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Kinematic discrimination of ataxia in horses is facilitated by blindfolding

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Abbreviations:

AIM	Automatic identification of markers
AUC	Area under the curve
ERH	Equine Referral Hospital
RVC	Royal Veterinary College
IMU	Inertial measurement unit
CV	Coefficient of Variation
s.d.	Standard Deviation
Se	Sensitivity
Sp	Specificity
ROC	Receiver Operator Characteristics

Summary

Background: Agreement amongst experienced clinicians is poor when assessing presence and severity of ataxia, especially when signs are mild. Consequently, objective gait measurements might be beneficial for assessment of horses with neurological diseases.

Objectives: To assess diagnostic criteria using motion capture to measure variability of spatial gait-characteristics and swing duration derived from ataxic and non-ataxic horses and to assess if variability increases with blindfolding.

Study design: Cross-sectional.

Methods: Twenty-one horses underwent measurements in a gait laboratory and live neurological grading by multiple raters. In the gait laboratory the horses were walked across a runway surrounded by a 12-camera motion capture system with a sample frequency of 240 Hz. They were walked normally and with a blindfold in at least 3 trials each. Displacements of reflective markers on head, fetlock, hoof, 4th lumbar vertebra, tuber coxae and sacrum derived from 3-4 consecutive strides were processed and descriptive statistics, receiver operator characteristics (ROC) to

determine the diagnostic sensitivity, specificity and area under the curve (AUC) and correlation between median ataxia grade and gait parameters were determined.

Results: For horses with a median ataxia grade ≥2, Coefficient of Variation for the location of maximum vertical displacement of pelvic and thoracic distal limbs generated good diagnostic yield. The hoofs of the thoracic limbs yielded an AUC of 0.81 with 64% sensitivity and 90% specificity. Blindfolding exacerbated the variation for ataxic horses compared to non-ataxic horses with the hoof marker having an AUC of 0.89 with 82% sensitivity and 90% specificity.

Main limitations: The low number of consecutive strides per horse obtained with motion capture could decrease diagnostic utility.

Conclusions: Motion capture can objectively aid the assessment of horses with ataxia. Furthermore blindfolding increases variation of distal pelvic limb kinematics making it a useful clinical tool.

Introduction

Ataxia is often recognised clinically as an irregularly irregular gait [1] and ataxia can also be defined as an interruption in the phase-dependent cyclical relationship between body segments in both spatial and temporal domains [2]. Ataxia can be the result of pathological disorders affecting the general proprioceptive system, cerebellum or vestibular system [3]. In the horse, the diagnostic workup is based on a thorough clinical and systematic neurological examination with neuroanatomical localisation [4]. This is followed, when appropriate, by laboratory testing of blood and cerebrospinal fluid [5], diagnostic imaging [6] and electrophysiologic examinations [7,8]. The ataxic horse remains a challenge, especially when the clinical signs are mild to moderate and even experienced clinicians disagree on the subjective assessment of gait and assignment of ataxia severity grades as well as whether the gait of a horse is normal or ataxic [9]. Development of objective criteria is therefore imperative to support the subjective assessment of gait as well as for detecting changes in gait over time in order to assess disease progression and response to

treatments. There is little research into use of gait laboratories for diagnostic purposes in ataxic horses. Cross-correlation of hoof motion pattern [10] and fuzzy clustering of motion capture signals [11] have been applied to ataxic horses walking and trotting on a treadmill. These techniques allow for discrimination between groups, but they have not been used diagnostically. Furthermore, horses often need several training sessions on the treadmill before the motion pattern is reproducible [12], treadmill exercise tends to stabilise gait and, given that [13], more subtle determinants of ataxia might be missed. Consequently, treadmill use in the assessment of ataxia is problematic: instead, over-ground gait analysis, might be preferable.

Various tests, such as walking a horse with a blindfold, are perceived to exacerbate clinical neurological signs and to help localise the anatomical location of the lesion [9,14]. Blindfolding is based on Romberg's test used in human medicine to assess the integration between vision, proprioception, vestibular and cerebellar systems [15]. In veterinary medicine, exacerbation of clinical signs after blindfolding is traditionally considered to be associated with vestibular ataxia [14,16]. Vision might have a feed forward effect on kinesthesia [17] and has recently been hypothesised to have a stabilising effect on postural control in horses [18] and spatial and temporal gait parameters in humans [19]. The influence of blindfolding on the gait of normal and ataxic horses has not previously been evaluated objectively.

