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Case Report

Effects of Mechanical Vibration in Equine Osteoarthritis: A Pilot Study

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Abstract: The use of therapeutic ultrasounds (TUs) is widespread in both human and veterinary medicine. In fact, mechanical vibration is the simplest and purest form of vibratory energy that is applied either in physical therapies or in rehabilitation medicine. In particular, the use of low-frequency TUs to treat equine conditions is a new and evolving field. In the equine industry, osteoarthritis (OA) is one of the most challenging causes of lameness. Despite its prevalence and the advancements in its treatment, there is still no therapy whose results are completely decisive. Little is described in the literature about the use of TUs in horses' joints, particularly regarding its use to treat OA. For these reasons, the aim of this study was to preliminarily assess the efficacy of low-frequency ultrasound in two horses with metacarpo/metatarso-phalangeal joint OA. The reduction in lameness was significant in both treated cases, pointing to the effective therapeutic action of TUs. However, to better evaluate the long-term effects in athlete horses, it is necessary to include in the research a greater number of cases and a control group.

Keywords: mechanical vibrations; ultrasound; equine; osteoarthritis; physical therapies

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1. Introduction

A mechanical vibration is a periodic back-and-forth motion of the particles of an elastic body or medium; the phenomenon occurs when a physical system is displaced from its equilibrium condition and responds to the stimulation with an internal motion that tends to restore equilibrium [1]. In biological systems, it has been demonstrated that vibrations, in particular ultrasound (US), are able to activate afferent nerve fibers of the muscle spindle and, hence, the alpha motoneurons; this elicits the so-called "tonic vibration reflex". It consists of the sustained contraction of the vibrated muscle and simultaneous relaxation of its prime antagonist [2].

Therapeutic ultrasounds (TUs) are widely used both in human and veterinary medicine to treat a broad series of pathological conditions [3]. However, scientific evidence of TUs' favorable clinical effects is limited for the majority of therapeutic purposes [4].

The first studies on TUs conducted on animals were recently published [4–7], and all of them provide a clinical and little speculative imprint on the effects of TUs. Despite this, in daily clinical practice, TUs are largely used by practitioners, who describe excellent empirical results for a wide variety of orthopedic conditions, especially in the field of equine medicine. Thus, if on one hand there is a lack of supporting literature, particularly in terms of frequencies and times of application [4], on the other hand, we must obey Plautus' famous Latin axiom "*quod factum infectum fieri nequit*" (what has been done we cannot consider not done).

The mechanism through which TUs treat various pathological conditions depends on the fact that tissue absorption of soundwaves results in both a thermal effect, with increased tissue temperature and blood flow, and a non-thermal effect [8,9]. Non-thermal effects include acoustic cavitation [9]. Observations also suggest that the absorption of mechanical energy may modify gene expression, growth factors, and collagens. Together, thermal and non-thermal effects on the target tissue have been proposed to result in increased local metabolism, circulation, extensibility of connective tissue, and tissue regeneration [10].

TUs have a frequency range of 0.75–3 MHz, with most devices set at a frequency of 1 or 3 MHz. The treatment parameters taken into account are:

Frequency: The number of compression–rarefaction cycles per unit of time, expressed in cycles per second, or Hertz (Hz). Increasing the frequency of TUs causes a decrease in their depth of penetration and concentration of ultrasonic energy in the superficial tissues. There is an inverse relationship between penetration and absorption [1].

Intensity: The power per unit area of the sound head, expressed in watts per square centimeter (W/cm^2). The World Health Organization (WHO) limits the average intensity output by therapeutic US units to 3 W/cm^2 [1].

Power: the amount of acoustic energy expressed in watts (W), per unit time. In order to characterize the exposure, the total power should be specified, as well as the following intensities: spatial average temporal average (SATA) intensity; spatial peak temporal peak (SPTP) intensity; spatial peak temporal average (SPTA) intensity; and, if applicable, spatial peak pulse average (SPPA) intensity and spatial average pulse average (SAPA) intensity [1].

Pulse: TUs can be delivered as continuous or pulsed waves. The continuous mode consists of the continuous delivery of ultrasound at a single frequency throughout the treatment period; the pulsed mode consists of the intermittent delivery of TUs during the treatment period, with a non-constant pressure amplitude (equaling zero for part of the time) [1].

In the equine industry, lameness due to osteoarthritis (OA) is the most prevalent cause of diminished athletic function and wastage in racing horses [11]. The scientific and therapeutic results of the use of TUs in human medicine seem very encouraging. In particular, low-intensity ultrasound stimulation (LIPUS) has been proposed as a tool to promote the chondrogenic differentiation of mesenchymal stem cells (MSCs) [12].

