The effect of acupuncture on objective and subjective gait parameters in horses

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Abstract:

Reasons for performing the study: Despite century long-standing application in human and veterinary medicine, it is still questioned whether acupuncture has any effect other than placebo.

Objectives: To investigate whether acupuncture can alter gait in horses as assessed by objective and subjective parameters.

Study design: Prospective, randomised, single-blinded, cross over study.

Animals: Eight adult horses.

Methods: Horses were randomly assigned to a treatment (three acupuncture treatments in 8 days) or control group. Subjective and objective gait analyses were performed before and after each treatment and at 1, 3 and 7 days after the last treatment (time-points 1–9, respectively). Horses were assessed at the trot in a straight line on a hard surface and on the lunge on the left and right reins on a soft surface (conditions 1–3, respectively). After 12 weeks, groups were reversed. Objective gait analysis was performed using inertial sensors and subjective analysis by two board- certified surgeons who reviewed video-recordings. Each limb was assessed for lameness before and after treatment. Lameness and global scores were assigned using 4-point scales. Assessors were blinded to treatment status. The effects of treatment (yes/no), time (1–9) and horse under conditions 1 –3 were compared using a linear mixed-effects model and a generalized estimating equation.

Results: Treatment decreased hip hike difference under all conditions [condition 1: control, 6.3 ± 6.4 mm versus treatment, –0.2 ± 6.4 mm (p = 0.007); condition 2: control, 9.7 ± 7.8 mm versus treatment, 2.8 ± 7.8 mm (p = 0.032); condition 3: control, 7.3 ± 6.3 mm versus treatment, –2.7 ± 6.4 mm (p = 0.003)]. Other parameters also improved significantly under conditions 1 and 3. Based on subjective gait analysis, treatment decreased lameness [odds ratio (OR) 0.51,

95% confidence interval (CI) 0.34–0.78; p = 0.002] but not global (OR 0.53, 95% CI 0.24–1.10; p = 0.12) scores.

Conclusions: Acupuncture can change horses' gaits to a degree appreciable by objective and subjective analyses.

Introduction

Acupuncture, which is part of traditional Chinese medicine, is increasingly incorporated into human and veterinary medicine. Traditional techniques have been adopted worldwide and numerous variations of acupuncture therapy have been developed. The mechanisms of action are still not fully understood. However, the use of modern investigation tools, including sophisticated imaging techniques, has suggested that acupuncture triggers a sequence of events involving the release of endogenous opioid- like substances, such as enkephalin, endorphin and endomorphin, which are measurable in plasma and cerebrospinal fluid, and which modulate pain signals (Wang et al., 2008 ; Kawakita and Okada, 2014). The limbic system has also been shown to play an important role in acupuncture-induced analgesia (Wang et al. 2008).

Although acupuncture is used in the treatment of a wide variety of diseases in people and veterinary species (Mittleman and Gaynor, 2000; Schofield, 2008; Yin and Chen, 2010; de Fourmestraux et al., 2014 ; Pfab et al., 2014), one of its most common applications is in the management of chronic pain, often arising from the musculoskeletal system. Large-scale clinical trials in people have demonstrated the efficacy of acupuncture in the treatment of lower back pain compared with standard care or patients not currently being treated (Brinkhaus et al., 2006 ; Haake et al., 2007). Over the last decades, acupuncture has been increasingly incorporated into equine medicine, but controlled clinical trials investigating its efficacy remain sparse. Anecdotally, acupuncture has been reported to be successful in alleviating a

variety of painful orthopaedic conditions, particularly back pain. However, the assessment of treatment failure or success is often subjective and influenced by the owner's and the attending veterinarian's expectations. The lack of controlled studies in which objective assessments are made has led to continued scepticism about whether acupuncture has any effects at all in horses (Ramey 2005). This pilot study tested the hypothesis that acupuncture treatment would result in alterations in gait in horses and that these would be appreciable by objective and subjective gait analyses.

