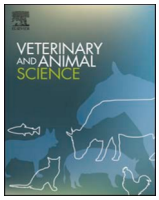
Veterinary and Animal Science 23 (2024) 100330

Contents lists available at ScienceDirect

Veterinary and Animal Science

journal homepage: www.elsevier.com/locate/vas



Effects of intra-articular administration of hyaluronic acid or platelet-rich plasma as a complementary treatment to arthroscopy in horses with osteochondritis dissecans

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ARTICLE INFO

Keywords:
Arthroscopy
Lameness
Synovial fluid
Osteochondrosis
Horse

ABSTRACT

Although arthroscopy is the treatment of choice for horses with osteochondritis dissecans (OCD), it is not yet known whether intra-articular therapies in the postoperative period can bring any benefit to the recovery of these animals. This study evaluated the effects of the intra-articular application of platelet-rich plasma (PRP), hyaluronic acid (HA) or lactated Ringer's solution (LR) in horses with OCD undergoing arthroscopy. Eighteen male and female Brazilian Sport horses aged between 2 and 6 years were evaluated. All animals presented OCD fragments in the middle crest of the tibia. Ten days after surgery, animals were randomly distributed into three groups and received intra-articular application of PRP ($n = 6$), HA ($n = 6$), or LR (control group, $n = 6$). Clinical, radiographic, ultrasound and synovial fluid evaluations were performed on the day of surgery and after 10, 30 and 60 days. An increase in the thickness of the joint capsule was observed 30 days after surgery in the three groups evaluated. In the control group, there was significant improvement in the flexion test 30 and 60 days after surgery, and in the PRP group, there was worsening of this parameter in the same evaluations. In the control group, there was a reduction in the degree of synovial effusion, and in the PRP and HA groups, there was increased effusion. There was a significant increase in the number of leukocytes in the HA group. Intra-articular use of PRP or HA ten days after arthroscopy did not promote positive effects on the recovery of horses with OCD.

1. Introduction

Osteochondritis dissecans (OCD) and osteochondrosis (OC) are terms frequently used to describe a defect in endochondral ossification that leads to osteochondral fragmentation and/or appearance of cysts in the joints of foals; however, they describe different stages of the disease. OC represents the initial phase of the process, while OCD reflects secondary changes that result in cartilage flap or osteochondral fragment formation (Semevolos, 2017).

Currently, it is known that OCD represents an enormous problem for the equine industry because in competition horses, lameness is caused mainly by lesions resulting from this disease, which drastically reduce the athletic performance and welfare of the affected animals (McIlwraith, 2005). Bone and joint disorders are also the main limiting factors that affect the reproductive potential and market value of horses. Therefore, it is of great importance to develop new strategies to improve the therapeutic management of OCD in horses (McIlwraith, 2013).

In horses, the joints most affected by osteochondrosis are the tibiotarsal joint, followed by the femoropatellar and femorotibial joint, scapulohumeral joint, and metacarpal/tarsal phalangeal joint. The treatment of chronic injuries or injuries with a slow clinical evolution has become a challenge, mainly due to the inadequate reorganization of the injured tissue, as well as the high rate of relapse, determining the end of the athlete's career or its inappropriate performance on return to sport (Harrison & Edwards, 1996).

The definitive diagnostic method and treatment of osteochondrosis dissecans is video arthroscopy of the joint. A minimally invasive technique, not causing extensive damage to the joint capsule, in addition to

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https://doi.org/10.1016/j.vas.2023.100330

Available online 23 December 2023

being efficient in removing fragments, cleaning degenerated tissues and inspecting all anatomical structures of the affected joint. When osteochondrosis dissecans is diagnosed and treated through video arthroscopy early, the condition has a good prognosis, allowing the animal to return to its normal athletic function. The progression of the process to osteoarthritis and the consequent chronic and/or irreversible injuries of the joint is avoided (Semevolos, 2017; McIlwraith, 2013).

In the tibial tarsal joint, the main clinical sign of osteochondrosis dissecans is synovial effusion, and video arthroscopy surgery is recommended in 100 % of cases to remove the fragments, since conservative treatment for this disease is not effective for the main clinical symptom. Likewise, conservative treatment does not prevent the progression of the osteoarthrosis process, since the negative stimulus of the articular fragment remains present (McIlwraith, 2013; Harrison & Edwards, 1996).