The purpose of this pilot study is to use a US device—the LF Esasound (Esacrom Italy)—in two horses with metacarpo/metatarso-phalangeal joint (fetlock) OA and determine the outcome of the treatment. The major promise of this non-invasive technology is to introduce methods and approaches that are currently only hypothesized and poorly studied in OA, and/or to reduce the morbidity associated with conventional therapy. It is only reasonable that completely non-invasive procedures are the natural progression of these new and emerging technologies.

2. Materials and Methods

2.1. Ethics

The study was authorized by the Ethics Committee of the University of Bologna, n.4508/23.

2.2. Animals

The two horses included in the pilot study were presented to the Equine Surgery Department of the Faculty of Veterinary Medicine of the University of Bologna. The horses included in the study needed to present unilateral or bilateral lameness due to OA of the metacarpo/metatarso-phalangeal joint. The inclusion criteria were:

1. Lameness assessed as follows: subjective evaluation by two expert equine veterinarians (RG and RR) using the AAEP scale grade from 0 to 5, and an objective gait analysis evaluation using a markerless smartphone computer vision method (Sleip AI);
2. Positive result to flexion test;
3. Response to intra-articular diagnostic analgesia;
4. Radiographic evidence of acute or chronic, localized, or diffuse OA.

The exclusion criteria are as follows:

- Presence of osteochondritis dissecans (OCD) or articular fracture;
- Lameness due to septic arthritis;
- Joint that undergoes surgical treatment 3 months prior to the study;
- Joint that has received an intra-articular joint injection of anti-arthritic medication 3 months prior to the study;
- horses that received systemic anti-inflammatory drugs 30 days prior the study.
- Two horses met both the inclusion and exclusion criteria.

2.3. Device

The LF Esasound Esacrom Italia (Esacrom Instruments, Imola Italia) was used in this study (Figure 1). The device consists of a continuous low-frequency ultrasonic machine, which also combines a shock wave effect (non-stationary hammer effect) and a thermal effect. The thermal effect is measurable as an increase in the target tissue temperature from 3 to 8 °C from the beginning to the end of the treatment session (as reported in human medical literature) [1]. As described above, the device develops a continuous discharge of pulsed and non-stationary shock waves. The pulsed mode provides for non-thermal effects that are mainly mechanical (acoustic cavitation) [1,13]. During the delivery of pulsed US, no acoustic energy is being emitted between pulses, and, for this reason, the US propagates through the medium as small packages of acoustic energy [1].

Figure 1. Low-frequency TU shock thermic device—LF Esasound (Esacrom Italy) [1].

2.4. Study Design

The injuries outlined in this study all occurred naturally, which adds a degree of difference in terms of their entity and their severity. The two horses also differed in terms of age and breed. These variables should be taken into account when considering the different outcomes of this study.

2.5. Assessment

2.5.1. Subjective Lameness Evaluation

Each horse was examined by two expert equine veterinarians (RG and RR) with more than 10 years of equine orthopedic experience. Firstly, a physical examination and a static limb inspection were performed. Then, the horses were subjected to lameness evaluations in motion. The horses were trotted straight twice and lunged in both directions. Both clinicians were asked to subjectively assess lameness using the AAEP scale grade from 0 to 5: grade 0 was defined as no lameness, grade 1 as slight lameness at trot only, grade 2 as moderate lameness at trot only, grade 3 as slight lameness at walk and severe lameness at trot, grade 4 as obvious lameness at walk, and grade 5 as lameness produces minimal weight-bearing in motion and/or at rest or a complete inability to move [14].