Materials and methods

Authorization was granted by the Ethics and Welfare Committee of the Royal Veterinary College, London (URN approval no. 2013 1244). Eight systemically healthy horses, as determined by physical examination (performed by BD) that belonged to the Royal Veterinary College Equine Referral Hospital teaching and blood donor herd were used. The number of horses was based on those in similar studies that demonstrated treatment effects with four to eight animals per group (Xie et al., 2001; Xie et al., 2005 ; Angeli et al., 2005). Horses were unshod, not regularly exercised and lived out on pasture year round. Any horse that was sound or mildly lame [≤2/10 on the lameness scale devised by Wyn Jones (1988)] on baseline evaluation was included in the study, but obviously lame horses (lameness score of >2/10) were excluded.

Horses were randomly allocated to either the treatment or control group by drawing numbers. After a 12 week wash-out period, groups were reversed. Assessors involved in obtaining and analysing the data for objective and subjective gait analyses were blinded to the treatment status of the horse.

In practice, acupuncture treatment commonly consists of the palpatory determination of treatment points followed by needling of those points. This was mimicked in the study design. After a baseline gait analysis, horses in the treatment group were assessed independently by two veterinarians trained in acupuncture (KOV-N and BD). Transpositional horse acupuncture points were used as described (Fleming 2000). Painful or tense areas were identified by palpation and intended treatment points recorded by each assessor separately. Additionally, body regions perceived to be painful were noted [neck, shoulder, proximal thoracic and pelvic limbs (distal to the shoulder and hip joints and proximal to the carpus and tarsus, respectively), distal thoracic and pelvic limbs (distal to the carpus and tarsus, respectively), back and abdomen]. Perceptions of pain or tension were based on avoidance reactions elicited by palpation. Identified painful regions and chosen acupuncture points were then compared and the final treatment plan consisting of 11 acupuncture points was agreed upon by discussion. Eleven points were chosen because, anecdotally, some acupuncturists have advised against treatment of more points per session. Bai-hui, a frequently used point deemed to have benefits against a multitude of problems, located at the level of the lumbosacral space (Fleming 2001), was treated in every horse by prior agreement. The palpatory assessment was carried out in all horses immediately before the first treatment. Control horses were brought into a stable from pasture and rested in the stable during the initial acupuncture assessment and treatment time (approximately 45 minutes), and on later days while treatments (approximately 30 minutes) were carried out.

Acupuncture at the identified points was administered using commercially available sterile single- use acupuncture needles size 0.3 x 30 mm and 0.3 x 50 mm (Tai-Chi Power 100; Wellkang Ltd, UK). Needles were left in place for 20 minutes before removal. Mild to moderate stimulation generated by rotating the needle and gentle up-and-down movements was carried

out three or four times during the 20 minute session. Treatments were repeated twice over the course of 8 days (treatments on days 1, 4 and 8) and were performed by the same person. Subjective and objective gait analyses were carried out before and immediately after each treatment [before and after the first treatment (T1 and T2, respectively), before and after the second treatment (T3 and T4, respectively), before and after the third treatment (T5 and T6, respectively)] and at 1, 3 and 7 days after the last treatment (T7, T8 and T9, respectively). The treatment and assessment block spanned a period of 15 days.