We therefore aimed to assess the diagnostic utility of over-ground motion capture for differentiation between horses with and without ataxia. We hypothesised 1) that the stride-to-stride variation in motion pattern of limbs and trunk is greater in ataxic horses compared with non-ataxic horses (as measured by displacement of the fetlock and hoof, duration of swing phase, maximum vertical and latero-medial displacement of the head, lumbar region and tubera coxae) and 2) that this higher

variation can be used diagnostically and 3) that blindfolding increases variation of the motion cycle in ataxic horses compared to non-ataxic horses and 4) that objective determinants of gait as measured with motion capture correlate with the median neurological grade designated for a horse by up to 6 raters.

Materials and Methods

Rater agreement of clinical assessment of the horses in this study and results of post mortem examinations have recently been published [9]. Kinematics and inertial sensor data from 7 of the horses used in this paper have previously been published as part of validation studies [20,21].

Horses

Horses were recruited from three sources: Group 1 included research horses with no known history of gait abnormalities that were purchased for an unrelated study of recurrent laryngeal neuropathy; Group 2 comprised horses referred to the Royal Veterinary College's Equine Referral Hospital (ERH) for neurologic evaluation of gait deficits. Horses were recruited to Group 3 if a decision for euthanasia had been made in first opinion practice because of perceived moderate to severe ataxia. Horses were excluded from Group 3 if they were considered too ataxic to travel. None of the horses had signs or histories compatible with vestibular or cerebellar dysfunction. The horses were examined in order of presentation to the ERH and none of the horses showed signs consistent with cerebellar or vestibular ataxia. Four horses used in the rater agreement study [9] did not have gait laboratory data either due to concern over safety of the equipment or to practical and logistical constraints. Kinematic gait assessment was obtained within 24 hours of the live clinical assessment for all cases.

Subjective assessment of ataxia

The horses were assessed during a live neurological examination and graded for degree of ataxia using a modified Mayhew ataxia grading scale [9,22]. The assessment was performed simultaneously by at least 4 of the same 6 raters of whom two were internists (DipACVIM), two were surgeons (DipECVS or DipACVS) and two were residents (one medicine and one surgery). For details of the physical examination, results and grading scale see Olsen *et al.* [9]. The median of all raters' ataxia grades was used as the final ataxia grade assigned to each horse [9].

Data acquisition and processing

Hemispherical reflective markers with a diameter of 26 mm were placed on each horse at the poll, over the presumed centre of mass (CoM) [23] on both left and right side, left and right tuber coxae, left and right supraglenoidal tubercle, left and right thoracic and pelvic limbs over the latero-distal extremities of the metacarpal/tarsal II and IV just proximal to the metacarpo-/metatarso-phalangeal joint and hoof markers over the lateral and dorsal and distal hoof walls (marker placement is illustrated in Supplementary Item 1). Reflective markers with a diameter of 36 mm were placed on the skin over the dorsal spinous process of the withers at T13, over the 4th lumbar vertebra and one over the 1st dorsal spinous process of the sacrum. Horses were walked at their preferred speed by an experienced handler along a 20 m indoor runway 5 times for each condition. The conditions were 1) walking normally without manipulation (normal walk); 2) walking wearing a blindfold (blindfold), and 3) walking with the head elevated. The order of walking condition was randomised (using random.org). A 12-camera, optical, motion-capture system (Qualisys Oqus 300 and 500 series[a]) was calibrated to collect 3D kinematic data covering an area of 6 m (length) x 2 m (width) x 2 m (height). Data were recorded at 240 Hz utilising commercial software (Qualisys Track Manager, version 2.3[a]). Each trial was pre-processed with labels and automatic identification of markers (AIM) followed by

manual tracking and exported to tab-separated-values (tsv). The data was batch-processed using custom written MATLAB scripts (R2012a[b]) and segmented into strides based on displacement of the hoof relative to the centre of mass [24]. The parameters stride time, stance time and swing time were derived from the data stream [24]. To facilitate comparison between strides and horses, each stride was interpolated to 100 equidistant points [25]. The parameters calculated were maximal displacement, the displacement at 50% of swing phase and duration of swing phase. For each interpolated stride, parameters were calculated for each of the axes; X: cranio-caudal; Y: latero-medial and Z: vertical.

