

Acoustic Myography: Its Assessment of Ground Reaction Forces as Measured through Forelimb and Hind Limb of the Horse during Mild Exercise

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Abstract

Background: Fractures in the limbs of racehorses are common, resulting among other factors, as the result of repeated ground reaction forces on bones and joints, leading to catastrophic failure. Aim: To quantify ground impact transmission through the limb bones of un-shod healthy horses using the non-invasive technique of acoustic myography (AMG). Methods: Four sites were selected for AMG measurements at the walk and trot, hoof wall (site 1) and sites 2 - 4, metacarpal 3, carpals and the radius of the left forelimb of two healthy horses. Measurements were on both rubber and concrete. AMG of the equine hind limb suspensory system was made and analyzed (amplitude and timing) for the proximal suspensory ligament (PSL) and the SDFT/DDFT. **Results:** AMG signal amplitude at site 1 (1.5 \pm 0.2 *versus* 1.1 \pm 1.5) was not found to be significantly different at the trot compared to the walk; however, sites 2, 3 and 4 were all significantly different when compared between the two gaits; site 2 P = 0.008; site 3 P = 0.006; site 4 P = 0.005. AMG signals recorded on the rubber surface had smaller amplitude than the equivalent signal and site on the concrete surface. Ground Reaction Force (GRF) transmission in the equine forelimb was 22 m/sec, whilst that of the hind limb suspensory system was 25 m/sec. Conclusion: Findings indicate that GRFs are transmitted proximally along the limb at considerable speeds, that they are dampened by tissues and structures in the limb, and that the GRFs are

present and detectable proximal to the fetlock joint.

Keywords

Horse, Foot, Limb, Acoustic Myography, Suspensory System

1. Introduction

It is a matter of physics (Newton's third law) that when a body stands motionless on a given surface, it exerts a contact force equal to the mass of that body and that an equal force in the opposite direction is exerted by the given surface on the motionless body, a so-called ground reaction force (GRF). Equally, in terms of biomechanics, when a body is in motion, the GRF increases due to acceleration forces, such that GRFs of up to three or more times body weight may occur [1]. Therefore, it is important that when in motion, a body is capable of absorbing incurred GRFs so as to prevent long-term injury to biological structures.

Bone fractures in racehorses have been reported as being the most common problem faced by the racing industry at the moment [2]. Musculoskeletal injuries in horses have been reported to be multifactorial, comprising not only a number of risk factors of biological nature (horse-related) but also of a non-biological nature (training program, shoe type, track surface, racing speed/time, etc.) [2]. In terms of incidence, a study spanning a period of 14 years and involving some 556,705 race entries revealed a risk of approx. 1.8% (10,000 entries) with most fractures occurring within long bones and joints of predominantly the forelimbs [3]. Away from the racecourse, such injuries are generally sustained in the field as a result of horse-to-horse kicking (43% of cases) either in terms of defense or aggression [4] [5], the majority of injuries being to the 2nd and 4th metatarsal/metacarpal bones (15.0% & 8.4%), radius (13.8%) and tibia (12.2%) [5]. Another study reported fractures due to kicking to be as high as 58% of all cases, arising from another horse, self-inflicted fractures, or box related injuries [6].

Indeed, the cyclical fatigue damage of horse limbs has been reported after as little as 10,000 strides of gallop [7], and there exists a correlation between high frequency components of GRF's and the development of tendonitis in racehorses [8].

Of course, it is worth noting that whilst catastrophic failure of limb bones can and does occur [9], they are most likely closely correlated with excessive GRFs on certain track surfaces. Although much of the GRF is in fact absorbed by biological tissues within the horse's limbs [10]. Indeed, Wilson and colleagues present calculations for the ability of an accessory ligament in series with muscle to usefully damp limb vibrations in horses, discovering that muscles absorb vibrational energy in the range 30 ± 100 Hz [10]. It has also been shown that equine accessory ligaments when healthy, absorb GRFs in a most efficient manner, but when injured and swollen lose this capability [11].

In light of this, this study has assessed foot-ground interactions in the equine forelimb using the technique of acoustic myography (AMG) [12]. As a technique, AMG enables a detailed and accurate measurement of active tissues, particularly those involved in movement, is independent of electrical signals measuring pressure waves solely, is portable, easy to use, inexpensive, re-useable and non-invasive [12]. Furthermore, it should be mentioned that this technique was used recently to measure the suitability of acoustic myography as a technique for assessing how ground reaction forces are dealt with in key sites up the human lower limb [13]. The main aim of this study was to measure the dampening of GRF in the forelimb bones of the horse with the hypothesis being that a measured GRF signal dissipates rapidly as it travels up a healthy equine limb.