Horses were assessed under three different conditions: at the trot in a straight line on a hard surface (condition 1), and at the trot in a circle on a soft surface on the lunge on the left rein (condition 2) and right rein (condition 3). Objective gait assessment was carried out using quantitative sensorbased gait analysis as previously described (Robartes et al. 2013). In brief, multiple 6 degrees of freedom inertial measurement units (IMUs) (MTx; Xsens Technologies BV, the Netherlands) and one combined IMU/global positioning system sensor (MTi-G; Xsens Technologies BV) were synchronized via a wireless transmitter unit (Xbus; Xsens Technologies BV). Data from each sensor were transmitted at 100 Hz per individual sensor channel to a laptop computer running commercial and custom-written analysis software (MATLAB Version 2012a; MathWorks Ltd, UK) (Pfau et al., 2005 ; Robartes et al., 2013). Sensors were attached to the horse over the poll to the halter with self-adhesive tape (Velcro; Velcro Ltd, UK), and along the midline to the withers, the mid-thoracic and lumbar region, the highest point of the pelvis (os sacrum) and just above the root of the tail and both tubera coxae with adhesive tape (Animal Polster; Snøgg AS, Norway). The areas of sensor attachment were clipped to minimize variations in sensor positioning between measurement days. Data were analysed from the sensor on the poll ('head nod') and the sensors over the sacrum and attached to the left and right tuber coxae ('pelvic hike'). The unit was fastened to the horse using a

surcingle. Sensor attachment was checked at regular intervals between exercises to ensure the sensor's firm attachment to the required anatomical landmark throughout the entire assessment (Robartes et al. 2013). Objective gait analysis referred to seven different variables which have been described in detail elsewhere (Starke et al. 2012). In brief, variables included a symmetry index for head and pelvic movement (H_SI and P_SI, respectively), a measure for thoracic (T_min) and pelvic (P_min) limb weight bearing, a measure for thoracic (T_max) and pelvic (P_max) limb push-off, and the hip hike difference (HHD).

For subjective gait analysis, video-recordings of horses trotting in a straight line were reviewed by two board-certified veterinary surgeons (DMB and AF-J). Only videos obtained before treatment (T1) and at 1 day (T7) and 7 days (T9) after the last treatment were reviewed. Each limb was assessed for lameness on a 4-point scale (0 = sound, 1 = minimally lame, 2 = mildly lame, 3 = moderately lame). The baseline video-recording (T1) was then compared with videos from T7 and T9 and a global score was assigned (1 = lameness better than baseline, 2 = lameness unchanged from baseline, 3 = lameness worse than baseline, 0 = cannot evaluate).

Data analyses:

Data were analysed using IBM SPSS for Windows Version 19.0 (IBM Corp., NY, USA). Normality of the data was assessed by the inspection of histograms. Objective gait assessments are presented as the mean ± standard error. Comparisons between subjective lameness scores are expressed as odds ratios (ORs) and 95% confidence intervals (CIs). Data for each of the three conditions were analysed separately. Baseline objective and subjective gait analyses data (T1) from the treatment and control groups were compared using a paired t test and McNemar chi-squared test, respectively, to rule out the existence of differences before treatment. A linear mixed- effects model was then used to investigate the effects of treatment, time (T2-T9) (fixed

factors) and horse (random factor) on outcome variables of objective gait analysis. Interactions between treatment and time were also evaluated. A generalized estimating equation (GEE) was used to investigate the effects of horse, treatment, time and observer on outcomes of subjective gait analysis (lameness score and global score). Condition was not taken into consideration in the subjective gait analyses as horses were evaluated only while trotting in a straight line. Inter-observer agreement between the subjective gait assessors in lameness evaluations and global scores was analysed using Cohen's kappa (κ-values of 0, 0.0–0.2, 0.21–0.4, 0.41–0.6, 0.61–0.8 and ≥0.81 were considered to indicate poor, slight, fair, moderate, substantial and excellent agreement, respectively) (Landis & Koch 1977). A p-value of ≤0.05 was considered to indicate statistical significance.

Results

Two Irish Sports horses, two ponies and four trotters (two geldings and six mares) with a mean ± standard deviation age of 14.8 ± 5.3 years were included in the study. Acupuncture treatment was well tolerated by all horses except that in one horse the insertion of the needle at one specific point resulted in violent shaking and foot stamping. This point was left untreated during all treatment sessions. One horse developed a foot abscess between T8 and T9 while serving as a control and was not submitted to the last assessment (T9). Treated acupuncture points included Bladder 23 in six horses, Large intestine 15 and Small intestine 9 in five horses each, Bladder 10, 13, 25 and 40 and Triple heater 16 in three horses each, Gallbladder 21 and Large intestine 11 in two horses each, and Large intestine 16, Triple heater 15 and Bladder 16, 20 and 22 in one horse each. The most commonly identified painful areas were the neck, shoulder, proximal thoracic limb and back.