Data analysis

The displacement and stride parameters were summarised by mean, s.d. and Coefficient of Variation (CV = s.d./mean) for each horse across each of the 3 axes. Statistical analysis was performed using R [26] with the packages ggplot2 for graphical data exploration and figures and pROC for calculation of sensitivity (Se), specificity (Sp) and area under the curve (AUC). Based on the median ataxia grade, the horses were assigned to a group of being normal or abnormal. The data were split multiple times with abnormal being a) grade 1 or greater; b) grade 2 or greater and c) grade 3 or greater. The results were calculated for the conditions walking normally and walking with a blindfold. In pilot experiments we discovered that when the horses were walking with their head elevated they had large variations in velocity (CV >15%) within each trial and this condition was therefore discarded from the study. In addition we compared results for normal walk and blindfold for thoracic limbs alone, pelvic limbs alone and thoracic and pelvic limbs together. Correlation between the median ordinal ataxia grade and continuous stride parameters was done using a Cumulative Link Model in R with the package Ordinal with a significance level set at $p \leq 0.05$.

Results

A total of 21 horses with a median age of 6 years (range 3 to 16 years) had kinematic analysis and neurological examination. Seven horses were assigned a median ataxia grade of 0, 3 had a median grade of 1, 6 had a median grade of 2 and five with a median grade of 3. Post mortem examination was performed on 13 of the 21 horses, however this number was too low to get sufficient power using pathology as a grouping factor for kinematic parameters. Detailed signalment for the horses, neurolocalisation and histopathology can be found in Supplementary Item 2.

For normal walk, a head marker was added after collection of 3 horses so that 18 of 21 horses were wearing a head marker walking with a blindfold. The blindfold obstructed the head marker in the first 10 horses after which an additional head marker was added for the next 11 horses. The tuber coxae markers were less stable on the Automatic Identification of markers (AIM) model and there was no consistent trace of the RTC marker for 3 of 21 horses walking normally and with a blindfold.

The mean and standard deviation for stride, stance and swing duration for both thoracic and pelvic limbs are listed in Table 1. A total of 2096 steps were included across the 21 horses for both conditions. For each horse a median of 51 steps for both pelvic and thoracic limbs across trials and for walking with and without a blindfold were analysed. Descriptive statistics for all data streams included in the study can be found in Supplementary Item 3. There was no statistically significant difference in either mean or s.d. for stride, stance or swing duration when the horses walked with a blindfold compared to normal walk (Table 1). Results of the diagnostic ROC analysis is summarised in Table 2. Included in the table are sensitivity, specificity and cut-offs for kinematic traces with an AUC greater than 0.7 for any of the ataxia grade groups. Mean values were not good discriminators between ataxic horses and non-ataxic horses. The location for maximum displacement in the vertical (Z) direction for markers on fetlock and dorsal hoof wall had better diagnostic yield compared to head and trunk mounted markers as well as CV of duration of swing phase when ataxic horses are compared to non-ataxic horses. Only location of maximum vertical displacement of the hoof marker

for the thoracic and pelvic limbs has an AUC >0.7 across all groupings for horses walking normally. For horses walking with a blindfold the AUC is >0.8 for the location of maximum vertical displacement and maximum displacement at 50% of swing phase for the dorsal hoof marker on the pelvic limbs with Se of 82% and Sp of 90% for detecting horses with a median ataxia grade ≥2 which is higher than walking without a blindfold where the Se is 73% and Sp is 70%. In addition the marker on a pelvic limb metatarso-phalangeal joint (fetlock) had a greater AUC, Se and Sp than that on a thoracic limb, when walking with a blindfold. In general, both AUC, Se and Sp were greater for horses with an ataxia grade greater than or equal to 3 compared to those with a grade of 0, 1 and 2. There was a significant ($p \leq 0.05$) link between the ataxia grade and the CV and s.d. for all data features except for maximum vertical location during swing for the head-marker (Supplementary Item 4).

