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The Clinical Effects of Platelet-Rich Plasma Therapy In The Treatment of Stifle Osteoartritis In Dogs

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Abstract:
Osteoarthritis (OA) in dogs is a progressive joint disease that causes chronic pain, impaired joint function and reduced quality of life. The limitations of current treatment options have increased interest in biological therapies. Platelet-rich plasma (PRP) is considered a promising therapeutic option for OA because of its ability to modulate inflammation and support cartilage tissue through the growth factors and bioactive molecules it contains. This review evaluates the preparation and administration methods, mechanisms of action, clinical and biochemical effects, and reported complications of PRP in the treatment of stifle OA in dogs. The available literature indicates that PRP treatment can modulate MMP-2 and MMP-9 activities, reduce pain and lameness, and improve weight-bearing and gait symmetry. However, variations in PRP preparation and administration protocols have prevented a definitive consensus regarding the optimal dose and treatment frequency. In conclusion, PRP may be considered a complementary or alternative biological approach for the treatment of stifle OA in dogs. Nevertheless, standardized controlled trials are needed to establish its efficacy and long-term outcomes.

Key Word*: osteoarthritis, platelet-rich plasma, cartilage tissue, stifle, dogs .*

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I. Introduction

Osteoarthritis (OA) is a common degenerative joint disease that significantly affects quality of life in both humans and animals. Age, obesity, joint trauma and genetic predisposition are among the main risk factors in the development of OA[1]. OA is characterised by progressive degeneration of the articular cartilage, joint inflammation and chronic pain, all of which contribute to impaired mobility[2]. This condition affects more than 20% of the canine population, with the stifle and elbow joints being among the most commonly affected sites and significantly reduce the quality of life of affected dogs [3,4,5]. Canine stifle OA commonly develops secondary to conditions such as cranial cruciate ligament disease or mechanical instability, and may result in progressive degenerative changes within the joint over time [3,5,6]. The primary goals of OA treatment in dogs are to reduce pain, preserve joint function and maintain quality of life. Current management strategies involve the integration of pharmacological treatments with multimodal interventions such as weight management, exercise and rehabilitation. However, given the progressive and chronic nature of OA, research has increasingly focused not only on controlling clinical symptoms but also on biological therapies targeting the underlying pathological processes within joint tissues. Non-steroidal anti-inflammatory drugs (NSAIDs) are commonly used as a firstline pharmacological treatment to reduce inflammation and pain and alleviate joint discomfort. However, longterm use of NSAIDs may result in adverse effects, including kidney damage, gastrointestinal ulcers and hepatic dysfunction [7,8]. Corticosteroids, which possess potent antiinflammatory and immunosuppressive properties, are also widely used to management of severe arthritis in cats and dogs. Nevertheless, prolonged corticosteroid use has been associated with adverse effects such as immunosuppression, hormonal disturbances and osteoporosis. Furthermore, these treatment approaches do not directly target the fundamental pathological changes observed in arthritis, such as cartilage damage and degeneration[2]. Although opioid analgesics may be used for pain management, their use carries a risk of dependence. In addition to pharmacological treatments, physiotherapy and surgical interventions are also used in the management of OA. While physiotherapy can contribute to improved joint function, it cannot halt disease progression; surgical interventions, on the other hand, are generally used as a last resort due to their high costs and potential risks [7]. These limitations highlight the need for novel therapeutic approaches with few or no adverse effects that, in addition to providing antiinflammatory and analgesic effects, support cartilage structure and may help counteract cartilage degradation[9]. Among these alternatives, platelet-rich plasma (PRP) is one of the alternative treatment options

being investigated for the treatment of OA due to its biological properties, which can promote cartilage regeneration and modulate intra-articular inflammation [3,5,10,11,12,13].

The aim of this review is to evaluate the preparation and administration methods, biological properties and mechanisms of action, clinical and biochemical effects, and reported complications of PRP therapy in the treatment of stifle OA in dogs, based on the current literature.

II. Protocols for the preparation and administration of platelet-rich plasma

The preparation and intra-articular administration of PRP for the treatment of stifle OA in dogs are based on the premise that autologous growth factors, when delivered into the joint, promote cartilage anabolism and modulate inflammation. The clinical efficacy of PRP treatment is linked to factors such as appropriate blood collection, the achievement of a high platelet concentration through a meticulous centrifugation protocol, and the thorough application of aseptic injection techniques [3,4,13,14,15,16].

The first step in PRP preparation is the collection of a venous blood sample from the patient under sterile conditions. The jugular vein is generally preferred. For manual double-centrifugation protocols, 16–20 mL of blood may be collected from dogs, whereas certain specialised kits or filtration systems may require 30–55 mL or even up to 120 mL [3,4,14,15,17]. Sterile tubes containing sodium citrate (Na-citrate) or Acid Citrate Dextrose-A (ACD-A) are used as anticoagulants to prevent blood coagulation [3,4,14,15].