No differences in variables of objective and subjective gait analyses were detected between groups at T1 (p-values 0.19–0.91 and p-values 0.24–0.47, respectively). No interactions between treatment and time were detected, and time had no effect in the mixed linear model (p-values 0.23–1.0). Treatment significantly affected some, but not all, parameters of head and thoracic limb movement (Fig. 1) and pelvic limb movement and HHD (Fig. 2) (Table 1). Overall, the movement of treated horses was more symmetrical than that of untreated horses.

Time did not have a significant effect on the subjective lameness analysis or global score, and no interactions between time and treatment were detected. The GEE indicated that treatment decreased the odds of subjective perceptions of lameness compared with equivalent perceptions in the control group (OR 0.51, 95% CI 0.34–0.78; p = 0.002), but did not have the same effect on the global score (OR 0.53, 95% CI 0.24–1.10; p = 0.12). Compared with observer 1, observer 2 assigned slightly higher scores for lameness (OR 1.9, 95% CI 1.1–3.2; p = 0.023), but did not do so for global scores (OR 1.7, 95%, CI 0.7–3.9; p = 0.22). Mean and median lameness grades and global scores before and after treatment are depicted in Table 2; Table 3. Means are displayed in addition to medians and ranges to illustrate numerical changes.
Agreement between acupuncturists in choosing the same points for treatment was 20% and 80% for identification of painful areas. Inter-observer agreement for subjective gait analysis was moderate (Kappa = 0.51; p<0.001).

Agreement between acupuncturists was 20% for the choice of points for treatment (excluding the Baihui point, on which agreement had been achieved prior to the start of the study) and 80% for the identification of painful areas. Inter-observer agreement for subjective gait analysis was moderate ($\kappa = 0.51$, $p < 0.001$).

Discussion

To the authors' knowledge, this is the first prospective, single-blinded, crossover study to evaluate the effects of acupuncture treatment on movement parameters in horses. The study was conducted as a pilot study to address the basic question of whether or not acupuncture treatment has any appreciable effect on gait in horses. Encouragingly, treatment effects were demonstrated for at least one objectively assessed pelvic limb parameter under all conditions and for all pelvic limb parameters under two conditions. However, several parameters showed no significant changes, which may indicate that acupuncture was not uniformly effective. Alternatively, the study may have been underpowered or its assessment methods not sensitive enough to detect more subtle differences. Objective gait analysis was chosen in the hope that this would detect changes in gait that might be difficult to appreciate by visual assessment. As with most methods, objective gait analysis is not perfect and parameters pertaining to the thoracic limbs may be particularly influenced by the horse's head movement in response to the environment.

Overall, treated horses moved in a more symmetrical manner, which suggests a lesser degree of discomfort. This was corroborated by the subjective assessment, which showed that lameness improved in the treatment group. The absence of significant changes in global scores may reflect the fact that assessors were unfamiliar with this form of assessment. When the study was planned, it was anticipated that treatment effects on horses' gaits would be difficult to appreciate by visual assessment and a global score was introduced to account for slight subjective improvements in movement. A possible explanation for the absence of differences is that the surgeons were more familiar with conventional grading of gait asymmetries and therefore more consistent in the assessments they made using a familiar method. Moreover, this scoring system has not been investigated previously and may not be the most suitable

method to detect gait changes. The fact that only a small number of horses were investigated may also have contributed.