In this context, the use of autologous material as an adjuvant in the healing process has been widely studied. Among them, many blood products have been studied, such as platelet-rich plasma (PRP) or plasma rich in growth factors, autologous conditioned serum, autologous blood preparations, and autologous protein concentrates (Ziltener et al., 2012; Chevalier, 2010). Hyaluronic acid (HA) is responsible for most of the viscoelastic properties of synovial fluid, and its intra-articular use has been proposed as a potential treatment for joint diseases for a long time (Howard & McIlwraith, 1993). In addition to the nutritional function of articular cartilage, hyaluronic acid has the ability to lubricate the joint, and although its half-life is only thirteen hours, the clinical benefits of its therapeutic use are maintained for weeks to months (Coronado et al., 2015; Stashak, 2002; Trotter & McIlwraith, 1996).

The objective of this study was to evaluate and compare the effects of intra-articular administration of hyaluronic acid and platelet-rich plasma in the postoperative period of horses with OCD lesions in the middle crest of the tibia.

2. Material and methods

2.1. Animals

Eighteen Brazilian Sport Horse breed, nine females and nine males, mean age of 3,5 ± 1,3 years and mean body weight of 500±100 kg, were included in this study. All animals presented OCD fragments in the middle crest of the tibia, diagnosed by radiographic examination (Fig. 1) and were referred for surgical arthroscopy treatment.

This study was approved by the Committee on Ethics in the Use of Animals of Faculty of Animal Science and Food Engineering, University of São Paulo (USP), approval number 1310211116.

2.2. Arthroscopy

After 6 h of fasting, the horses received 2 % xylazine (0.5 mg kg1 IV) as pre-anesthetic medication; after 10 min, the animals received guaiacol glyceryl ether (100 mg kg1 IV) for myorelaxation. Anesthetic induction was conducted with 10 % ketamine (1 mg kg1 IV) and midazolam (0.1 mg kg1 IV) mixed in the same syringe; anesthesia was maintained with isofluorane and controlled ventilation. The same surgeon performed all surgical procedures. The animals were positioned in dorsal decubitus, and the region of the tibiotarsal joint was clipped. The surgical site was prepared for surgery by scrubbing the area with chlorhexidine scrub for 3 min, followed by chlorhexidine solution and 70 % isopropyl alcohol application. After antisepsis was completed, the sterile disposable field cloths were positioned to isolate the prepared region, and all material for video arthroscopy surgery was connected. At this point, synovial fluid was collected.

The dorsomedial access was used, using as a reference, 2 cm below the medial malleolus of the tibia, in the region lateral to the saphenous vein and medial to the cranial tibial, peroneus tertius and long digital extensor tendons. After making an incision of approximately 2 cm of skin, the joint is inflated with sterile lactated Ringer's solution, with the aid of an arthroscopic inflation pump. When the joint is well pressurized, entry begins, carefully, with the sharp trocar, and as soon as the joint capsule is surpassed, it is switched to the blunt trocar.

Once in the joint, all compartments of the tibiotarsal joint, the lateral, dorsal, ventral and medial aspects were evaluated to observe and determine signs of degeneration. After this initial assessment, the lens was positioned on the intermediate crest of the tibia bone to document the size of the dissecting osteochondrosis fragment, and with a 40×10 hypodermic needle, the best instrument access was determined laterally to release the fragment, in order to allow its removal. The skin and subcutaneous tissue were incised with a #11 scalpel blade, and then the joint capsule was also incised. Once the instrument portal was created, the fragment was evaluated with the explorer, loosened with a periosteal elevator, and removed with Ferris Smith Router forceps (Fig. 1). Once the fragment was removed, debridement of the degenerated tissue of the tibial sagittal crest bed was performed, in order to establish a suitable location for the fibrocartilage healing process. The instruments were removed and the skin was sutured with 0 nylon, in a simple separate pattern. The animals received compressive bandages, which were changed every 2 days, and the stitches were removed after 14 days. The animals rest in a stable for 60 days, leaving only for a brief walk.

2.3. Treatments

On the tenth day after arthroscopy (D10), the animals were randomly divided into three groups and received an intra-articular injection of 4 ml of PRP (Arthrex ®, PRP Group, $n = 6$), 2 ml of hyaluronic acid (Fermathron ®, HA Group, $n = 6$), or 4 ml of Lactate Ringer's Solution (Control Group, $n = 6$). The injections were performed in the tibiotarsal joint medial to the saphenous vein, following the routine antisepsis protocols for joint puncture.