2.5.2. Objective Lameness Evaluation Using an Artificial Intelligence System

At the same time, another veterinarian performed the objective gait analysis using a markerless smartphone computer vision method (Sleip AI). The system is based on a smartphone application downloaded on an iPhone 14 Pro. For this purpose, the smartphone was placed on a tripod and the video streams were first saved locally and later uploaded to the software for data processing [15]. The horses were trotted by different grooms up and down twice along a 30 m long, straight, flat, well-lighted surface to enable the 25 contiguous strides recommended by the smartphone computer vision method. Then, the horses were lunged in a 12–15 m circle by different grooms for at least 45 s in each direction to enable the 25 contiguous strides recommended by the smartphone computer vision method. The system does not require any markers placed on the horse, because the networks learn to detect the points of interest on the horse's body visible in the images' true training on large data sets [16]. Once the video is recorded, it is uploaded to the system and finally analyzed. The tool gives the operator the possibility of immediately analyzing the video, with results times that vary based on the network connection, or uploading it later [15]. In the study, all the videos were recorded and then uploaded and evaluated later. Once the video is analyzed, different screens with different information are shown for each horse. Firstly, there is a summary view that gives an overview of the asymmetries detected and the certainty of the analysis. In this section, it is also possible to consult a MinDiff/MaxDiff presentation graphic. In fact, the system is based on kinematic principles, according to which the symmetry of the horse's midline is the most sensitive parameter to quantify weight-bearing lameness. Weight-bearing lameness can cause impact lameness when the horse lands with less force, which can be visualized in the MinDiff, or push-off lameness when the horse pushes off with less force, and this can be visualized in the MaxDiff. MinDiff is the difference between two minima in the vertical position of the right and left halves of a stride, and MaxDiff is the difference between two maxima of the right and left halves of a stride [15]. MinDiff and MaxDiff will be near zero in horses with perfect movement symmetry, while, with lameness, these two values will have increasingly positive or negative values [17]. The system also shows a gait analysis screen with a ray plot of the head and pelvic movement for each stride. Every line in the graphic represents a stride, and the length of each line indicates the magnitude of asymmetry [15]. The direction in which the ray is pointing is indicative of the side and timing (impact through push-off) of the lame limb [17]. On another screen, it is also possible to see the stride-by-stride analysis video with synchronized biomechanical data [15,17].

2.5.3. Intra-Articular Diagnostic Analgesia

Intra-articular diagnostic analgesia with 8 mL of mepivacaine hydrochloride 2% was administered to confirm the source of pain. Horses with OA of the metacarpo/metatarso-phalangeal joint were included if, 15 min after the application, an improvement > 80% of the base lameness resulted. The horses were then re-examined through a subjective lameness examination.

2.5.4. Radiographic Examination

The clinical relevance of any lesion(s) was evaluated and confirmed by radiographic examination.

An overall evaluation of the severity of an individual injury was therefore made using a combination of clinical records, diagnostic imaging, and patient indications.

2.6. Selected Cases

Case 1: The first case was an 18-year-old KWPN mare. She had a history of a traumatic event to the left front fetlock that occurred during a show-jumping competition. After the trauma, a radiographic examination showed no evidence of articular pathology of the affected fetlock. In the acute phase, the mare was treated with systemic Phenylbutazone (4.4 mg/kg) twice a day for a week, followed by a total lack of exercise for 60 days. During the re-evaluation, the mare was markedly lame (grade 3 AAEP scale), both in the straight line and in the circle on both hands. At this point, an intra-articular therapy with triamcinolone acetonide (20 mg) and high molecular weight hyaluronic acid was performed. The intra-articular therapy was followed by a new period of total lack of exercise for 60 days. After 60 days, the mare presented a grade 1 lameness (AAEP scale) in the right circle, and no lameness in the left circle and straight line. A multi-directional shoeing was also applied, which resulted in the total absence of lameness. A gradual training program was started, but, 15 days after the beginning of the activity, the mare presented a grade 3 lameness (AAEP scale) both in the straight line and in the circle on both hands. A new radiographic examination was performed, which revealed a medial joint collapse associated with major osteophytosis. The US evaluation also showed marked synovitis and capsulitis. An intra-articular therapy with platelet-rich plasma (PRP) was therefore chosen: three injections at one-week intervals, with 6 months of absolute rest. At the clinical evaluation 6 months later, there was a worsening of the clinical status resulting in non-weight-bearing of the affected limb. From this moment, the worsening of the clinical situation forced the mare to undergo continuous cycles of anti-inflammatory therapies (local and systemic), which, however, did not lead to any benefit. A month after the last anti-inflammatory therapy that did not result in any improvement, it was chosen to include the mare in the study protocol. At the admission, a complete physical examination and lameness evaluation was performed. The subjective examination was made by two expert equine veterinarians (RG and RR) who detected a left front limb lameness (grade 4 AAEP scale), with the impossibility of lunging the mare on the right hand. The objective examination with the marker-less smartphone computer vision method revealed, in the straight line, a main impact (9.6) and push-off asymmetry (4.2), with also a minor mixed asymmetry for the right hind limb (0.6 push-off; 0.8 impact). In the left circle, the AI system found a mixed impact (4.9) and push-off (1.2) asymmetry for the left front limb; for the right hind limb, the AI system found a minor, probably compensatory, asymmetry (0.5 impact; 1 push-off). In the right circle, which was very difficult to assess, the AI system showed a mixed impact (8.7) and push-off (2.3) asymmetry for the left front limb, with a minor, probably compensatory, mixed asymmetry of the right hind limb (0.7 impact; 0.8 push-off) (Figure 2).

