact that all the bilaterally forelimb had a hindlimb asymmetry ipsilateral to the primary lameness after diagnostic analgesia. If it had been a compensatory lameness, the hindlimb lameness would have changed when the forelimb lameness changed after diagnostic analgesia. Instead, more horses showed an ipsilateral pelvic asymmetry to the primary lame forelimb afterward. The increased number of horses could be due to that some horses have a right-sided compensatory pelvic asymmetry after diagnostic analgesia due to the secondary lameness in the contralateral forelimb in a combination with that some horses have a true right-sided hindlimb lameness.

The argument that the horses with a head and wither asymmetry pattern indicating different forelimbs have a hindlimb lameness is also supported by the fact that all bilaterally forelimb lame horses and an ipsilateral lameness with a pelvic asymmetry above thresholds have a head and wither asymmetry indicating different forelimbs. To fully understand which secondary lameness was causing the hindlimb asymmetry, one would have needed to continue the examination with diagnostic analgesia. The phenomena could also be described by natural variation in the population and that it occasionally gives the illusion of a difference pattern due to a small study population.

For the pelvic movement among the horses with a bilateral forelimb lameness there was no overall pattern indicating a lower frequency of compensatory hindlimb asymmetry from an initial perspective (e.g., the pattern of pelvic asymmetry above thresholds before diagnostic analgesia) compared to the unilaterally forelimb lame horses. Neither was any initial indications found of an alternative compensatory pelvic asymmetry pattern compared to the group with unilaterally lame horses (e.g., no difference between the occurrence of a PDmax and PDmin asymmetry indicating the same or different hindlimbs).

#### Bilateral hindlimb lameness (Group 1b)

The group of horses with bilateral hindlimb lameness had a higher frequency of horses with a head and wither asymmetry pattern indicating different forelimbs, which is the compensatory movement pattern one can expect for a primary hindlimb lameness (Persson-Sjodin et al. 2023). The reason why the frequency seems to be higher in the group with bilateral hindlimb lameness could be due to a small study population. This argument is also supported by the fact that there is a higher occurrence of horses with bilateral hindlimb lameness with an ipsilateral forelimb lameness with a head asymmetry above the thresholds and a head and wither asymmetry pattern indicating different forelimbs, which also is the expected compensatory pattern for a hindlimb lameness (Persson-Sjodin et al. 2023). That, in short, means that no indicators were found for a bilateral hindlimb lameness since all the identified initially movement patterns are shown as a primary hindlimb lameness with its expected compensatory movement patterns.

# 5.2.2 Group 2 – Hindlimb lame horses with a head and wither asymmetry pattern indicating the same forelimb.

# Hindlimb lameness for PDmax, with a head and wither asymmetry pattern indicating the same forelimb (Group 2a)

Some of the horses with a hindlimb lameness and a head and wither asymmetry indicating the ipsilateral forelimb seem to be bilaterally lame in the hindlimbs since they had a change of lame side after diagnostic analgesia. This could explain the

deviating pattern from the majority of unilaterally hindlimb lame horses as seen in other studies (Persson-Sjodin et al. 2023). However, a head and wither movement pattern indicating the same forelimb were not found among the majority of the other bilaterally hindlimb lame horses in group 1b, which is noteworthy. This could indicate that it is not the bilateral hindlimb lameness affecting the head and wither pattern, but that there may be another co-existing factor to the bilateral lameness in these cases which affects the movement pattern of the head and withers.

A possible explanation for a hindlimb lameness with a head and wither asymmetry indicating the same forelimb could be that the horses are lame in both a forelimb and a hindlimb. The result shows a higher frequency of horses with an initially ipsilateral forelimb asymmetry above threshold among the horses with a hindlimb lameness and head and wither asymmetry indicating the same forelimb. The argument that it is a real ipsilateral forelimb lameness affecting the head and wither asymmetry pattern is supported by the fact that the ipsilateral forelimb lameness is just slightly reduced after diagnostic analgesia of the primarily lame hindlimb in some cases, when a compensatory lameness would be expected to disappear. This population of horses is though very small, which makes it harder to identify patterns.

## Hindlimb lameness for PDmin, with a head and wither asymmetry pattern indicating the same forelimb (Group 2b)

In the group of horses with hindlimb lameness for PDmin and a head and wither asymmetry pattern indicating the ipsilateral forelimb, just as in the group of hindlimb lame horses for PDmax, one horse seems to have a bilateral hindlimb lameness as well, since it had a change of the lame side after diagnostic analgesia. The vertical movement of the head and wither movement could be affected by the bilateral hindlimb lameness and therefore a head and wither asymmetry pattern indicating the ipsilateral forelimb could be an indicator in some cases. One can though question why the same result was not found among the bilateral hindlimb lame horses in group 1b. This could, as for group 2a, indicate that it is not the bilateral hindlimb lameness affecting the head and wither pattern, but that there may be another co-existing factor to the bilateral lameness in these cases which affects the movement pattern. The head and wither asymmetry pattern indicating the same forelimb among the horses with a hindlimb lameness could also be due to an ipsilateral forelimb lameness as mentioned above.