According to the standard manual double-centrifugation protocol, the first centrifugation is performed at 96 g for 10 minutes to separate the cellular components from the sodium citrate-anticoagulated blood. The resulting plasma layer is carefully transferred to another tube and centrifuged a second time at 380 g for 20 minutes. The platelet concentrate obtained through this process is filtered and prepared for injection[14].

According to the buffy coat-focused double centrifugation protocol, citrated blood tubes are centrifuged at 2800 rpm for 20 minutes. This process separates the blood into three main layers: red blood cells, the buffy coat and platelet-poor plasma (PPP). Eighty per cent of the plasma at the top is aspirated and discarded. The buffy coat layer, containing platelets and mononuclear cells, is carefully resuspended in the remaining 0.75–1.0 mL of plasma using a sterile pipette. The resuspended mixture is centrifuged at 1300 rpm for 15 minutes in a sterile Falcon tube. Following centrifugation, the supernatant (PPP) is removed and the platelet pellet at the bottom of the tube is resuspended in the remaining small volume of plasma using a vortex mixer to obtain the final PRP[3].

According to the modern laboratory double-spin protocol, anticoagulated blood is centrifuged at 900 x g for 5 minutes to separate the plasma from the cells. The collected plasma is centrifuged a second time at 1500 x g for 15 minutes. Approximately two-thirds of the supernatant PPP fraction is discarded. The lower fraction, containing the platelet pellet, is carefully collected while avoiding erythrocyte contamination, and subsequently homogenised. This method yields a platelet concentration 3.81 times higher than that found in whole blood[16].

According to the pure PRP protocol, leukocyte-rich PRP obtained from commercial kits is subjected to an additional centrifugation step, spinning at 1250 g for 3 minutes. During this step, red and white blood cells settle at the bottom of the tube, while the platelets remain in the supernatant, resulting in the production of pure, leukocyte-free PRP[5].

According to the protocol for PRP preparation using a filtration device, whole blood containing an anticoagulant is passed through the C-PET filtration system by gravity. The platelets retained on the filter surface are washed (flushed) with a hypertonic (2%) NaCl solution and recovered directly into a syringe[4].

PRP, prepared according to various protocols, is administered under sedation or general anaesthesia during intra-articular injection to minimise the risk of injury to intra-articular structures and ensure complete immobilisation of the dog [3,4,14,15]. The area surrounding the stifle joint is extensively shaved and disinfected with an antiseptic solution before injection [6,14]. A sterile needle, usually 23 G, is inserted into the stifle joint cavity. Before the injection, a small amount of synovial fluid is aspirated from the joint, and the volume aspirated is recorded. While maintaining the needle position, the syringe containing synovial fluid is removed and replaced with a syringe containing PRP [4,15]. The PRP is then slowly injected into the joint. Depending on the breed of dog, the volume of intra-articular injection generally ranges from 2 to 5 mL. During the procedure, the injection is continued until a distinct resistance is felt against the needle plunger [4,5,14,15].

During the PRP treatment period, dogs should not receive NSAIDs, corticosteroids or joint supplements, as these may interfere with platelet activation, local inflammatory modulation and the release of growth factors [3,4]. Many protocols recommend discontinuing NSAIDs at least one week before treatment and avoiding their use throughout the treatment period[4]. Another important consideration is that restriction of excessive and uncontrolled physical activity following injection. However, to facilitate the homogeneous distribution of PRP within the joint and to maintain the joint range of motion, patients are recommended to undergo a controlled, leashed walk at a gentle pace for 10–15 minutes daily[15].

III. The biological structure and mechanisms of action of platelet-rich plasma

Platelet-rich plasma is an autologous biomaterial containing a high concentration of platelets relative to whole blood, obtained from the patient's own blood using a double-centrifugation method [3,17,18]. In addition to their role in haemostasis, platelets are a rich source of growth factors and cytokines due to their abundant alpha granules [3,16,19,20]. When administered intra-articularly or activated locally, platelets release several key growth factors, including platelet-derived growth factor, transforming growth factor-beta, insulin-like growth factor-1, vascular endothelial growth factor, basic fibroblast growth factor, and epidermal growth factor [3,14,15,17]. These growth factors and bioactive proteins contribute to processes such as the proliferation, migration of mesenchymal stem cells and chondrocytes and synthesis of cartilage matrix within the joint [3,15,17]. Furthermore, they may contribute to an analgesic effect by regulating local inflammatory signals [3,6]. The autologous nature of PRP may provide a safety advantage by reducing the risk of immunological reactions associated with allogeneic biomaterials [3].