Intra-articular diagnostic analgesia of the left front fetlock was administered, and according to both clinicians, there was an 80% improvement in the initial lameness, while for the objective method, there was a 65% improvement in the impact factor and a 93% improvement in the push-off factor for the left front limb lameness, and an almost 100% improvement in the right hind limb lameness. Radiographic examination in the four standard radiographic projections (LM, DPr-Pa (Pl)DiO, D45°L-Pa (Pl)MO, D45°M-Pa (Pl)LO [18]) was performed to confirm the diagnosis and to assess the OA changes. In this severe case, the following findings were present: soft tissue swelling, periarticular osteophytes at the osteochondral margins of the III metacarpal bone (MCIII) and the first phalanx (P1), erosion of the dorsal surface on the distal end of MCIII, supracondylar lysis of the palmar cortex of MCIII, and subchondral bone sclerosis and collapse of the medial aspect of the joint [19].

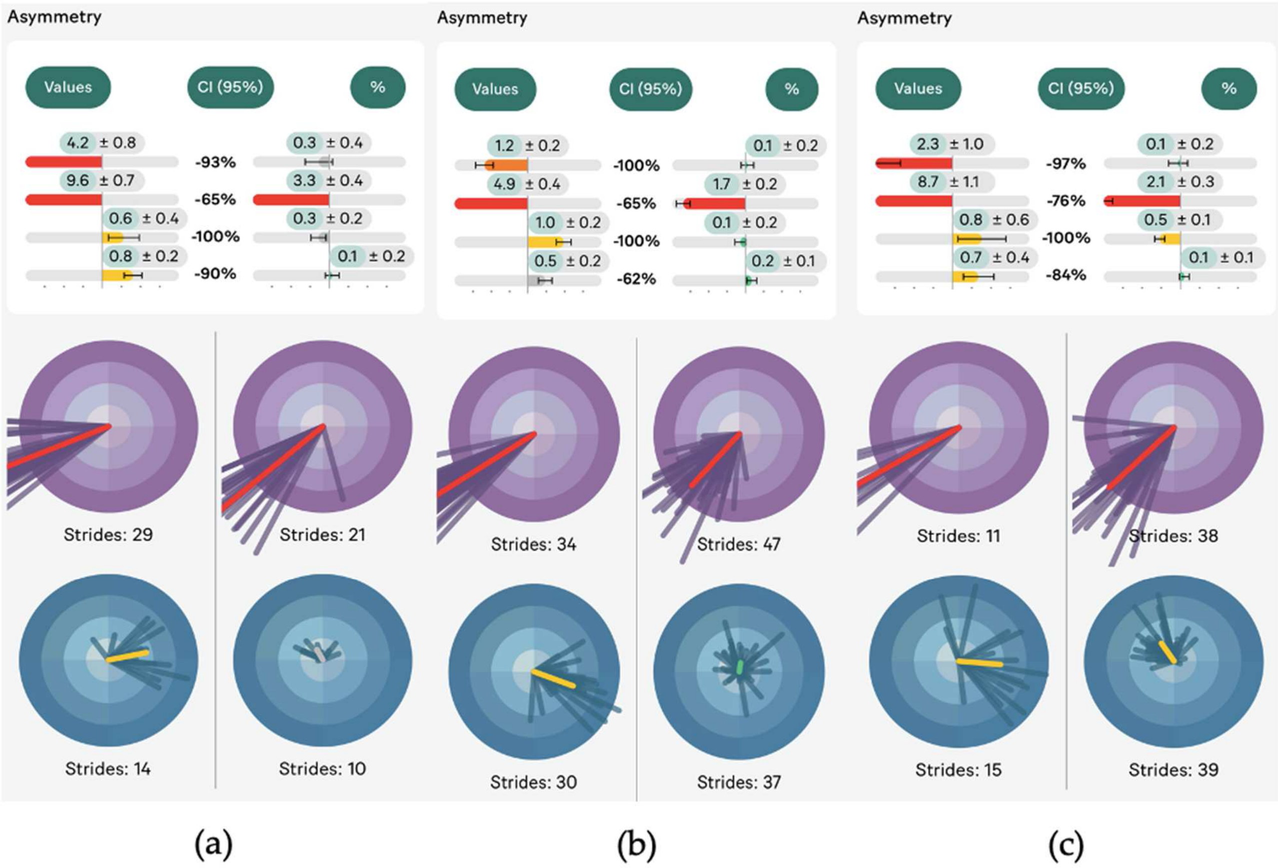

Figure 2. Objective lameness analysis from the Sleip AI system. The graphics show the movement analysis before and after the treatment, respectively, in the straight line (**a**), left circle, (**b**) and right circle (**c**), comparing the two situations and showing the percentage of improvement or worsening for each single stride component. The colors depend on the severity of the asymmetry, red is the most serious.