PRP was prepared following the manufacturer recommendations. A straight needle was used to withdraw 1.5 mL ACD-A into a syringe. Puncture of the left jugular vein was performed, and blood flashed in the tubing. Slowly withdraw was performed by pulling back on the red wings of the Arthrex ® syringe. The syringe was filled to a maximum of 16 mL venous blood at a rate of 1 mL every 2 s. After blood draw was completed, the butterfly needle was manually pinched against the luer tip of the ACP syringe. The syringe was sealed with the red cap. The syringe was gently inverted to mix blood and ACD-A and was placed into one bucket of the centrifuge and an appropriate-size counterbalance in the opposite bucket. The centrifuge was runned at 1500 rpm for 5 min. The syringe was removed from the centrifuge, taking care to keep it in an upright position to avoid mixing the plasma and red blood cells. In order to transfer 4 mL of Arthrex ® PRP from the larger outer syringe into the small inner syringe, the syringe's red wings were slowly pushed down and the small inner syringe was unscrewed. The ACP was immediately used.

2.4. Radiographic evaluation

A radiographic evaluation of the tibio-tarsal joint, with a dorsomedial-plantar-lateral oblique projection, was performed on the day of surgery (D0) to confirm the diagnosis of OCD with a fragment in the middle crest of the tibia. The exam was repeated 30 days after arthroscopy (D30) to evaluate the existence of radiographic changes and possible differences between the groups.

2.5. Ultrasound evaluation

On the day of surgery (D0), ultrasound was performed to measure the thickness of the joint capsule. The measurement was performed on the medial surface of the tibiotarsal joint, medial to the saphenous vein, as the joint space is large in this region, allowing for better visualization,

Fig. 1. Ilustration of OCD in the intermediate crest of the tibia (A); arthroscopy surgery to OCD removal (B); OCD fragment (C); area of degeneration caused by the OCD (D); PRP (E); Hyaluronic acid (F); Ringer's lactate solution (G); study design.

and would not be used as a portal site for video arthroscopy. The evaluation was repeated on D30 postoperatively at the same site as the first evaluation.

2.6. Flexion test

The flexion test of the tarsal tibial joint, as described by Stashak (2002), was performed on the day of surgery (D0) and repeated after 30 (D30) and 60 (D60) days. The response to the flexion test was graded and classified on a scale of 0 to 3, being 0 - no pain response; 1 - mild pain response; 2 - moderate pain response; and 3 - severe pain response.

2.7. Evaluation of synovial effusion

The macroscopic evaluation of the degree of synovial effusion of the tibio-tarsal joint was performed before surgery (D0) and repeated 10 (D10), 30 (D30) and 60 (D60) days postoperatively. A classification from 0 to 3 was adopted as follows: 0 – absence of synovial effusion; 1 - mild synovial effusion; 2 - moderate synovial effusion; and 3 - severe synovial effusion.

