Trends in Biotechnology

CellPress

Review

Platelet-Derived Products in Veterinary Medicine: A New Trend or an Effective Therapy?

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Platelet-derived products (PDPs) have gained popularity, mainly due to their high concentrations of bioactive molecules such as growth factors and cytokines, which play important roles in tissue healing and regeneration. PDPs are obtained through minimally invasive procedures and their therapeutic effect has been widely recognized. In veterinary medicine, however, the lack of standard protocols to generate PDPs is a major hurdle for assessing the clinical relevance of PDP-based therapies and for their widespread usage. The aim of this review is to analyze the technical and scientific specificities of PDPs in terms of preparation methodologies, classification categorization, nomenclature, and biological properties to advance their future biotechnological potential in veterinary contexts.

Highlights

Platelet-derived products (PDPs) are envisioned as economic, safe, fast, and easily obtained formulations in both tissue engineering and regenerative therapeutics.

PDPs are sources of growth factors, cytokines, and structural proteins involved in tissue healing, obtained from minimally invasive and simple blood harvesting.

Consensus and recommended guidelines are needed to promote well-characterized veterinary PDPs, starting with standardizing nomenclature, classification, and preparation methods, in both clinical and research scenarios.

Quality and safety criteria should be defined for PDP manufacture, envisioning explicit scientific research in veterinary patients and contributing to definitive evidence for the efficacy of platelet therapy.

Platelet lysate displays strong promise in becoming a bio-fabricated allogeneic therapy.

Introducing Platelet-Derived Products: Starting from Blood Harvesting

Platelet-derived products (PDPs) obtained by separating whole blood have been explored as **autologous** (see Glossary) or **allogeneic** sources of growth factors (GFs), cytokines, and structural proteins, with healing and regenerative potential for both tissue engineering (TE) approaches and regenerative strategies [1,2].

The different hemoderivatives applied in human and veterinary settings can be roughly classified into platelet-poor and platelet-rich products, distinguished mainly by the presence of platelets [3]. Depending on the platelet and fibrin content, blood-derived biomaterials include platelet concentrate (PC), which is sometimes referred to as platelet-rich plasma (PRP), platelet-poor plasma (PPP), fibrin glue (FG), and platelet-rich fibrin (PRF), among others [2–5]. FG was the first PDP used in humans (since the 1960s), followed by PRP, which has recently spread to veterinary medicine [3,6,7]. All of these fractions can be derived from simple blood harvesting (Figure 1).

The preparation of PRP or PC usually involves two sequential centrifugations of anticoagulated whole blood (Figure 1A), designated respectively as soft spin (to separate erythrocytes from plasma) and hard spin (to concentrate the platelets) [8].

The first centrifugation separates the blood into three phases (Figure 1B): a lower layer rich in red blood cells (RBCs); a middle layer rich in white blood cells (WBCs) and platelets, also known as the buffy coat (BC); and an upper layer, corresponding to the plasma, which is a hemoderivative used for FG production that still contains some residual platelets in suspension [9].

The second centrifugation of the plasma (Figure 1C), which may or may not contain the BC depending on the applied methodology (Box 1), produces two fractions: the PRP/PC (Figure 1D) and the PPP fractions (Figure 1E) [3,10].

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The PRP/PC can be used directly, or it can be activated to obtain other PDPs such as platelet gel (Figure 1D,ii) and plasma-rich in growth factors (PRGF; Figure D,i) or to disrupt the platelets' membranes to obtain acellular platelet lysate (PL) (Figure 1D,iii) [11,12].

After the PDPs are activated, the platelet cytoskeleton reorganizes and platelet granules are moved to the cell center, fusing with the **open canalicular system** and releasing GFs and cytokines into the extracellular environment [13]. The PPP, poor in platelets but with a physiological level of fibrinogen, can be supplemented with calcium salts or exogenous **thrombin** to activate fibrinogenesis, producing fibrin membrane (FM) (Figure 1E,i) [14]. The spontaneous clotting of whole blood during or before its centrifugation produces the PRF and autologous serum (AS) (Figure 1F,i,ii) [3,15].

This review results from the increasing demand for platelet-derived biomaterials in veterinary medicine, focusing on clinical veterinary approaches to platelet therapy. This article is centered on the technical aspects of platelet-based therapy in the veterinary field and it discusses the lack of standard protocols to produce different PDPs and the lack of categorization of these products.

Platelets in Tissue Healing: Much More Than Coagulation

Beyond the major role of platelets in hemostasis and thrombogenesis, they play a pivotal action in wound healing [13], being involved in the control of the inflammation cascade, recruiting WBCs and progenitor cells, and motivating angiogenesis [13]. Also known as thrombocytes, they are generated in bone marrow, being formed from small cytoplasmic fragments of the megakaryocytes, regulated by thrombopoietin and other cytokines [16].

Despite being nonnucleated, platelets have intracellular compartments termed 'granules' and lysosomes, very relevant for their function. Two granule types can be identified: dense and alpha granules. The first ones are lysosome-related organelles and contain small molecules such as calcium, magnesium, serotonin, histamine, and purines such as ADP and ATP [17]. Alpha granules are the most abundant organelles, containing multiple GFs, mitogens, and pro- and anti-inflammatory cytokines [13].