Research into the use of acupuncture is hampered by many factors. The absence of standard treatment protocols has led to wide variations in opinions with regard to application, frequency and duration of treatments. The anatomical landmarks describing the locations of acupuncture points are sometimes vague and decisions on which and how many points to treat are usually based on the subjective assessment and personal beliefs of the acupuncturist. The lack of a good control procedure and difficulties in finding objective methods with which to evaluate the (often subtle) treatment effects further complicate the design of studies. The design of this study was chosen to resemble a treatment approach taken by many equine practitioners who use acupuncture as adjunctive therapy. This entails the careful palpation of all body parts and the application of pressure to acupuncture points to identify areas that might benefit from treatment (Still 2013). Chronic muscular pain in people is assessed in a similar fashion, whereby tender muscle groups are identified by careful palpation (Gerdle et al. 2014). This method is limited by the subjective nature of the assessment and the inability to objectively verify findings. As the verification of palpatory findings was not an objective of the present study, this was considered acceptable.

Choosing an adequate control procedure is challenging because acupuncture in areas not associated with established treatment points, as well as minimal [superficial needle insertion that does not elicit the so-called de-qi sensation assumed to be essential for effective acupuncture by some (Kawakita & Okada 2014)] or simulated (using a wooden toothpick to prick the skin) acupuncture techniques have been shown to be as effective, or only marginally less effective, as traditional treatment. Equally, palpation and the application of pressure to

acupuncture points represent a recognized treatment (acupressure) and these procedures can hence not be considered as physiologically inert placebo interventions (Kawakita & Okada 2014). For this reason, control groups in studies involving people often include a standard treatment protocol (conventional treatment without acupuncture) or no treatment, if this is ethically justifiable. The aim of this pilot study was to evaluate the effects of acupuncture treatment as it might be carried out by practising veterinarians and thus control horses were left undisturbed in a manner that mimics the no-treatment approach in people. The disadvantage of this design is that it was not possible to differentiate between the effects of palpation and those of needling. Given that horses were palpated only once, before the first treatment, and that gait changes persisted throughout the study period, it appears more likely that needling was responsible for the observed changes. However, future studies are required to confirm or refute this assumption.

The study design did not allow for the identification of the mechanism by which the treatment effect was achieved. Electroacupuncture increased cere- brospinal fluid opioid concentrations and decreased thoracolumbar pain scores in horses, and acupuncture and electroacupuncture provided cutaneous analgesia in an experimental setting (Skarda et al., 2002; Xie et al., 2005). It is therefore considered most likely that a generalized analgesic effect may have contributed to the findings reported here. However, it is also possible that pain associated with needle insertion was sufficient to change the gait in treated horses. If this were the case, one would expect the most profound differences to become apparent at the time-points immediately after treatment (T2, T4 and T6). The lack of an effect of time suggests that this was not the case. Alternatively, it is possible that repeated trotting, particularly on hard ground, worsened the gait of the control horses. Again, the absence of a significant time effect argues against this possibility and supports the theory of an analgesic effect. A number of statistical tests were

performed to increase the detection of differences despite the relatively small number of study subjects in this preliminary investigation, accepting a small risk that differences might be detected by chance. Given the consistency of the results, a random effect can probably be discounted.

The lack of a treatment effect in most objective measures of thoracic limb movement may derive from sensor positioning. As discussed, head movement is often more variable and is influenced by other external factors. Particularly in horses that are not schooled in lunging, more head movement might be expected in response to their environment. It is also possible that the study was underpowered to detect a difference or that treatment of the chosen points simply does not affect thoracic limb movement. A longer period of treatment, the inclusion of a greater number of horses or the use of different acupuncture points might have influenced this result. Another limitation of the study was the use of horses without overt lameness. It is possible that the use of horses with known painful orthopaedic conditions might have better highlighted differences between the groups. Clinical cases were not used in this preliminary study because it would have been difficult to recruit a sufficient number of cases with similar complaints of comparable severity within the time frame of the study. In addition, the withholding of treatment in the control group would not have been ethically justifiable. Furthermore, natural disease processes often change over time, making it more difficult to establish subtle treatment effects (Steiss et al. 1989). Some horses in the study showed low-grade lameness. No attempts were made to identify any underlying orthopaedic conditions as this was not one of the study objectives. However, some conditions may respond to acupuncture treatment better than others, and these effects could not be appreciated with the chosen study design.