2.8. Synovial fluid analysis

After classifying the degree of synovial effusion, antisepsis was

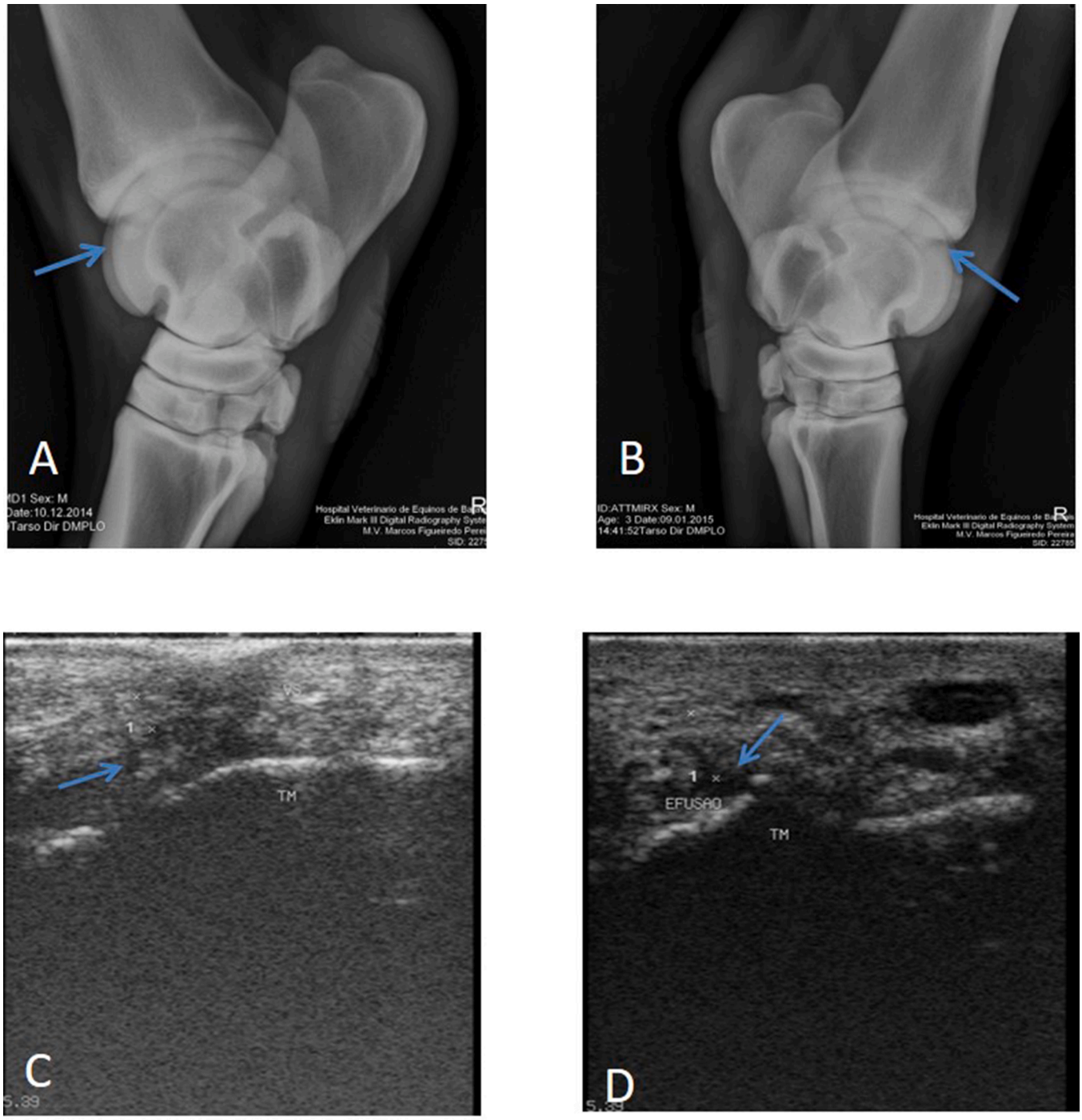

Fig. 2. Ilustration of radiographic and ultrassonographic images of OCD in the intermediate crest of the tibia in D0 (A; C) and in D30 (B; D).

performed, and synovial fluid was collected using a 40×10 needle in the medial bursa of the tibiotarsal joint, medial to the saphenous vein. Two ml of synovial fluid was collected for visual and cytological laboratory evaluation. Samples were collected on D0 (immediately before surgery), D10 (10 days after arthroscopy and immediately before intra-articular administration of the treatments) and D30 (30 days after arthroscopy and 20 days after intra-articular treatment).

2.9. Statistical analysis

The results were subjected to the Kolmogorov–Smirnov normality test. The normal data, protein concentration and lymphocytes were analysed in relation to D0 by means of analysis of variance (ANOVA), followed by the Bonferroni posttest. The data referring to the thickness of the joint capsule were subjected to the paired *t*-test.

The values that did not follow the normal distribution, leukocyte concentration, neutrophils, segmented neutrophils, monocytes, joint effusion, and flexion test were analysed in relation to D0 using the Kruskal–Wallis test, followed by Dunn's correction.

The time-point comparisons between the groups were performed using two-way analysis of variance followed by Tukey's posttest. The normal variables are expressed as the mean ± standard deviation, while the nonparametric variables are expressed as the median and the minimum and maximum values. For all data, a significance level of $P < 0.05$ was considered.

3. Results

3.1. Radiographic evaluation

The surgical procedure and adjuvant therapies did not cause negative radiological changes in the joint on D30 (Fig. 2).

3.2. Ultrasound evaluation

The thickness of the joint capsule increased significantly in the three Groups 30 days after surgery (D30) (Table 1 and Fig. 2). The HA group had lower values for joint capsule thickness in the first evaluation (before surgery), but on D30, the thickness increased, equalling the other groups.

3.3. Flexion test

The animals that received treatment with lactated Ringer's solution (control group) showed improvement in the joint flexion test on D30 and D60 compared to the test performed before surgery (D0). The PRP group, on the other hand, showed a worsening of the response to the flexion test on D30 and D60, and the group treated with hyaluronic acid (HA) showed no change in the flexion test performed on D0, D30 and D60 (Table 2).