Discussion

Accurate assessment of equine neurological gait deficits is crucial for rider safety, for investigating effects of treatments such as surgery or physical therapy, for determining disease progression, making decisions for euthanasia and offering a prognosis. Here we investigate the potential for motion capture (kinematics) to differentiate between ataxic and non-ataxic horses. We used the median ataxia score for a group of raters as a reference standard to determine presence and severity of ataxia. We show that vertical displacement of the hoof and fetlock as well as swing duration have good diagnostic yield and that the pelvic limbs show more discriminatory capacity than the thoracic limbs. We also show that blindfolding increases the variation of vertical motion of

the distal limb for the ataxic horse compared to the non-ataxic horse; therefore our data support a stabilising effect of vision on posture and gait [18,19] in the so-called feed-forward hypothesis [27]. Our data also reveal that blindfolding exacerbates gait deficits associated with presumed general proprioceptive dysfunction.

Strobach *et al.* [10] evaluated 17 ataxic and 17 non-ataxic horses walking on a treadmill and found no significant differences in stride duration or stride length between the two groups but reported significantly lower duty factor and decreased maximum of the vertical flight arch for the ataxic horses. Their study also looked at auto-, and cross-correlation analysis of hoof marker signals and found a significant, but narrow difference between normally coordinated horses and ataxic horses. The authors evaluated mean and s.d. but not the diagnostic capability of cross-correlation. Auto- and cross-correlation functions are heavily affected by velocity and require many sequential strides in a steady state so analysis of 3 sequential strides in the measurement frame in the gait laboratory is insufficient. Based on our data, it appears that vertical displacement and swing duration are of greater diagnostic value when assessing horses over-ground. Keegan *et al.* [11] compared 12 ataxic and 12 normally coordinated horses walking on a treadmill and reported a correct classification for 100% of horses as ataxic or normal using fuzzy clustering (a form of cluster analysis) of the medio-lateral and dorso-ventral displacement of a lumbar marker combined with vertical displacement of a fetlock marker. Keegan *et al.* [11] did not report diagnostic utility beyond the fuzzy clustering; for comparison, we did not detect diagnostically discriminatory results when assessing lumbar displacement of markers, but did get good AUC, sensitivity and specificity for fetlock displacement in the vertical direction. Both previously reported studies [10,11] were conducted on a treadmill, which might have altered certain characteristics of gait, especially at the walk [28,29].

Medio-lateral (Y) excursion of the distal limb is often assessed clinically in the neurologic evaluation of gait, in particular, when evaluating possible ataxia. Indeed Ishihara *et al.* [30] found that ataxic horses had a significantly increased variation of the medio-lateral ground reaction force; in contrast, we did not find a significantly increased variation in the medio-lateral kinematic marker traces of the trunk during swing. We [21] and others [31] have previously shown a large inaccuracy when comparing medio-lateral displacement between inertial sensors and motion capture. The discrepancy is thought to be due to the low amplitude of the distal limb movement in the medio-lateral direction. We did not analyse the medio-lateral displacement of the distal limb in the current work due to the inherent technical challenge of quantifying and distinguishing the medio-lateral movement of the limb independently from any lateral drift in the horses' direction. The uncontrolled manifold hypothesis suggests that motor control stabilises the centre of mass though multi-joint synergies that limit variation of the displacement trajectory and the limbs not interfering with each other. This leads to complex feedback and feed-forward systems that stabilise the trajectory [32]. The medio-lateral excursion amplitude is low and the control mechanisms described above likely make the kinematic stride-to-stride variation during swing too large to be of diagnostic value. In smaller quadrupeds the neurologic examination includes hopping in the lateral direction which directly facilitates assessment of the proprioceptive pathways; this could be measured in future studies however it cannot be accomplished safely in the pelvic limbs of many horses. Future studies could assess the distal limb kinematics during pertubations such as medio-lateral manoeuvering including thoracic limb hopping and circling. Due to the challenges of standardising such measurements it is possible that clinical assessment might remain superior in assessing changes in medio-lateral motion, even though such assessment would remain subjective.