Among hindlimb lame horses for PDmax and a head and wither asymmetry indicating the same forelimb there is a higher frequency of horses with an ipsilateral forelimb lameness compared to the hindlimb lame horses for PDmax with a head and wither asymmetry indicating different forelimbs. This argument is supported by that the ipsilateral forelimb lameness seems to be a real lameness since it was hardly reduced after diagnostic analgesia of the primary lame hindlimb. Though to be able to ensure that the ipsilateral forelimb lameness is real or due to compensatory mechanisms, one would have needed to confirm it with diagnostic analgesia of the suspected lame limb.

# 5.2.3 Group 3 - Forelimb lame horses with a head and wither asymmetry pattern indicating different forelimbs.

# Forelimb lameness with a head and wither asymmetry pattern indicating different forelimbs (Group 3a)

The deviating head and wither asymmetry pattern among the horses with a forelimb lameness and a head and wither asymmetry pattern indicating different forelimbs could be due to a bilateral forelimb lameness since the results show that some of the horses have a bilateral forelimb lameness. The question is though why there was not a higher occurrence of horses with a head and wither asymmetry pattern indicating different forelimbs among the bilateral forelimb lameness in group 1a compared to the unilateral forelimb horses. It could indicate that the bilateral forelimb lameness is not affecting the head and wither asymmetry pattern, but instead that a co-existing factor to the bilateral lameness may occur in these cases.

The head and wither pattern could also be affected by a combination of forelimb and hindlimb lameness. The higher occurrence of ipsilateral hindlimb lameness among the forelimb horses with a head and asymmetry pattern indicating the different forelimbs compared to those with a head and wither pattern indicating the same forelimb supports the idea that an ipsilateral hindlimb lameness could affect the head and wither pattern among these horses. The argument that the horses have a real ipsilateral hindlimb lameness is also supported by the fact that none of the horses with forelimb lameness had a diagonal hindlimb lameness, which is the expected compensatory movement pattern which also was seen in a higher occurrence among the forelimb. To fully ensure if the head and wither pattern was affected by the bilateral forelimb lameness or a real ipsilateral lameness one would have needed to proceed with an examination with diagnostic analgesia of the suspected lame limbs.

#### 5.2.4 Conclusions

No clear indicators for multiple limb lameness in the initial phase of movement analysis during a lameness examination were found, but some cases give a suggestion of possible indicators. An ipsilateral forelimb and hindlimb lameness could in some cases appear as a forelimb lame horse with a head and wither pattern indicating different forelimbs and in some cases as a hindlimb lame horse with a head and wither pattern indicating the same forelimb. This means that if a forelimb or hindlimb lame horse with a deviating head and wither pattern is presented at the clinic, it is possible that the horse has a multiple limb lameness of the ipsilateral pair of limbs.

Whether a deviating head and wither asymmetry pattern is indicative for bilateral lameness is more unsure, since the tendencies are only shown in both the group of horses with a forelimb lameness and a head and wither asymmetry indicating different forelimbs and among the horses with a hindlimb lameness and a head and wither asymmetry indicating the same forelimbs (e.g., forelimb or hindlimb lame horses with a deviating head and wither asymmetry pattern), but not among the bilaterally forelimb or hindlimb lame horses. This might mean that a deviating head and wither asymmetry pattern could be due to a bilateral lameness, but it might also mean that in some cases it looks like a bilateral lameness due to a co-existing factor such as a third lameness.

#### 5.3 Limitations in the study

Some horses are expected to have a multiple limb lameness, but only the primary lameness was confirmed with diagnostic analgesia. To ensure if the suspected secondary lameness was a real lameness, one would have needed to confirm the lameness with diagnostic analgesia, which was not possible due to the retrospective design of the study.

The horses were also divided into groups depending on their primary lameness that was confirmed by diagnostic analgesia (unilateral or bilateral), but there is a possibility that the horses also had a lameness on other limbs than the primary lameness, which could affect the movement pattern and therefore result in a misinterpretation of their movement pattern.

The study is also limited by a small study population, which makes it more difficult to find patterns in the population and to use statistical methods.

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#### Popular science summary

Disorders from the locomotion apparatus is the most common disease category in horses and a common cause for euthanasia. A frequent symptom of these disorders is lameness in one or several limbs and to be able to treat such disorders the correct diagnosis needs to be made. However, it is challenging for veterinarians not only to treat the disorders, but also to detect and correctly locate the lameness. The detection of lameness presents its difficulties due to limitations in the human visual ability to perceive asymmetries, due to bias and the complexities of orthopaedic diseases and biomechanics.