Table 1
Mean ± standard deviation of joint capsule thickness (mm) of animals with OCD treated with lactate Ringer's solution (LR), hyaluronic acid (HA), and platelet-rich plasma (PRP). The evaluations were performed on D0 (before arthroscopy) and on D30 (thirty days after arthroscopy and twenty days after intra-articular treatment).

Evaluation	Groups	D0	D30	P value
Capsule thickness	LR	5.4 ± 1.1	7.5 ± 1.5*	0.0194
	HA	3.6 ± 0.4[a]	7.8 ± 1.1*	0.0001
	PRP	5.5 ± 1.0	8.8 ± 0.8*	0.0007

*indicates difference in momentum relative to D0. Significance level $P < 0.05$.
[a] indicates the difference between groups. Significance level $P < 0.05$.

Table 2
Median (minimum and maximum) of the response to the joint flexion test of animals with OCD treated with Lactate Ringer's solution (LR), hyaluronic acid (HA), and platelet-rich plasma (PRP). The evaluations were performed on D0 (before arthroscopy) and repeated on Days 30 (D30) and 60 (D60).

Evaluation	Groups	D0	D30	D60
Flexion Test	LR	1 (1-2)	0 (0-1)*	0 (0-1)*
	HA	0 (0-0)	0 (0-0)	0 (0-0)
	PRP	0 (0-1)	1 (1-1) *	1 (1-1) *

*indicates difference in momentum relative to D0. Significance level $P < 0.05$.

3.4. Evaluation of synovial effusion

In the evaluation of joint synovial effusion, in relation to D0, the group treated with lactated Ringer's solution (LR) showed reduced values on D30 and D60. On the other hand, the HA and PRP groups showed an increase in joint synovial effusion in relation to D0. In the HA group, an increase was observed on D30, and in the PRP group, an increase was observed on D10 and D30 (Table 3).

3.5. Synovial fluid analysis

In the laboratory analysis of synovial fluid, although there were significant changes between times and groups, the values remained within the normal range for the species. On D30, the HA group showed a significant increase in leukocyte count compared to the counts on D0 and D10 (Table 4 and Fig. 3).

Comparing the three groups, the number of leukocytes in the HA group on D30 was significantly higher than that found in the LR and PRP groups (Table 4 and Fig. 3). The mean values of protein concentration did not differ over time or between groups.

4. Discussion

This study demonstrated that the use of intra-articular PRP or hyaluronic acid in the recent post-operative period, as a complementary treatment to arthroscopy in horses with osteochondritis dissecans promoted an increase in synovial effusion, when compared to Ringer's lactate solution.

Ultrasound is an auxiliary diagnostic technique in cases of OCD and has been widely used to evaluate the appearance of the synovial fluid, changes in the bone silhouette, and the thickness of the joint capsule (Raes et al., 2010). In the present study, the ultrasound evaluation performed 30 days after surgery showed a significant increase in the thickness of the joint capsule in the three groups evaluated, suggesting that there was no influence of treatment on this parameter and that the capsule thickening is probably due to arthroscopy, as demonstrated by Barcelos (2012).

The improvement in the flexion test shown by the animals of the control group at 30 and 60 days after arthroscopy indicates a positive effect of the surgical procedure on the clinical condition. On the other hand, the worsening observed in the PRP group suggests that this intra-articular therapy performed 10 days after arthroscopy may have caused a deleterious effect. Therefore, great criteria must be used in the

Table 3
Median (minimum and maximum) of the evaluation of synovial fluid effusion of animals with OCD treated with lactated Ringer's solution (LR), hyaluronic acid (HA), and platelet-rich plasma (PRP). The evaluations were performed on D0 (before arthroscopy) and repeated on Days 10 (D10), 30 (D30) and 60 (D60).

Evaluation	Groups	D0	D10	D30	D60
Synovial effusion	LR	1 (1-1)	1 (0-1)	0 (0-1)*	0 (0-1)*
	HA	0 (0-0)	0 (0-0)	1 (1-1)*	0 (0-0)
	PRP	1 (0-1)	1 (1-1)*	1 (1-1)*	1 (0-1)

*indicates difference in momentum relative to D0. Significance level $P < 0.05$.

Table 4
Medians (minimum and maximum) of leukocyte count (leucocytes/mm3) and mean ± standard deviation of protein concentration (g/dL) of synovial fluid of horses with OCD treated with lactate Ringer's solution (LR), hyaluronic acid (HA), and platelet-rich plasma (PRP). The collections were performed on Days 0 (D0, before arthroscopy), 10 (D10, before intra-articular administration of the treatments), and 30 (D30).