Foss *et al.* [33] analysed the gait of 10 clinically normal and 9 dogs with cervical spondylomyelopathy (CSM) and found a significant difference in the stride duration of the thoracic limbs but not the pelvic limbs. In contrast, we did not find a difference in absolute swing time between ataxic and non-ataxic horses; we did however find a diagnostically relevant increase in CV of swing time for horses

walking without a blindfold with an ataxia score greater than or equal to 2 compared to horses with an ataxia score of 0 and 1. This difference may be associated with the majority of CSM dogs having a C6-C7 lesion generating a two-engine gait [33] whereas this was not the case for the horses in this study.

Ishihara *et al*. [30] performed kinetic (force-plate) analysis of gait for 12 normal horses, 12 horses with lameness and 12 horses with spinal ataxia. Using the mean lateral force peak and coefficient of variation of the lateral force peak the study obtained an AUC of 0.94 for horses with ataxia versus clinically normal horses. A combination of force plate data and kinematic data might be advantageous to further improve the diagnostic yield of data obtained in a gait laboratory but force plates are not widely available outside research environments. In the present study we utilise the "reference standard" motion capture for objective gait analysis and quantification of displacement.

Inertial Measurement Units (IMUs) are small affordable and portable sensors that enable collection of longer stride series without the constraints of a treadmill or gait laboratory. IMUs are compatible with motion capture and can obtain accurate and precise vertical and cranio-caudal displacement of sensors on the head, trunk and fetlocks [21,34–36] as well as stride time characteristics [20,37]. Recent attempts towards the use of hoof-mounted IMUs revealed unacceptably large measurement errors for displacement [31]. All parameters measured in this study can be translated to a portable IMU-based system and several IMU-based systems are available for objective assessment of lameness in horses [38–43]. Three-dimensional accelerometers mounted on the sternum have been described to assess ataxia after sedation with alpha2 adrenergic agonists [44–46]. Sedation leads to a subjective perception of ataxia [47] although the movement pattern after sedation is markedly different from movement patterns in horses with spinal (general proprioceptive) ataxia [10].

Sedation-induced gait deficits resulted in more pronounced truncal sway, lower head carriage and tetraparesis compared to proprioceptive deficits where horses with low-grade ataxia and therefore the parameters developed by Lopez-Sanroman *et al*. [44,46] are, in our opinion, unlikely to be useful for the clinically ataxic horse.

Coordination involves complex interaction of proprioceptive and motor pathways, and their control at the levels of the brain and spinal cord. Gait incoordination results from dysfunction of these interactions between touch, proprioceptive feedback and their integration with feed-forward information derived from vision [48]. Indeed people without neurological disease had significantly increased CV stride time and stride length when walking with their eyes closed and the difference in CV between eyes open and eyes closed is greater at slower walking speeds [19]. Here we show that variation of location of the maximum vertical displacement of the distal limb during swing phase, in particular coffin and fetlock joint, is greater in ataxic horses and normal horses and the variation increases when the horses are blindfolded. Further it is an indication of the feed-forward effect of the eyes and the proprioception tested in Romberg's test in humans [49,50]. More research is needed into the effect of vision on the gait of quadrupeds to understand the role it plays in normal animals and in compensation for proprioceptive deficits.

In our study there was a large variation in agreement between raters assessing ataxia grades [9]. Given that there is no reference standard that can designate a horse as normal or abnormal nor currently any reliable severity scale, we used the median grade of all raters as an approximation of each horse's true ataxia grade. We considered whether histopathological assessment might be a better gold standard, but the classification of disease post mortem, is also subjective and likely misses functional deficits, or for practical reasons, might miss subtle or isolated lesions. Further, the

relationship between histopathological changes and ataxia severity has not been determined, but it likely depends on multiple variables, many of which could not be controlled in a study of clinical cases. A higher case number would enable assessment of combination of multiple criteria from gait analysis that might improve diagnostic accuracy for objective assessment of ataxia. Such data analysis requires a training data set and another data set for application. A wider variation of neuroanatomical localisation would facilitate more knowledge of pelvic limb abnormalities and their presence and severity in horses with C1-T2 myelopathies compared to T3-L3 myelopathies that might not have obvious or measurable changes of gait of the thoracic limbs.