2. Materials & Methods

2.1. Ethics

The study was carried out in accordance with the Helsinki Declaration. The owners of the horses used, gave their informed consent prior to the start of this study. This study was entirely non-invasive in its nature.

2.2. Subjects

Since this study was not only aimed at, but was also designed to assess the technical capabilities of AMG equipment in terms of its ability to detect ground reaction forces within the limbs of horses, the focus has been on the recordings themselves rather than a large number of subjects. Two subjects were included in the present study for the AMG and bone measurements (Horse 1 and Horse 2). The number of horses used in this study reflects the limited number of AMG units available in combination with the time available at Equitopia CA and the research time allotted to DI and SL. In addition to the AMG and bone measurements, four horses were recruited for equine hind limb suspensory system recordings (Horses A, B, C & D), again with a focus on the technique and its potential rather than a large cohort trial. The inclusion criteria were that the horses were sound and unshod with no history of lameness, they had an intact hoof wall and a normal appearing frog and heel bulb (not atrophied), they were also in regular work. Horses were excluded if they had been lame for any reason within the previous 12 months. The horses were on average 11.7 years old (range 9 - 16 years) with a mean weight of 550 Kg.

2.3. Experimental Protocol

Both AMG bone measurement subjects were walked and trotted on a hard concrete walkway, as well as on a rubber surface for a period of approximately 5 mins. A repeat recording was made for Horse 1 exactly 10 days after the first recording, with the sole intention of assessing repeatability.

2.4. Acoustic Myography

Acoustic myography (AMG) is a biomechanical method capable of recording energy waves generated in the equine limb as the result of foot impact [11] [12] [14]. AMG recordings were carried out with a CURO unit and CURO sensors (CURO-Diagnostics ApS, Denmark; formerly MyoDynamik ApS) and followed in real time on an iPad Air (Apple Inc, Cupertino, CA, USA) via the App "CURO Equine" and a dedicated data recording system that is freely available (<u>https://www.curo-diagnostics.com/</u>) and the details of which have been published [11] [15] [16]. Using this setup, it was possible for us to see the actual recordings while the horses were physically active. We used 20 mm sensors with a frequency recording range of $0.5 - 20 \pm 0.5$ kHz, and the sampling rate was 2 kHz. Recorded data was stored in the CURO unit and after completion of measurements transferred to the CURO software (<u>https://app.myodynamik.com</u>) for analysis.

Ultrasound-gel coated sensors were placed centrally on the dorsal hoof wall and laterally on the 3rd metacarpal bone (SITE 2), the carpus (SITE 3) and the radius (SITE 4) of the equine left fore-limb. With regard to measurements of the equine hindlimb suspensory system, sensors were placed on the PSL as previously described [11] and on the SDFT/DDFT approximately 3 cm and 9 cm distal to the PSL sensor, and horses were walked for approximately 5 minutes on a hard surface only. Connecting cables from the sensors were connected to the CURO unit which was placed in a pouch mounted on a surcingle fastened to the horse. The sensors and the wires from the sensors were secured with a flexible adhesive bandage (Snøgg; Norway) (see Figure 1).



Figure 1. The Acoustic Myography sensor setup, with sensor and cable attachment using flexible bandage (Snøgg). A 20 mm sensor was placed centrally on the hoof wall and on the 3^{rd} metacarpal, carpals and radius of the left fore-limb. Note that this horse was unshod as mentioned in the methods.

2.5. Exercise

The exercise session included walking and trotting on a hard concrete surface, as well as a more shock absorbent rubber surface. The walk and trot lasted for approx. 5 mins and measurements were made after a 10 min period of warm-up that comprised walking on a lunge. With regards to the equine hind limb suspensory system, measurements were made whilst the horse walked on a hard surface only and measurements were made over a 5-minute period of such exercise. These measurements were taken whilst the horse was led by a handler.