Evaluation	Groups	D0	D10	D30
Total Leukocytes	LR	0.2 (0.1-0.2)	0.35 (0.2-0.6)	0.3 (0.2-0.7)[a]
	HA	0.2 (0.2-2.3)	0.2 (0.1-0.3)	1.15 (0.2-2)*[b]
	PRP	0.1 (0.1-0.2)	0.35 (0.1-0.4)	0.2 (0.1-0.3)[a]
Protein	LR	1.3 ± 0.2	1.1 ± 0.6	1.0 ± 0.2
	HA	1.0 ± 0.2	0.7 ± 0.1	1.6 ± 0.6
	PRP	1.1 ± 0.7	1.2 ± 0.6	1.7 ± 1.0

*indicates a difference in momentum relative to D0. Significance level $P < 0.05$. ab indicate differences between groups. Significance level $P < 0.05$.

interpretation of clinical results with the use of PRP. In a systematic review of the therapeutic use of PRP in orthopaedic injuries in humans and horses, Brossi et al. (2015) evaluated 60 clinical studies and found positive results in 46.7 % and negative results in 43.3 %. However, all the publications that presented positive results in the equine species did not have a control group.

According to Moraes et al. (2015), the reports on the efficacy of PRP in the joint and its anabolic and catabolic effects in the synovial environment still need to be better clarified because there are still few well-designed clinical studies that prove the therapeutic efficacy in horses with arthropathies, as well as how the responses of the environment articulate the *in situ* application of PRP. The tissue repair process is active in case of osteoarthritis and is led by some growth factors (Chevalier, 2010). However, when the joint presents an associated inflammatory process, the chondrocytes fail in this response in relation to insulin-like growth factor 1, and present an unconventional response to transforming growth factor β. Platelets have growth factors such as transforming growth factor β (TGF-β), insulin-like growth factor 1 (IGF-1), platelet-derived growth factor (PDGF), vascular endothelial growth factor (VEGF), fibroblast growth factor (FGF), epidermal growth factor (EGF) and platelet-derived epidermal growth factor (PDEGF). These growth factors promote increased gene transcription, proliferation and cell differentiation (Ziltener et al., 2012). It is hypothesized that these growth factors contained in PRP may contribute to the regeneration of articular cartilage, since TGF-β increases the phenotypic expression of chondrocytes, chondrogenic differentiation of mesenchymal stem cells, disposition of extracellular matrix, in addition to decreasing suppressive effects of the inflammatory mediator interleukin 1 on proteoglycan synthesis in cartilage. PDGF increases chondrocyte proliferation and proteoglycan synthesis and IGF-1 stimulates proteoglycan synthesis, while delaying its catabolism (Ziltener et al., 2012; Chevalier, 2010).

Additionally, regarding the intra-articular use of hyaluronic acid, the results of this study differ from those of some articles that described a modulating action of the inflammatory process. Hyaluronic acid has the ability to lubricate the joint, in addition to the function of nourishing the articular cartilage; and has been proven to have beneficial therapeutic characteristics for the joint environment; but its efficiency may be compromised by the local inflammatory process (Coronado et al., 2015). In a study that evaluated the effects of hyaluronic acid on clinical signs and synovial fluid, compared with a control group that received saline solution, it was demonstrated that intra-articular application of hyaluronic acid in healthy joints of horses is able to promote a response of mild to moderate inflammation, often associated with lameness and joint effusion (Yasui et al., 1992; Wang et al., 2006; Onodera et al., 2015; Johnston et al., 2019), as demonstrated in the present study. Furthermore, when comparing the cell count in synovial fluid, it was found in this study that treatment with hyaluronic acid had a higher total leukocyte count compared to the other experimental groups, suggesting a pro-inflammatory property in the joint environment of horses.

Although OCD does not often promote lameness in horses, synovial effusion may be present, which is the clinical feature that most bothers owners and trainers (Van Weeren, 2006). In the present study, it was demonstrated that the use of intra-articular PRP in the early post-operative period promoted lameness on the flexion test and that HA promoted an increase in synovial effusion when compared to the control group, which received intra-articular lactated Ringer's solution. Thus, this study contrasts with the literature that reports positive results with the intra-articular use of PRP (Carmona et al., 2007, 2009) and hyaluronic acid (Neuenschwander et al., 2019) in horses, but it agrees with the studies that demonstrated a mild to moderate inflammatory response after the administration of PRP (Textor & Tablin, 2013) and hyaluronic acid (Johnston et al., 2019) in healthy joints of horses.