A week after the complete evaluation, the treatment protocol was started.

Case 2: The second case was an 8-year-old Italian Warmblood mare. The mare had a history of poor show-jumping performance. The mare underwent a clinical examination, followed by diagnostic anesthesia, and a radiographic examination. A diagnosis of bilateral lameness due to a degenerative arthropathy in each metatarsophalangeal joint was made. Intra-articular treatment was then administered using 10 mg of triamcinolone acetonide and high molecular weight hyaluronic acid in both hind limb fetlocks. As a result of the treatment, the mare showed an improvement, allowing her to return to training. However, after a few times in training, there was a gradual worsening of the initial condition. Several intra-articular treatments with hyaluronic acid were performed, which did not yield positive results. A month after the last intra-articular treatment, which did not result in any improvement, it was chosen to include the mare in the study protocol. At the admission, a complete physical examination and lameness evaluation was performed. The subjective examination was made by two expert equine veterinarians (RG and RR), who detected a bilateral hind limb lameness (grade 2 AAEP scale for the left hind limb and grade 3 AAEP scale for the right hind limb). The objective examination with the markerless smartphone computer vision method revealed, in the straight line, a main left hind limb mixed impact (0.7) and push-off asymmetry (0.7), with also a minor mixed asymmetry for the left front limb (0.4 impact-0.5 push-off). In the left circle, the AI system found a mixed impact (0.6) and push-off (0.5) asymmetry for the left hind limb; for the right front limb, the AI system found a minor, probably compensatory, asymmetry (0.4 impact; 0.3 push-off). In the right circle, the AI system showed a mixed impact (0.8) and push-off (0.8) asymmetry for the left hind limb, with a minor, probably compensatory, push-off asymmetry of the right front limb (0.4 push-off) (Figure 3). Intra-articular diagnostic analgesia of both hindlimb fetlocks was administered, and both clinicians and the smartphone vision method identified an 80% improvement in the initial lameness. Radiographic examination in the four standard radiographic projections (LM, DPr-Pa (Pl)DiO, D45°L-Pa (Pl)MO, and D45°M-Pa (Pl)LO [18]) was performed to confirm the diagnosis and to assess the OA changes. A week after the assessment, the treatment protocol was started. In this case, the OA findings were soft tissue swelling and periarticular osteophytes at the osteochondral margins of the first phalanx (P1) [19]. In addition, a US examination was executed, highlighting signs of synovitis in both hind limb fetlocks. A week after the complete evaluation, the treatment protocol was started.