Study limitations

Whilst the Se and Sp is excellent in this study population, it should be acknowledged that, as with any test, a low disease prevalence would affect the positive and negative predictive values of the diagnostic test: as such it should be applied to a larger population and tested as a screening tool compared to the neurologic examination, and preferably spinal cord histopathology.

We also recognise that use of our gait laboratory has an inherent disadvantage: although the walk-way in our facility is 20 m long, the motion capture measurement area only spans 6 m, which limits the number of consecutive strides per trial. The use of IMUs could enable study of many more strides per trial and thereby improve diagnostic utility, decrease the standard deviation and increase the power of similar or future studies. Further, trials likely were conducted at slightly different walking velocities because we allowed the horses to walk at their preferred speed as we felt this was most clinically-relevant; however, this factor might have influenced results. A future study examining influence of walking speed on selected gait variables in ataxic horses would be helpful.

In conclusion, we provide evidence for good diagnostic yield of using a gait laboratory in horses with neurological gait deficits, through analysis of fetlock or hoof displacement. We also show a significant link between the median ataxia grade and gait parameters. If implemented into current motion capture or inertial sensor systems for routine gait analysis outside the gait laboratory, this could have a significant impact on the objective assessment of ataxia in horses and knowledge of disease progression change over time and effects of treatment, as well as in the training of veterinary practitioners and students.

Authors' declaration of interests

T. Pfau owns a company (Equigait) with the commercial focus of IMU based systems for the use in lameness assessment in horses. This study was performed with custom algorithms independent from Equigait's software.

Ethical animal research

This manuscript has been approved by the Royal Veterinary College and assigned manuscript number CSS-01474. Owners gave informed consent for their horses' inclusion in the study.

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Authorship

E. Olsen was responsible for study design, study execution, data analysis and interpretation, and preparation of the manuscript. N. Fouche and H. Jordan were involved in study design, data analysis and interpretation, and preparation of the manuscript. T. Pfau and R. Piercy were involved in study design, and preparation of the manuscript. All authors gave their final approval of the manuscript.

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Table legends

Table 1: Descriptive data for stride time, stance and swing duration in ms.

Table 2: Receiver Operator Characteristics displayed as sensitivity, specificity, Area Under the Curve (AUC) and cut-offs for coefficient of variation (%) for gait parameters derived from ataxic and non-ataxic horses when walking with and without a blindfold. The analysis was performed for thoracic limbs (TL), pelvic limbs (PL) and thoracic limbs with pelvic limbs (TL&PL). The data were analysed 1) for horses with an ataxia grade greater than or equal to 1 (n = 14) compared to those with grade 0 (n = 7); 2) ataxia grade greater than or equal to two (n = 11) compared to those with grade 0 and 1 (n = 10) and finally 3) horses with ataxia grade greater than or equal to 3 (n = 5) compared to those with ataxia grades of 0,1 and 2 (n = 16).

Supplementary Information

Supplementary Item 1: Location of reflective markers.

Supplementary Item 2: Horse signalment, neurological examination findings and spinal cord histopathology.

Supplementary Item 3: Descriptive statistics.

Supplementary Item 4: Correlation statistics.

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Table 1: Descriptive data for stride time, stance and swing duration in ms

Leg	Condition	Stride duration Mean (s.d.)	Stance duration Mean (s.d)	Swing duration Mean (s.d.)
TL	Normal	1236 (105)	820 (83)	416 (30)
TL	Blindfold	1217 (145)	803 (117)	415 (59)
PL	Normal	1239 (105)	808 (82)	432 (49)
PL	Blindfold	1223 (140)	802 (111)	421 (67)
TL&PL	Normal	1238 (105)	814 (83)	424 (37)
TL&PL	Blindfold	1220 (143)	802 (114)	418 (63)

TL: Thoracic limb, PL: Pelvic limb, TL&PL: Thoracic and Pelvic limbs, s.d.: Standard Deviation

Table 2: Receiver Operator Characteristics displayed as sensitivity, specificity, Area Under the Curve (AUC) and cut-offs for the descriptive statistics for gait parameters derived from ataxic and non-ataxic horses when walking without and with a blindfold. The analysis was split into thoracic limbs (TL), pelvic limbs (PL) and thoracic limbs with pelvic limbs (TLPL). The data were analysed 1) for horses with an ataxia grade greater than or equal to 1 (n = 14) compared to those with grade 0 (n = 7); 2) ataxia grade greater than or equal to two (n = 11) compared to those with grade 0 and 1 (n = 10) and finally 3) horses with ataxia grade greater than or equal to 3 (n = 5) compared to those with ataxia grades of 0,1 and 2 (n = 16).