2.6. Ultrasound

The selected bone sites were scanned using ultrasound (Sonosite EDGE II; FUJIFILM Sonosite, Inc. Bothell, Washington USA) so as to ensure that the recorded signals were not due to underlying tissue movement, and equally to confirm a good placement close to the intended structures. The ultrasound unit was equipped with a high frequency linear probe (50 mm long; HFL50) and set to the frequency range of 6 - 15 MHz. In brief, the hair at the level adjacent to the sensor was marked, 70% isopropyl alcohol was applied to the site of interest to act as a contact medium, the probe was then placed on the sensor site in a transverse position and the depth setting was set to 1.5 cm. Immediately after each measurement the recorded image was captured and saved for future analysis. Images were analysed using the Sonosite EDGE II software in terms of skin-to-bone depth. For details of the ultrasound images see **Figure 2**. The 20 mm sensors and cables were securely attached using a flexible bandage (Snøgg) to minimize any unintentional movement during exercise and recording.

It should be noted that the equine hind limb suspensory measurement sites were based on previously identified and tested locations [11].



Figure 2. Ultrasound images for locations 2, 3 and 4 in **Figure 1**. Sensor 2: Left front medial proximal MC3; Sensor 3: Left medial C3; Sensor 4: Left medial radius at proximal physis, with skin to bone depths (A-A) of 0.2 cm, 0.5 cm and 0.2 cm, respectively.

2.7. Statistics

All statistics were performed using GraphPad InStat 3 for Mac (Version 3.0b, 2003; Graph-Pad Inc., La Jolla, CA). Data were initially tested for normal distribution and equal variance, and then subsequently analysed using a *t*-test. Differences between means with a *P* value > 0.05 were considered non-significant. Values are presented as the mean \pm the standard error of the mean.

3. Results

3.1. Recording Sites

All The selected sites were found to be free of extraneous tissue as can be seen from the ultrasound images for each of the 3 bone sites (see **Figure 2**); and sites had a skin-to-bone depth of 0.2 cm for site 2 medial metacarpal 3, 0.5 cm for site 3 carpal 3 and 0.2 cm for site 4 radial physis.

3.2. AMG

All AMG signals proved to be very repeatable, so the three recordings for Horse 1 from the first day of measurement and the two signals from the second day were pooled to provide a mean of 5 walk and trot sequences all made on the concrete surface (see Table 1(a)).

AMG signals generated on both the hard rubber and concrete surfaces at the walk illustrate how the large amplitude signal initially recorded in the hoof, was quickly dampened and dissipated by the time it reached the 3rd metacarpal bone (site 2) and was essentially undetectable in the 3^{rd} and 4^{th} sites of the carpal and radius bones, respectively. The signals recorded for the concrete surface had initially greater amplitudes for the first 2 sites than those signals recorded on the rubber surface but were quickly dampened to a minimal level proximally on the limb (see Figure 3 & Figure 4). Thus, on a rubber surface small shock waves were recorded in the 3rd metacarpal (site 2), as the initial signal at site 1 (0.55 V) was reduced by approx. 40% at site 2 (0.34 V) compared to the original signal, during both the walk and trot. The signals were then further dampened by 0.05 V (approx. 10% of initial signal) and by 0.23 V (approx. 42% of initial signal) at sites 3 & 4, respectively, for the walk and trot. In this way, the initial signal was reduced by some 90% from its initial amplitude by the time it reached site 4. On the concrete surface, while an appreciably greater shock wave was generated in the limb with some dampening in the 3rd metacarpal, the signal (0.84 V) generated at site 1 was reduced by 60% at site 2 (0.35 V) for the walk and by 25% (0.64 V) for the trot. The signals were then further dampened at sites 3 & 4 by 0.15 V during the walk and by 0.30 V during the trot.

The recordings taken for both the hard rubber and the concrete surfaces revealed that a detectable signal persisted at the trot at all 4 recording sites measured in contrast to those taken at the walk. It is important to mention that the signals recorded for the concrete surface were initially greater in amplitude for the first 2 sites than those for the rubber surface. **Table 1.** (a) Hard surface (concrete) S-scores for the AMG measurements made over 2 days for Horse 1. Repeats (Rep 1-3) were made on day 1 and repeats (Rep 4-5) were taken 10 days later. Values for the mean of the 5 repeats are \pm SD, and the final column presents the S-score mean values as amplitude in volts (Amp (V)). NB: the value of <0.01 for the trot at site 1 (Rep 1-3) indicates that the signal was slightly in excess of the 6 dB scale (1 V), whilst those for the same site (Rep 4-5) were just below the cut-off of 6 dB. (b) Mean values for S-scores for the AMG measurements for concrete *versus* rubber with statistical significance (*P*-values). NS = not significantly different.