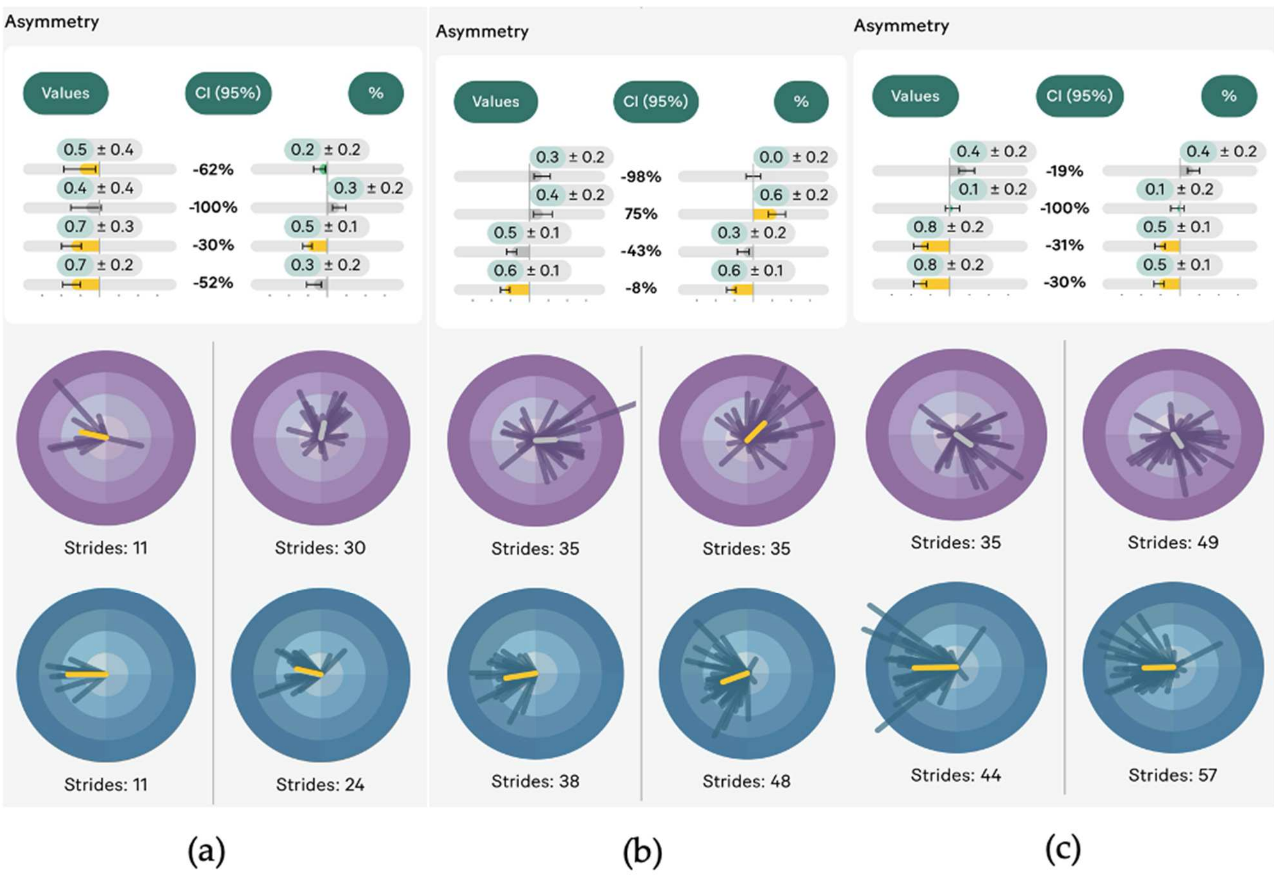

Figure 3. Objective lameness analysis from the Sleip AI system. The graphics show the movement analysis before and after the treatment, respectively, in the straight line (**a**), left circle (**b**), and right circle (**c**), comparing the two situations and showing the percentage of improvement or worsening for each single stride component. The colors depend on the severity of the asymmetry, red is the most serious.

2.7. Treatment Protocol

Firstly, the safety of the practitioner, horse, and equipment were always considered. Horses were prepared for the treatment in such a way as to prevent uncontrolled kicking, distress, or pain. The stress and/or pain of each subject was estimated during the treatment procedures, where such traits as (1) tendency to move, (2) moving forward or backward and or side by side, (3) repeated, insisted pawing, and (4) sudden, hasty limb subtraction were relied on. The device does not require treatment areas to be shaved. US-specific gel was applied to both the contact surface of the machine and directly to the treatment area. This allows a better US penetration and also assists the transducer head to smoothly glide over the selected treatment area. Subsequent applications of US gel were made when it was felt that insufficient gel was present at the treatment site. The device parameters used to treat the metacarpo/metatarso-phalangeal joint were a power of 60 W and a pulse of 80 (hammer effect), with 5 min of treatment per single joint. These parameters were used both according to the manufacturer's instructions and derived from experience in treating shoulder and elbow arthropathies in human patients [1], and additionally, with respect to exposure time carried out on rats, mice, and pigs [20–22]. The treatment period included two treatments per week for three weeks, as indicated by the literature on the use of TUs [1,13,23].

No sporting activities were allowed for the whole treatment duration.

2.8. Re-Assessment

A week after the last treatment, both horses were re-evaluated by physical examination, static inspection, and dynamic subjective and objective lameness evaluation. Three months after the last treatment, to confirm the results' continuity, a telephone follow-up was conducted.

3. Results

Case 1: One week after the last treatment, the mare presented, in the straight line and lunge in both directions, grade 2 lameness (AAEP scale) according to one veterinarian (RR), and in the straight line and lunge in both directions, grade 3 lameness (AAEP scale) according to the other veterinarian (RG). The mare did not show any lameness at the walk for both clinicians. The objective movement analysis revealed in the straight line an improvement from the original lameness to a 3.3 impact and 0.3 push-off asymmetry. For the left circle, the AI system found an improvement in the original lameness to a 1.7 impact left front limb asymmetry, with the disappearance of the front push-off and the right hind limb asymmetry. For the right circle, the AI system showed an improvement in the original lameness to an impact asymmetry of 2.1, with the disappearance of the front push-off asymmetry, and an improvement of the right hind limb asymmetry to a 0.5 push-off asymmetry, with the disappearance of the impact asymmetry. The total estimated improvement rate is about 70% (Figure 2).