However, some limitations should be noted in this study. First, the small number of horses used in this clinical study makes it difficult to interpret the results as unique or really representative for all horses.

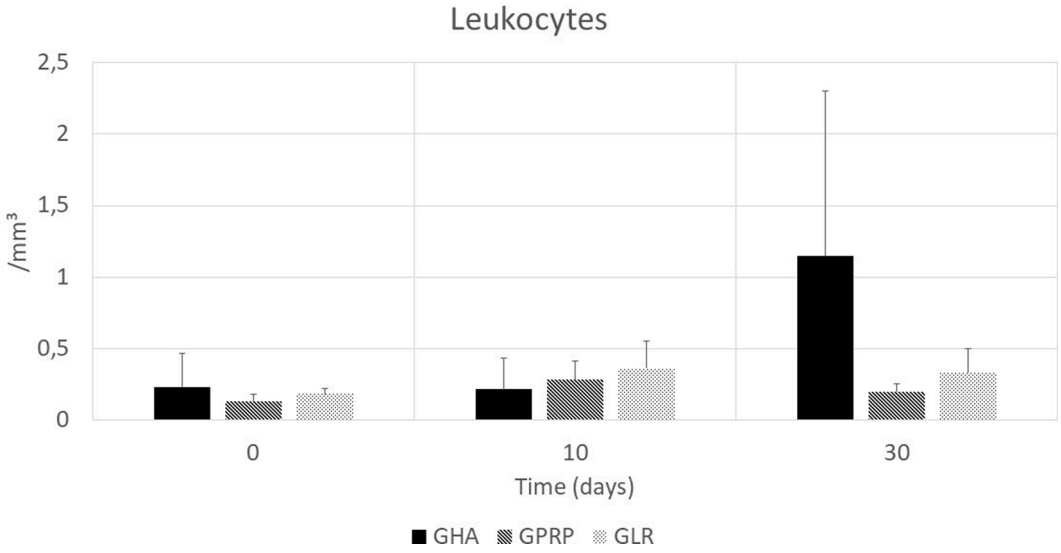

Fig. 3. Means and standard deviations of the total leukocyte count (leucocytes/mm3) of the synovial fluid of horses with osteochondrosis dissecans treated with lactated Ringer's solution (GLR), hyaluronic acid (GHA), and platelet-rich plasma (GPRP). The evaluations were performed on Day 0 (D0), on Day 10 (D10) before administration of the treatments and on Day 30 (D30).

Second, we did not measure inflammatory cytokines in this study aiming to evaluate local inflammation.

Thus, it is concluded that the intra-articular use of PRP or HA is not indicated as complementary therapy after the arthroscopy procedure for the treatment of OCD in horses.

Financial support

The authors thank São Paulo Research Foundation (FAPESP), Grant 2021/13378-8, and National Council for Scientific and Technological Development (CNPQ), Grant 309701/2022-8 for financial support, and CAPES code 01.

Ethical animal research

All procedures were approved by the Ethics Committee on Animal Use of the Faculty of Food Engineering and Animal Science, University of São Paulo, approval number 1310211116.

CRediT authorship contribution statement

Marcos F. Pereira: Resources, Supervision, Data curation, Writing – original draft, Writing – review & editing. **Gesiane Ribeiro:** Conceptualization, Resources, Data curation, Formal analysis, Investigation, Methodology, Validation, Writing – original draft, Writing – review & editing. **Alessandra Gonzales:** Investigation, Resources, Methodology, Writing – original draft, Writing – review & editing. **Julia A. Arantes:** Resources, Writing – original draft, Writing – review & editing. **Renata G.S. Dória:** Resources, Data curation, Formal analysis, Writing – original draft, Writing – review & editing.

Declaration of Competing Interest

No competing interests have been declared.

Acknowledgments

None.

References

Barcelos, K. M. C. (2012). Exame ultrassonográfico da articulaçãõ metacarpofalangeana de equinos praticantes de polo na zona Oeste do Estado do Rio de Janeiro: Protocolo e mensuraçõẽs. *Veterinaria e Zootecnia, 19*(3), 407–415.

Brossi, P. M., Moreira, J. J., Machado, T. S. L., et al. (2015). Platelet-rich plasma in orthopedic therapy: A comparative systematic review of clinical and experimental data in equine and human musculoskeletal lesions. *BMC Veterinary Research, 11*(98), 1–17.