The ideal wash-out period following acupuncture treatment is not known. Effects induced by electroacupuncture in horses with chronic thoracolumbar pain have been reported to last at least 14 days (Xie et al. 2005), and in people, pain relief for myofascial pain lasted at least 1 month (Rayegani et al. 2014). Given these findings, the present authors anticipated that only minimal residual effects, if any, would remain after 12 weeks.

The optimal number of treatments is not known. Anecdotally, many acupuncturists feel that at least three applications are necessary before it can be determined whether or not a person or animal is going to benefit from acupuncture. In earlier studies, the mean numbers of treatments necessary to see obvious improvement in horses with chronic back pain were three and four, respectively (Martin and Klide, 1987; Xie et al., 2005). More frequent treatments may therefore be necessary before the full beneficial effect becomes evident. Equally, choosing appropriate points can influence the response, as has been demonstrated in horses, in which the use of different points induced different analgesic effects (Xie et al. 2001). The best way of determining treatment points is unknown and most acupuncturists rely on a palpatory assessment, as performed in the present study. Hence, it is conceivable that the experience and individual ability of the acupuncturist may also play a role. However, a recent evaluation in people investigating the influence of style of acupuncture, location and number of needles, number, frequency and duration of sessions, and experience of acupuncturists concluded that the only variables associated with better outcomes in terms of pain relief were larger numbers of needles and more treatment sessions (MacPherson et al. 2013).

Considering the fact that the acupuncturists in the present study had 361 points from which to choose, the achievement of 20% agreement was positively surprising. However, anecdotally, certain points are more commonly used than others, potentially because they are frequently

perceived to be beneficial, easily accessible, well tolerated or a combination of these factors. Most of the acupuncture points chosen in the present study were amongst those that are anecdotally considered more popular, which probably explains the level of agreement. The high level of agreement in the identification of painful body parts suggests that both acupuncturists provoked similar responses in the study horses and interpreted them similarly. Alternatively, the large muscle groups of the neck, shoulder, proximal thoracic limb and back may be palpated more easily than, for example, the abdomen, and reactions that can be interpreted as indicating discomfort in other body parts may be more difficult to appreciate. That the level of agreement in the subjective gait assessments was only moderate is not surprising because any lameness was generally very mild. Presumably, agreement would increase with the use of lamer horses.

In summary, the results of this pilot study suggest that acupuncture treatment, as it is commonly performed in equine veterinary practice, changes the gait of horses in a positive manner. The changes could be appreciated by both objective and subjective gait analyses. These results should be confirmed in a larger population and further studies into underlying mechanisms of action, potential clinical benefits and best applications are necessary.

Figure legends:

Figure 1A and B: Lameness (Table A: 0= sound; 1=mildly lame; 2=moderately lame; 3=severely lame) and global scores (Table B; 1= lameness unchanged compared to baseline; 2= lameness better than baseline; 3= lameness worse than baseline; 0= cannot evaluate) divided by subjective gait assessors.

1A

	Lameness score	Observer 2				Total
		0	**1**	**2**	**3**	
Observer 1	**0**	126	29	0	0	155
	1	10	21	2	0	33
	2	1	0	1	2	4
	3	0	0	0	0	0
Total		137	50	3	2	192

1B

	Global score	Observer 2				Total
		0	**1**	**2**	**3**	
Observer 1	**0**	0	1	2	1	4
	1	0	1	1	3	5
	2	0	3	26	3	32
	3	0	2	3	3	8
Total		0	7	32	10	49

1   Table 1: A comparison of objective gait parameters in eight horses left untreated (control) or treated with acupuncture in a crossover design.