Through phone contact 3 months after the end of the treatment, the owner reported the permanence of the results, which allowed the mare to live in a paddock without needing any anti-inflammatory therapies.

Case 2: A week after the last treatment, for both clinicians, the mare was free from straight-line lameness. The horse presented, according to both clinicians, a grade 1 lameness (AAEP scale) in the left hind in the left circle, and a grade 1 lameness (AAEP scale) in the right hind in the right circle. The objective movement analysis revealed, in the straight line, an improvement from the original lameness to a mixed left hind limb impact (0.3) and push-off asymmetry (0.5), with the disappearance of the left front limb asymmetry. For the left circle, the AI system found an improvement for the left hind limb to a mixed impact (0.6) and push-off (0.3) asymmetry, while for the right front limb, the AI system found a worsening to a 0.6 push-off asymmetry with the disappearance of the impact factor asymmetry. In the right circle, the AI system showed an improvement in the original lameness to a mixed impact (0.5) and push-off (0.5) asymmetry, and also maintenance of the minor, probably compensatory, push-off asymmetry of the right front limb (0.4 push-off). The total estimated improvement rate is about 30% (Figure 3). Through phone contact 3 months after the end of the treatment, the owner reported the permanence of the result. Due to the mare's pregnancy at that time, it was not possible to evaluate her sporting performance after the treatment.

4. Discussion

TUs are used both in companion animals and athletes to treat various diseases and injuries affecting tendons, ligaments, muscles, joints, and bones. There is limited information about the use of TUs in horse joints in the literature, particularly regarding the treatment of

OA. To date, few veterinary medicine studies on the topic have been published. Moreover, the little scientific knowledge on the effects of TUs on dogs OA comes from a single cohort study without a control group and with a high risk of bias [24].

In particular, there are no studies about OA in horses, although there are two studies on traumatic arthritis in donkeys [25,26]. In the four donkeys assigned to TU, the authors observed earlier improvement of lameness, faster reduction of joint swelling, and less pain at flexion than in the other four, non-treated donkeys. Synovial cytological and biochemical analyses indicated less inflammation in the TUs group. In addition, the histomorphological examination revealed signs of improved healing and reduced signs of cartilage degeneration in TU-treated donkeys [25,26].

The present study was mainly designed starting from the need to clarify the use of TUs in horses and to better understand its clinical effects on spontaneous onset OA. To begin with, we inquired about the operating mechanism of TUs. We know from experiments conducted on healthy horses that the temperature changes in the tendons of treated horses [27]. In fact, in the present study, at the end of a 10- to 20-minute treatment session, the temperature had increased by 2.5–5.2 °C in the superficial and deep digital flexor tendons of the thoracic limb (one 10 min session, continuous mode of 3.3 MHz). Another study showed that a 1° increase in temperature triggers metabolic activity, 2° to 3° decreases muscle spasms and increases blood flow, and 4° changes the viscoelastic properties of collagen [1,13]

US propagation in biological tissue is fundamentally a non-linear process. This non-linearity becomes more dominant with the increase in intensity, where secondary phenomena, which are not predicted from a purely linear propagation model, begin to emerge [28]. These effects include cavitation, radiation forces, and micro-streaming [29]. Except for cavitation, the magnitudes of the other mechanisms are often insignificant [30].

The cavitation effect is the most widely known. It consists of the formation, growth, and implosion of gas bubbles within the fluid subjected to an ultrasonic field. The gas bubbles are generated and then expand and contract in tissue when ultrasonic pressure goes from a positive peak to a negative peak [31]. Despite the fact that cavitation nuclei are not present in human and animal tissues and blood, we assume a priori that nuclei exist, and we focus instead on the dynamics of an acoustically small gas bubble in a Newtonian fluid undergoing radial oscillations—all key approximations intended to simplify the model while retaining most of the essential underlying physics [4,32]. In particular, the action of a specific intense ultrasonic field within a liquid is called “acoustic cavitation” and is generally divided into two categories: stable and transient.

Stable cavitation occurs from low to moderate pressure amplitudes (less than w1 MPa with an inverse dependence on frequency). A radially oscillating bubble inside a liquid medium can grow in an acoustic pressure field as a result of rectified diffusion. In this process, during the expansion half cycle, due to the decreasing pressure inside the bubble, some of the liquid surrounding the bubble can diffuse through the boundary layer, turning into vapor inside the bubble. In the subsequent contraction half cycle, as a result of the increase in pressure inside the bubble, some of this vapor will condense and diffuse out. This diffusion is “rectified” because the surface area of a bubble in the expanded state is much greater than in the contracted state, providing a greater area for diffusion of the gas into the bubble than for diffusion out of it. Rectified diffusion favors bubble growth over subsequent oscillations as a result of the increase in gas inside the bubble [33].