Biochemical effects and Matrix Metalloproteinase Inhibition

In stifle OA, pro-inflammatory cytokines produced by intra-articular macrophages and chondrocytes stimulate proteolytic enzymes that lead to the degradation of the cartilage extracellular matrix. The most important of these enzymes are matrix metalloproteinases (MMPs) zinc-dependent endopeptidases that break down cartilage collagen and other structural proteins and, in particular, MMP-2 and MMP-9, which are known as gelatinases. Zymography and ELISA studies have reported that intra-articular PRP injections can exert direct inhibitory effects on the biochemical mechanisms of cartilage-degrading enzymes. In a study by Arıcan et al., it was reported that following PRP treatment administered to the stifle joints of dogs, there was suppression of MMP-9 activity and a marked reduction in MMP-2 activity in the initial phase. Although MMP-2 activity increased again in the following days, clinical improvement was maintained[14]. These findings indicate that PRP may modulate destructive processes within the joint and supports its potential chondroprotective effect at the biochemical level [3,14].

IV. The Clinical Effects of Platelet-Rich Plasma

The clinical effects of PRP in canine stifle osteoarthritis are assessed using objective parameters such as force platform and pressure-sensitive gait analyses, as well as subjective pain scores. In clinical studies, the Canine Brief Pain Inventory and various clinical assessment scales, completed by dog owners and veterinarians, have been used [3,4,14].

In a study conducted by Aminkov et al., intra-articular PRP was administered three times at 10-day intervals to dogs with knee and elbow osteoarthritis, and a significant reduction in lameness scores and pain scores was reported. It was noted that this clinical improvement persisted for 6 to 8 months [3].

It has been found that a single dose of intra-articular PRP also significantly improves pain intensity and quality of life, particularly in cases of early- and mid-stage OA [4,14].

Force plate analyses show that PRP improves patients' ability to bear weight on their knee joints [4,5]. In a randomised controlled clinical trial conducted by Fahie et al., it was reported that 12 weeks after a single dose of intra-articular PRP injection, a statistically significant 12 per cent increase in maximum vertical force values was measured in the affected limbs of treated dogs, while no clinical improvement was observed in the control group [4].

In a study by Venator et al., it was reported that in dogs with chronic stifle OA lasting for more than 12 months due to chronic cranial cruciate ligament disease, a single-dose PRP injection significantly improved symmetry in the vertical force distribution of the hind limbs for at least 4 to 12 weeks and contributed to the balancing of weight distribution [5].

In the literature, HA-PRP conjugates which combine hyaluronic acid (HA) with PRP have also been evaluated in a stifle OA model. In a study by Lee et al., gross and histopathological examinations revealed that HA-PRP treatment, administered every two weeks over a 20-week follow-up period, provided a statistically superior long-term chondroprotective effect compared to standard HA treatment administered weekly, in terms of preserving cartilage surface regularity, preventing proteoglycan loss and reducing chondrocyte clustering [15].
A comparison of single-dose and repeated PRP protocols in the treatment of stifle OA in dogs is presented in Table 1.

Table 1. Comparison of single-dose and repeated PRP protocols in the treatment of knee osteoarthritis in dogs.

Comparison Criterion	Single-Dose PRP Protocol	Repeated PRP Protocol
Administration Protocol	Single intra-articular injection [4,5,6,14].	A series of intra-articular injections administered at defined intervals [3,15].
Duration of Effect	Clinical improvement is generally observed for up to approximately 3 months [4,5,6].	The duration of symptomatic and clinical improvement may extend to 5-8 months [3,15].

Indication	More effective in mild to moderate, early-stage OA cases [6,14].	Preferred for chronic, advanced stage OA or OA associated with recurrent joint instability [3,5].
Clinical Improvement	Reduction in pain, with limited to moderate improvement in limb use and gait symmetry over a limited duration [5,6].	Marked improvement in pain and functional scores [3].

V. Complications

It has been reported that serious complications are rare following intra-articular PRP injections for stifle OA [4,15,17]. As PRP is an autologous biomaterial, the risk of immunological reactions associated with allogeneic materials is low; however, it is not possible to completely rule out procedure-related complications, such as infection, following intra-articular injections. The most common minor side effects are mild joint stiffness, a temporary increase in lameness, or local sterile synovitis reactions, which resolve spontaneously within the first few days following treatment and do not require any further intervention [3,15,17].

VI. Conclusion

In light of clinical and biochemical evidence, PRP is regarded as a biological agent that can modulate the activity of proteolytic enzymes such as MMP-2 and MMP-9, reduce pain both subjectively and objectively, and correct weight-bearing symmetry in the treatment of stifle OA in dogs [3,4,5,14,15]. In particular, for patients unsuitable for surgical intervention, where the use of NSAIDs is contraindicated, or in cases where adequate pain control cannot be achieved with conventional methods, PRP offers a rational treatment alternative [5]. However, due to the heterogeneity of existing studies in terms of sample sizes, PRP preparation methods, administration protocols and evaluation methods, it is not possible to reach a definitive conclusion regarding the long-term chondroprotective effect of PRP and the optimal administration protocol. Therefore, PRP may be considered as a biological approach in the treatment of stifle OA in dogs, particularly as an alternative or adjunt to conservative treatment options; however, there is a need for more robust and standardised controlled trials regarding its clinical use.

Declaration

Ethics Committee Approval

This article does not require ethics committee approval.

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