2   Parameters consisted of a symmetry index for head and pelvic movement (H_SI and P_SI, respectively), a measure for thoracic (T_min) and

3   pelvic (P_min) limb weight bearing, a measure for thoracic (T_max) and pelvic (P_max) limb push off, and the hip hike difference (HHD)

	Condition 1			Condition 2			Condition 3		
	Control	Treatment	p-value	Control	Treatment	p-value	Control	Treatment	p-value
H_SI (%)	98±6	107±6	0.07	99±12	101±12	0.63	100±15	101±15	0.85
T_min (mm)	-2.3±1.2	-4.52±1.3	0.21	-7.56±3.6	-9.58±3.6	0.35	4.69±6.9	4.28±6.9	0.53
T_max (mm)	-2.7±3.7	0.9±3.7	0.009	-7.9±7.3	-10.6±7.2	0.44	6.1±8.2	11.4±8.2	0.11
P_SI (%)	92±9	99±9	0.007	91±9	96±9	0.076	91±7	103±7	<0.001
P_min (mm)	2.5±3.8	-1.5±3.8	0.009	8.0±4.1	4.5±4.1	0.12	1.8±3.7	-5.7±3.7	0.002
P_max (mm)	-4.19±3.4	-1.5±3.3	0.002	0.02±3.9	2.1±3.9	0.058	-6.6±2.7	-2.9±2.7	0.002
HHD (mm)	6.3±6.4	-0.2±6.4	0.007	9.7±7.8	2.8±7.8	0.032	7.3±6.3	-2.7±6.4	0.003

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5   Condition 1: trotting in a straight line on a hard surface; conditions 2 and 3: trotting in a circle on a soft surface on the lunge on the left and right

6   reins, respectively. *Significantly different from control ($p \leq 0.05$).

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10 Table 2: Mean ± standard deviation (SD) and median (range) lameness scores assigned by
11 DMB and AF-J before (T1) and at 1 day (T7) and 7 days (T9) after the last of three
12 consecutive acupuncture treatments in eight horses receiving acupuncture (treatment) or left
13 untreated (control) in a crossover design
14

	Lameness score				OR (95% CI)	p-value
	Treatment		Control			
	Mean ± SD	Median (range)	Mean ± SD	Median (range)		
T1	0.21 ± 0.4	0(0–1)	0.26 ± 0.5	0 (0–2)	0.53 (0.24–1.1)	0.002
T7	0.17 ± 0.4	0(0–1)	0.39 ± 0.6*	0 (0–3)*		
T9	0.26 ± 0.6	0 (0–3)	0.31 ± 0.5∗,†	0 (0–2)*,†		

15 Lameness score: 0 = sound; 1 = minimally lame; 2 = mildly lame; 3 = moderately lame. OR,
16 odds ratio; 95% CI, 95% confidence interval. ∗Significantly different from control (p ≤ 0.05).
17 †n = 7 horses.
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19 Table 3: Mean ± standard deviation (SD) and median (range) global scores assigned by DMB
20 and AF-J before (T1) and at 1 day (T7) and 7 days (T9) after the last of three consecutive
21 acupuncture treatments in eight horses receiving acupuncture (treatment) or left untreated
22 (control) in a crossover design

	Global score				OR (95% CI)	p-value
	Treatment		Control			
	Mean ± SD	Median (range)	Mean ± SD	Median (range)		
T1	Baseline	Baseline	Baseline	Baseline	0.53 (0.24–1.1)	0.12
T7	1.6 ± 0.7	2.0 (0–3)	2.4 ± 0.7	2.5 (1–3)		
T9	2.0 ± 0.9	2.0 (0–3)	1.9 ± 1.0*	2.0 (0–3)*		

23 Global score: 1 = lameness better than baseline; 2 = lameness unchanged from baseline; 3 =
24 lameness worse than baseline; 0 = cannot evaluate). OR, odds ratio; 95% CI, 95% confidence
25 interval. *n = 7 horses.

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