On the contrary, transient cavitation occurs when the process described above occurs quickly, and the bubble experiences rapid growth over a few cycles, followed by a collapse during the positive half cycle as a consequence of the inertia of the spherically converging liquid [34].

There is an optimum range for the initial size of a bubble that can undergo transient cavitation at the threshold pressure amplitude of a wave with frequency (f). In general, this optimum size is in the order of $Rr/3$, where Rr is the radius of the bubbles resonating at frequency f, as in the following formula:

$$Rr = 3.28/f \ (Rr \geq 0.01 \text{ mm}).$$

Accordingly, it is reasonable to expect that the influence of transient cavitation will be more significant at lower ultrasonic frequencies [34,35].

The effects of cavitation depend on the characteristic parameters of the liquid. This is demonstrated by the model developed by Holland and Apfel [36]. This theory presented an approximate analytic prediction of the thresholds of cavitation as a function of the frequency of insonification, the initial nuclei radius, the specified final collapse temperature, and the host parameters (surface tension, viscosity, and density). Thus, the connection between TUs and this theory depends on the nature of the damage and the sensitivity of the subject. It is important to search for regions where cavitation could occur in vivo along with the possible biological manifestation of this phenomena. The existence of optimal-size nuclei, corresponding to the applied US parameters, should be investigated [36]. Based on this, it can be hypothesized that their effect in horse synovial fluid or blood, for example, will be different from that in humans or other animal species.

The present pilot study was created with the aim of understanding if all the effects described above really work in reducing the pain and symptoms of equine OA. In our opinion, beyond the scarcity of studies present in the literature, the main problem is the standardization of the vibration platforms used to treat patients, application times, and intensity. In addition, some studies used pulsed US and other continuous US. Moreover, even though there is more in the literature about the effects of TUs in human medicine, the effectiveness of US for treating people with pain, musculoskeletal injuries, and soft tissue lesions remains questionable. In fact, most papers are systematic reviews of randomized controlled trials, in which the US was used to treat people with those conditions. Each trial was designed to investigate the contributions of active and placebo US to the patient outcomes measured. Unfortunately, no conclusion was drawn by any of the studies [12,37].

In the present study, regarding treatment times, we based ours on the indications provided by veterinary medicine studies—2–6 treatment sessions weekly for up to 3–4 weeks [4]. We chose two treatments a week for three weeks to see if there were improvements with a few weekly treatments and in a few weeks.

We chose the platform based on the evidence that continuous TUs have better efficacy in human medicine, where the maximum efficiency of the signal for each treated tissue is always guaranteed [1]. Devices that deliver continuous TUs allow for increasing the temperature of the target tissue by 3–8 °C by the end of the treatment session. The thermal effect generated by continuous TUs triggers biological changes to occur in the target tissues, resulting in the relief of sub-chronic and chronic pain, muscle spasms, and joint contractures [1].

Another peculiarity of this device is that it has a non-stationary hammer effect (shock wave), which can help break down any mineralization in the joint capsule. Concerning the power, intensity, and frequency, we set the device according to the manufacturer's instructions and the experience of two human orthopedic specialists who used the device in their patients' shoulder arthritis with excellent results.

The results of this first study are encouraging. In fact, neither patient responded to conventional therapies, whereas both horses showed an improvement in lameness, according to two expert clinicians and the video analytical system used. The artificial intelligence system (AI system) defined a computed percentage improvement of almost 70% in the first case and of almost 30% in the second case, representing the movement analysis. However, following this experience, the need to conduct a clinical study with a greater number of cases, a standard degree of lameness, and a control group undergoing traditional therapies for OA is evident. Tests should also be conducted regarding frequency, power, and intensity to find the best combination.

A further aspect to investigate is that of the thermal effects in OA, considering the heterogeneity between studies and the low number of studies for each combination of species and indication. In athlete horses, experimental tests should be carried out on the temperature of the tissues after the treatment, and it is also important to investigate how long this effect lasts.

Regarding the application time, we based our study on the state of the art on the harmful effects of US concerning exposure time carried out on rats, mice, and pigs [20–22].

5. Conclusions

In conclusion, it can be stated that the proposed treatment had an effective therapeutic action in horse OA. However, little is known about its exact mechanism(s) of action, and many potential applications need to be explored, taking care to ensure adequate size samples in the studies, accurately select the type of patients to enroll, and ensure the presence of a control group, always respecting up-to-date protocols, to eventually codify the role of vibration therapy in physical and rehabilitation veterinary medicine.

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