A lameness is visible in the vertical movement of the body when the horse is trotting. As the horse trots, it will alternately land and push off from the diagonal pair of limbs. This will make the horse's body to move downwards before impact and during the first half of the stance of one diagonal pair and henceforth move upward in the second half of stance phase and after pushing off. The upward and downward movement will then happen again for the other pair of diagonal limbs. When a horse is sound, the upward and downward movement is symmetric between the two diagonal pair of limbs, which means that the horse reaches the same height in both the highest and lowest position above ground twice during a stride. When a horse is lame it will use adaptive mechanisms to decrease the loading in the painful limb as it moves, which results in asymmetries in the upward and downward movement between the two diagonal pair of limbs. When assessing lameness in a horse, such asymmetries are taken into examination; a forelimb lameness can be detected through asymmetries in the movement of the head while a hindlimb lameness can be detected through asymmetries in the movement of the pelvis.

The adaptive mechanisms to decrease the loading of the painful limb often result in an illusion of a lameness in another limb, so called compensatory lameness. It is most common with a compensatory same-sided forelimb lameness to a hindlimb lameness and a diagonal hindlimb lameness to a forelimb lameness. A compensatory lameness is not due to pain but only adaptive movements. These compensatory lameness's add complexity to the lameness detection, especially since the compensatory movements can appear as a more severe lameness than the real lameness and therefore disguise it. Studies have shown that veterinarians sometimes tend to mistake the compensatory lameness for the real lameness and therefore miss the actual lameness, especially when a horse has a hindlimb lameness. When a horse has a lameness in several limbs, the lameness detection gets even more complex, since one lameness often is evident in the initial assessment while the other lameness emerges first after diagnostic analgesia. Additionally, the compensatory movements from lameness in several limbs can make the horse look sound, which further complicate the assessment.

The movement pattern of horses with lameness on one limb is well described in studies as well as the compensatory mechanisms in horses with lameness on one limb, while the movement pattern of horses with lameness on several limbs is not. If one knew the initial factors in the horses' movements that indicates lameness in several limbs, it could then decrease the risk to miss a painful disorder as well as facilitate the examination for the veterinarian.

The aim of this study was to investigate horses considered or suspected to be lame on several limbs, to investigate whether specific movement patters can be inductors of multiple limb lameness. The study included 193 horses recorded with an objective gait analysis system called Qualisys, which is a camera-based system that records and analyses the movement of the horse. The horses were recorded during routine lameness examinations by veterinarians at four equine hospitals in Europe between October 2015 and November 2020. The horses included in the study showed a movement asymmetry above chosen thresholds for lameness and their lameness was reduced by at least 70% during diagnostic analgesia.

The horses in the study were furthermore divided into groups depending on whether they were considered lame in one or several limbs, or if they were suspected to be lame in several limbs because they had a deviating movement pattern of the head and wither from what is expected from a horse with lameness in one limb. Among the horses that were considered lame on several limbs the movement pattern was analysed with the assumption that they had a movement pattern that could be explained by the lameness on several limbs, while the horses suspected to be lame on several limbs were analysed with the assumption that they also had another lameness which could cause the deviating movement pattern.

The result of the thesis shows no clear indicators of lameness on several limbs but gives indications of that a horse with a forelimb and hindlimb lameness on the same side can either look as a forelimb lame horse with a head asymmetry indicating the lame forelimb and a wither asymmetry indicating the opposite forelimb, or as a hindlimb lame horse with a head and wither asymmetry indicating lameness in the same forelimb. The result also shows that a bilateral forelimb horse in some cases also have a head and wither asymmetry indicating different forelimbs as well as a hindlimb lame horse in some cases have a head and wither asymmetry indicating the same forelimbs. It is though uncertain whether it is the bilateral lameness causing the deviating head and wither asymmetry pattern or another (third) lameness. This thesis is limited by the difficulties to ensure that the horses only have the expected lameness. The possibility that the horses were lame in other limbs than the primary lameness was a cause to possible errors in the study, since it can result in a misinterpretation of the movement pattern and therefore erroneously comparing between the movement pattern of horses with lameness on one and several limbs.

#### Acknowledgements

First and foremost, I want to thank my supervisor Marie Rhodin for her guidance, support, and encouragement throughout this project. I would also like to extend my sincere thanks to Emma Persson Sjodin and Ylva Mellbin for their significant contributions to the data collection and data processing.

I also want to thank the veterinarians and veterinary assistants at the University Equine Clinic at the Swedish University of Agricultural Sciences in Uppsala for generously sharing their time and expertise, as well as making the days at the clinic an enjoyable time.

Finally, I want to thank my friends and family for listening patiently to my thoughts through excitement and frustration; and for always giving tons of encouragement. Especially I want to thank my dear classmate Ida Johansson, my partner André Sihm and dear friend Matteo Carone not only for proofreading but also for their interest to participate in discussions about the project, always willing to share their ideas and knowledge, not least about data processing and the usage of Excel.

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