Carmona, J. U., Arguelles, D., Climent, F., et al. (2007). Autologous platelet concentrates as a treatment of horses with osteoarthritis: A preliminary pilot clinical study. *Journal of Equine Veterinary Science, 27*(4), 167–170.

Carmona, J. U., López, C., & Prades, M. (2009). Uso de concentrados autólogos de plaquetas obtenidos mediante el método del tubo como tratamiento de artropatías en caballos. *Archivos de Medicina Veterinaria, 41*, 175–179.

Chevalier, X. (2010). Intraarticular treatments for osteoarthritis: New perspectives. *Current Drug Targets, 11*, 546–560.

Coronado, I., Iturriaga, V., Bornhardt, T., et al. (2015). Application protocol of hyaluronic acid on temporomandibular joint degenerative diseases. *Avances en Odontoestomatologia, 31*, 77–84.

Harrison, L. J., & Edwards, G. B. (1996). Radiographic investigation of osteochondrosis. *Equine Veterinary Education, 8*, 172–176.

Howard, R. D., & McIlwraith, C. W. (1993). Sodium hyaluronate in the treatment of equine joint disease. *Compendium: Continuing Education For Veterinarians, 15*, 473–481.

Johnston, G. C. A., Wood, K. A., Jackson, K. V., et al. (2019). Evaluation of the inflammatory response to two intra-articular hyaluronic acid formulations in normal equine joints. *Journal of Veterinary Pharmacology and Therapeutics, 00*, 1–12.

McIlwraith, C. W. (2005). Use of synovial fluid and serum biomarkers in equine bone and joint disease: A review. *Equine Veterinary Journal, 37*, 473–482.

McIlwraith, C. W. (2013). Surgical versus conservative management of osteocondrosis. *The Veterinary Journal, 197*, 19–28.

Moraes, A. P. L., Moreira, J. J., Brossi, P. M., et al. (2015). Short- and long-term effects of platelet-rich plasma upon healthy equine joints: Clinical and laboratory aspects. *Canadian Veterinary Journal, 56*, 831–838.

Neuenschwander, H. M., Moreira, J. J., Vendruscolo, C., et al. (2019). Hyaluronic acid has chondroprotective and joint-preserving effects on LPS-induced synovitis in horses. *Journal of Veterinary Science, 20*(6), E67.

Onodera, Y., Teramura, T., Takehara, T., et al. (2015). Hyaluronic acid regulates a key redox control factor Nrf2 via phosphorylation of Akt in bovine articular chondrocytes. *FEBS Open Bio, 5*, 476–484.

Raes, E. V., Vanderperren, K., Pille, F., et al. (2010). Ultrasonographic findings in 100 horses with tarsal region disorders. *The Veterinary Journal, 186*(2), 201–209.

Semevolos, A. S. (2017). Osteochondritis dissecans development. *Veterinary Clinics: Equine, 33*, 367–378.

Stashak, T. S (2002). *Adams'Lameness in horses* (5th ed.). Hoboken: Wiley-Blackwell.

Textor, J. A., & Tablin, F. (2013). Intra-articular use of a platelet-rich product in normal horses: Clinical signs and cytologic responses. *Veterinary Surgery,, 42*(5), 499–510.

Trotter, G. W., & McIlwraith, C. W. (1996). Clinical features and diagnosis of joint disease. McIlwraith, CW, Trotter GW. *Joint disease in the horse* (pp. 120–145). Philadelphia: W.B. Saunders.

Van Weeren, P. R (2006). Etiology, diagnosis and treatment of OC(D). *Clinical Techniques in Equine Practice, 5*, 248–258.

Wang, C. T., Lin, Y. T., Chiang, B. L., et al. (2006). High molecular weight hyaluronic acid down-regulates the gene expression of osteoarthritis-associated cytokines and enzymes in fibroblast-like synoviocytes from patients with early osteoarthritis. *Osteoarthritis Cartilage, 14*, 1237–1247.

Yasui, T., Akatsuka, M., Tobetto, K., et al. (1992). The effect of hyaluronan on interleukin-1 alpha-induced prostaglandin E2 production in human osteoarthritic synovial cells. *Agents and Actions, 37*, 155–156.

Ziltener, J. L., Allet, L., Sclison, P., et al. (2012). How effective are injections of platelet-rich plasma (PRP) for the treatment of sports injuries: A critical review of the literature. *Journal of Sports Medicine & Doping Studies S2*, 1–6.