(a)

	S-score	Rep 1	Rep 2	Rep 3	Rep 4	Rep5	Mean	SD	Amp (V)
WALK	Site 1	1.5	1.3	1.5	1.8	1.6	1.5	0.2	0.84
	Site 2	6.0	6.7	7.2	6.3	6.5	6.5	0.4	0.35
	Site 3	8.8	8.4	8.2	8.4	8.6	8.5	0.2	0.16
	Site 4	8.7	8.5	8.6	8.5	8.4	8.5	0.1	0.15
TROT	Site 1	2.7	2.7	0.01	0.01	0.01	1.1	1.0	0.88
	Site 2	4.4	3.7	3.0	4.1	4.2	3.9	0.5	0.64
	Site 3	6.3	6.2	6.0	7.2	7.1	6.5	0.5	0.35
	Site 4	7.6	7.7	7.4	7.1	6.8	7.3	0.4	0.27
-									

(b)

	S-score	Mean ± SD Concrete	Mean ± SD Rubber	Statistical Significance Concrete vs Rubber
	Site 1	1.5 ± 0.2	4.6 ± 0.0	<0.001
	Site 2	6.5 ± 0.4	6.8 ± 0.1	NS
WALK	Site 3	8.5 ± 0.2	9.5 ± 0.0	<0.001
	Site 4	8.5 ± 0.1	9.2 ± 0.1	<0.001
	Site 1	1.1 ± 1.0	4.4 ± 0.0	<0.05
TDOT	Site 2	3.9 ± 0.5	6.6 ± 0.1	<0.001
TROT	Site 3	6.5 ± 0.5	7.5 ± 0.2	< 0.05
	Site 4	7.3 ± 0.4	7.7 ± 0.1	NS

The results are reported as the S-score, which represented the inverse ratio of the amplitude of the signal obtained: a low S-score corresponded to a larger amplitude signal, while a high S-score corresponded to a smaller amplitude signal (see **Figure 5**).

3.3. Gait Effects

Site 1 values (1.5 \pm 0.2 *versus* 1.1 \pm 1.0) were not found to be significantly different at the walk compared to the trot; however, at sites 2, 3 and 4, S-scores were all significantly different when compared between these two gaits; site 2 *P* = 0.008; site 3 *P* = 0.006; site 4 *P* = 0.005, respectively.

On a more compliant rubber surface significant differences between values were noted compared with those obtained for concrete (see Table 1(b)).



Figure 3. AMG signals recorded at the walk for both a hard rubber and a concrete surface for each of the four placement sites illustrated in **Figure 1**. Note how the large amplitude signal initially recorded in the hoof is quickly dampened and dissipated by the time it reaches the 3^{rd} metacarpal and is essentially undetectable in the carpal and radius bones. The signals recorded for the hard concrete surface are of an initially greater amplitude for the first 2 sites *cf* those from the rubber surface, but also quickly damp to a minimal level. Note this horse was unshod as mentioned in the methods.



Figure 4. AMG signals recorded at the trot for both a hard rubber and a concrete surface for each of the four placement sites illustrated in **Figure 1**. Unlike the walk, trotting results in a persistent signal that is detectable throughout the 4 sites measured in this study. Note too how the signals recorded for the hard concrete surface are of an initially greater amplitude for the first 2 sites *cf* those from the rubber surface. The conduction velocity of the GRF from the hoof up through the sensor on MC3 (site 2) occurred at 24 m/sec, through to C3 (site 3) at 21 m/sec, and to the Radius (site 4) at 19 m/sec: a mean conduction velocity of 22 m/sec. Note this horse was unshod as mentioned in the methods.

3.4. Speed of Transmission

From the data presented in **Figure 4**, the speed of the generated shock wave was measured between the foot impact and the more proximal recording sites on the forelimb. The initial GRF reached the 3rd metacarpal by 13 msec after impact, whilst by 28 msec it was in the carpal joint and at 35 msec it was detectable in the radius. With the sensor separation distances from site 1 to site 4 being 31.7 cm, 42.5 cm and 65.8 cm, respectively, the conduction velocities at the three sites were calculated as being 24 m/sec, 21 m/sec and 19 m/sec, respectively, with mean conduction velocity value of 22 m/sec.