					Ataxia grade ≥ 1				Ataxia grade ≥ 2				Ataxia grade ≥ 3			
Walk	Limbs	Marker	Data feature	Direction +statistic	**AUC**	**Se**	**Sp**	**cut-off**	**AUC**	**Se**	**Sp**	**cut-off**	**AUC**	**Se**	**Sp**	**cut-off**
Normal Walk	TL	Hoof	Max Displacement	Z, SD	0.75	71.4	71.4	0.8	**0.82**	**63.6**	**90.0**	**0.9**	**0.86**	**80**	**93.8**	**1.2**
	TL	Fetlock	Max Displacement	Z, SD	**0.87**	**64.3**	**100**	**1.2**	0.71	63.6	80.0	1.2	**0.84**	**100**	**75.0**	**1.2**
	PL	Hoof	Duration of swing	X, CV	0.69	42.9	100	0.0	0.78	54.5	100	0.0	0.86	100	68.8	0.0
	PL	Hoof	Displacement at 50% of Swing	Z, SD	**0.76**	**71.4**	**71.4**	**0.7**	**0.73**	**72.7**	**70.0**	**0.8**	**0.94**	**80.0**	**100**	**1.1**
	PL	Fetlock	Max Displacement	Z, CV	0.61	64.3	71.4	0.1	0.69	72.7	70.0	0.1	**0.95**	**80.0**	**100**	**0.1**
	TLPL	Fetlock	Max Displacement	Z, SD	0.71	64.3	85.7	1.9	0.72	72.7	80.0	1.9	0.88	100	75.0	1.9
	TLPL	Head	Max Displacement	Z, CV	0.73	57.1	100	0.8	0.74	81.8	75.0	0.7	0.64	60.0	78.6	0.9
Walk with blindfold	TL	Hoof	Max Displacement	Z, SD	0.70	42.9	100	1.6	0.81	54.5	100	1.6	0.65	60.0	81.2	1.6
	TL	Fetlock	Max Displacement	Z, SD	0.84	78.6	85.7	1.5	0.79	63.6	90.0	1.7	0.56	60.0	75.0	1.8
	PL	Hoof	Duration of swing	X, CV	0.64	0.43	0.86	0.1	0.68	36.4	100	0.1	0.41	60.0	56.2	0.1
	PL	Hoof	Displacement at 50% of Swing	Z, SD	**0.83**	**85.7**	**71.4**	**1.0**	**0.89**	**81.8**	**90.0**	**1.2**	**0.89**	**100**	**75.0**	**1.2**

PL	Fetlock	Max Displacement	Z, CV	**0.80**	**78.6**	**85.7**	**0.1**	**0.76**	**81.8**	**80.0**	**0.1**	**0.74**	**100**	**62.5**	**0.1**
TLPL	Fetlock	Max Displacement	Z, SD	0.78	50.0	100	2.6	0.83	63.4	100	2.6	**0.94**	**100**	**87.5**	**2.6**
TLPL	Head	Max Displacement	Z, CV	0.78	66.7	100	6.4	0.79	100	50.0	8.0	0.68	75.0	71.4	0.9

AUC: Area Under the Curve, Se: Sensitivity, Sp: Specificity, cut-off: Cut-off value optimised to highest simultaneous sensitivity and specificity, CV: Coefficient of variation, proportion, SD: Standard deviation. TL: Thoracic limbs, PL: Pelvic limbs, TL&PL: Thoracic and Pelvic limbs. RTC: Right tuber coxae. Ataxia was assigned on a 0-4 scale as described in Olsen *et al* (2014). Max Location: The normalised time point where maximal displacement occurred during swing phase. *: Ordinal regression using cumulative link and the model ataxia grade ~ data feature. Bolded text: Diagnostically relevant and consistent across groupings. Z: Vertical, X: Cranio-caudal, Y: Latero-medial. Maximum displacement.