Figure 5. S-score AMG measurements. (a) a low amplitude AMG signal represents a high S-score, (b) a high amplitude AMG signal represents a low S-score, and (c) the S-score is calculated from the AMG signal amplitude using the equation shown, where 999 represents the full 6dB scale of 1 Volt. Note that the S-score is inverted, such that a HIGH score represents a signal with a low amplitude, and a LOW score represents a high amplitude signal, being calculated thus. A signal amplitude of 250 mV would generate an S-score of ((999-250)/999) \times 10 = 7.4.

3.5. Hind Limb Signals

With regard to the AMG signals made from the hind limb suspensory system, it was found that the SDFT/DDFT signals were clear and repeatable. Since the distance between sites 1 and 2 was known and time differences were measurable, the velocity with which this signal was transmitted to the SDFT/DDFT could be calculated (see Table 2).

It should be noted that a mean signal transmission velocity of 25 m/sec was arrived at for the four measurements made on the hind limb suspensory system in this study. For details of the sensor placement and the signal recordings for Horse A, see Figure 6.

4. Discussion

This study uses a novel technique to non-invasively assess the dampening of GRF in the forelimb of the horse as the generated shockwaves progressed from foot-ground impact to more proximal recording sites. This technique also highlights the importance of ground surface composition when horses move faster than at the walk.

Measurements of limb forces in horses, and the limb's ability to dampen GRF's illustrate the AMG's usefulness for examining equine locomotion to decipher many of the physiological events occurring simultaneously within different sites along the limbs, as well as for training individual animals and designing race courses and their compositional features. To date force plates have been used to assess GRF, but this requires clean impacts by the foot on the force plate at a set speed [17] [18]. While instrumented horseshoes avoid the possible variation and imprecision of limb contacts with the force plate during measurement, early examples were fragile and subject to damage [19]. More recent ones, though, risked the added weights of the shoes affecting the horse's normal gait [20]. To this end, the technique of AMG, as used in the present study, proved to be a quick and easy tool for collecting repeatable data with horses during locomotion. These findings are in agreement with those of human subjects [21]. Moreover, the AMG technique used in this study has a number of other benefits/advantages, being cheaper than competitor techniques such as force plates or force monitoring shoes, which furthermore cannot be used on different surfaces or are not sensitive enough for such studies.

Table 2. AMG signal recordings taken from the suspensory ligaments of the equine hindlimb, at the sites illustrated in **Figure 5**. The results of four horses (A, B, C, D) are presented, for which a delta signal for the recording made over the measured recording distance has been calculated to give a signal transmission velocity in the suspensory ligaments. The mean signal transmission velocity was found to be 25 m/sec. Horses were measured whilst walking on a hard rubber surface.

Horse	Recording site	Recording distance (cm)	Recorded signal delta (msec)	Signal transmission velocity (m/sec)
А	SDFT/DDFT	6	2.0	30
В	SDFT/DDFT	8	4.0	20
С	SDFT/DDFT	7	3.0	23
D	SDFT/DDFT	7	2.5	28



Figure 6. AMG signals recorded at the walk on a hard rubber surface for Horse A (see **Table 2**). The three recording sites are shown. Sites 1 and 2 were 6 cm apart and the signal delta was 2 msec. Note that in relation to the ground reaction force (GRF; signal = red line, direction of travel = green arrow) the order of recording in terms of the AMG signal was site 3, site 1 and site 2. PSL denotes signals recorded from the proximal suspensory ligament, SDFT/DDFT denotes signals recorded from the superficial and deep digital flexor tendons, respectively. At the bottom of the figure is a typical PSL recorded trace from a healthy equine limb; it is characterized by a clearly and narrowly defined signal with a small amplitude. In this case the recorded signal has a S-score of 8.3 which indicates a healthy PSL capable of adequately damping the GRF.

Studies of the transmission of GRFs through the distal forelimb of horses have reported a dampening effect of the signals within the foot and digit [22]. In support of these observations, the relative amplitude of the GRF is much attenuated at the metacarpus compared to the hoof wall [23] [24]. In fact, *in vitro* studies of the shock waves generated by hoof impact with a hard surface described the subsequent amplitude attenuation as being related to the digital bones and interphalangeal joints [23] [25]. Moreover, these authors went on to state that such GRF amplitudes were smaller above the proximal interphalangeal and metacarpophalangeal joints, both of which are distal to site 2 in the present study.

In an elegant study undertaken by Bobbert and colleagues (2007) [26], the calculated and measured forelimb average vertical reaction forces whilst walking were 6.9 ± 0.5 N/kg and 7.1 ± 0.3 N/kg, respectively, and whilst trotting these same values increased to 11.5 ± 0.9 N/kg and 11.7 ± 0.9 N/kg, respectively, all of which were higher than those obtained for the hind limbs. In terms of percentage change, the measured vertical reaction forces increased by 65% during trotting as compared to walking [26], and this percentage change was similar to that seen in the AMG signal measured at site 2 (the medial aspect of metacarpal 3). At this site, the AMG amplitude was 0.35 V whilst walking and 0.64 V whilst trotting on a hard surface representing an 82% increase.

The mean S-score increased from site 1 to sites 2, 3 and 4, both at the walk and trot. That is to say that the signal amplitude at each site decreases from approximately 0.8 V to 0.3 V or less, being the inverse of the S-score [11] [13]. It is likewise interesting to note that the S-score is higher for each site for both the walk and trot when measured on the softer rubber surface – that is to say, the signal amplitude is smaller than the equivalent signal and site for the hard concrete surface, indicating that the surface absorbs and dissipates a portion of the GRF conducted through the limb.

It is apparent from the present study that at the walk *cf* the trot, most of the GRF is absorbed and dampened within the foot and the 3rd metacarpal bone. Relatively little GRF can be detected at the walk in either the carpals or radius bones on either a rubber or a concrete surface. Thus, in this respect, the present findings not only support those of previous investigators [23] [25], but also extend them by taking measurements higher up the equine forelimb than the metacarpophalangeal joint.

Wilson and colleagues present calculations for the speed with which hoof impact vibrations are dampened on both a hard concrete and rubber matting surface, reporting that dampening occurs within 100 msecs of hoof impact [10]. Our own data (see **Figure 4**) are in full agreement with these calculations by Wilson and colleagues, showing that the GRF signal is not only greatly dampened by the time it reaches the carpals, but that this occurs within a time frame of approx. 20 msecs.

It has been postulated that the GRF is only experienced by tissues and struc-

tures below the fetlock [24], yet the present study has clearly shown that this is not true, particularly for certain gaits and ground surfaces. Whilst at the walk, it is true that very little GRF is detectable in the present study at sites 3 and 4, above the metacarpal site; the same cannot be said for either a rubber or concrete surface when horses trot. Here it is likewise interesting to note that on a rubber surface there is less GRF signal in the hoof and metacarpal site compared to that on a concrete surface, but whilst dampened, the signal remains equally detectable at sites 3 and 4 for both surfaces.

These findings reinforce the importance of considering ground surface material for horses working at high speeds, as well as those recovering from or rehabilitating injuries, especially those within the hoof capsule.

We have previously published results for the equine hind limb proximal suspensory ligament, in which AMG was used to detect and assess the functional health of the ligaments by means of their ability to act as damping harmonic oscillators [11]. The present study examined four horses to measure the speed of signal transmission as it travels up the suspensory system, doing so by measuring the signal at two locations on the same tissue (SDFT/DDFT). The speed of signal transmission is commensurate with that recorded in the skeletal tissues; 25 m/sec (suspensory system) cf22 m/sec (bones), which is a reassuring finding. However, despite this observation, it would seem that the PSL plays a unique role as part of the hindlimb suspensory system as **Figure 6** shows quite clearly that the damped GRF signal is first observed in the PSL before appearing in the SDFT/DDFT, despite the fact that the later sensors were lower down the leg than that on the PSL, and therefore much closer to the signal source. This phenomenon was noticed for all four of the horses in this study, which now warrants greater attention and research focus.

5. Conclusion

This study highlights the importance of a healthy foot and suspensory system in the horse since both structures combine to greatly reduce the GRF effect on the limb bones and joints. Moreover, in the light of; 1) reports that repetitive impulsive loading of the horse's foot is a source of stress to tissues in the limb that can over time result in bone damage and failure [27], and 2) the ease of measurement of GRF's in the equine limb with the AMG equipment, we propose further studies of the effects of gait, shoeing, training regimen and track surface.

Author Contributions

The project design was arrived at by DI, RB and AH based on preliminary trials by JCC, RB, DI, VSE and AH. Data collection was performed by DI and SL, with suspensory ligament measurements by JCC, VSE and AH. Analysis was performed blinded by AH. WA assisted with the AMG and suspensory measurements as well as the dissection of the limb shown in **Figure 6**. All authors contributed to the interpretation and writing of this manuscript.

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Author Information

AH is in the process of establishing a company to produce and market the Acoustic MyoGraphy equipment (CURO-Diagnostics ApS).

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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