Amongst the GFs stored in alpha granules are **vascular endothelial growth factor (VEGF)**, **platelet-derived growth factors (PDGF)**, **fibroblast growth factor (FGF)**, **epidermal growth factor (EGF)**, **brain-derived neurotrophic factor (BDNF)**, **hepatocyte growth factor (HGF)**, and **insulin-like growth factor-1 (IGF)** [4,17–19], which are released upon activation. These polypeptides are essential for intercellular communication, through the mediation of cell metabolism, relocation, proliferation, differentiation, and apoptosis, triggering vasculature remodeling [20]. Additionally, nano-sized platelet-extracellular vesicles (P-EVs) such as exosomes and microvesicles have been recently studied, mainly due to their RNA and protein cargos [21]. These display proangiogenic, antiapoptotic, and anti-inflammatory properties, regulating signaling pathways via receptor–ligand interactions or content delivery [22]. The latest research shows that P-EVs have similar results in the epithelization of cutaneous wounds to those of PRP/PC [23].

Presenting the Hemoderivatives with Therapeutic Interest

The Suitability of Platelet-Poor Hemoderivatives

Platelet-enriched products are not the only blood-derivatives with clinical and biotechnological significance. Platelet-poor hemoderivatives have emerged as surplus products raised from the manufacture of platelet-enriched fractions, constituting a target of reinvestment for the regenerative field, namely in human medicine. Among platelet-poor products already used in clinics are the PPP, FG, FM, and AS. According to the authors' knowledge, FM has not been used in veterinary patients.

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PPP is a plasmatic solution depleted of platelets and other blood elements, being rich in fibrinogen [24]. Its use has been considered advantageous as a sealant for hemostasis, being inexpensive and readily available [3,25]. The presence of angiogenic factors and biomolecules in PPP has been associated with bone repair in human and canine studies [3,26].

FG, also assessed as fibrin sealant, is another blood-derived biomaterial, described as a semi-rigid to elastic clot, acting as a biological tissue glue through which cells can migrate [9]. Commercial FG is produced from fibrinogen precipitate and it can be prepared using several techniques: the most common one is through the thawing of fresh frozen plasma at 2–4°C, leading to the isolation of cold-insoluble proteins, which are later separated from the remaining plasma fraction by centrifugation (cryoprecipitate) [27]. It has been used for human and veterinary surgical purposes, in ophthalmology, orthopedics, and cardiovascular and reconstructive surgery [27].

Repair of human corneal ulcers has been recently reported using autologous FM, produced by the jellification of PPP fraction with calcium salts, combined with solid PRP/PC [14,28].

AS is frequently prescribed for veterinary patients diagnosed with **keratomalacia**, as a corneal wound promoter [29]. AS attainment implies blood coagulation before its centrifugation (Figure 1F,i), being used because of its biochemical composition, analogous to normal lacrima. Besides the lubrication of the ocular surface, AS still contains GFs, antiproteinase, and anticollagenase properties, supporting epithelial healing and inhibiting stromal loss [29].

PRP, PC, and PG: Clarifying the Nomenclature

PRP/PC is described as a liquid platelet concentrated fraction before activation [30]. After activation, PRP/PC acquires a solid viscoelastic fibrin network, called platelet gel (PG), acting as a temporal **scaffold** for the proteins of the cells. PG has been explored for corneal healing and bone and soft tissue regeneration [12,31].

The activation method may influence GF release and contribute to the physical properties of the PG, ranging from liquid to solid gel. Several agents may be used for PDP activation, such as thrombin, calcium salts (e.g., $CaCl_2$) [3,32], **batroxobin** [33], or collagen [34]. Thrombin and $CaCl_2$/thrombin were proven to cause a faster human PC jellification (15 minutes), whereas $CaCl_2$ promoted clot formation within 30 minutes, with a progressive GF discharge up to 24 hours [35].

The definition of PRP and PC is still controversial. While some researchers consider PRP to be any platelet suspension in plasma (such as the upper phase obtained from whole blood after the soft spin), others attribute this definition to any platelet-enriched fraction, with a concentration capable of producing therapeutic effects [30]. Following this line, PRF, PG and liquid PRP/PC formulations (henceforth designated PC) are all categorized as 'PCs', making any distinction between formulations impossible.

The lack of consensus in this terminology and indefiniteness of these concepts, associated with the limited disclosure of their composition (in terms of platelet, RBC, and WBC content), has led to confusion in the interpretation of scientific outcomes concerning the real therapeutic effect of such platelet-rich hemoderivatives [34]. The terminology PRP has been assumed to designate all platelet preparations, prepared by diverse protocols, presenting different compositions, and, therefore, reaching different biological outcomes [34].

In 2012, a proposition of human PRP definition and preparation methodology, settled in an accurate and simple terminology system, was made [31]. According to this, all the products defined as

Glossary

Allogeneic: transfer of cells, tissues, or organs between genetically different subjects, within the same species, but sufficiently unlike to interact antigenically.
Autologous: transfer of cells, tissues, or organs between one donor and a recipient site, in the same individual.
Batroxobin: a thrombin-like serine protease, produced from snake venom, with fibrinogenolytic activity over the platelet-rich plasma, promoting the formation of clots, without affecting the integrity of the platelets.
Brain-derived neurotrophic factor (BDNF): mainly occurs inside platelets and only a minor amount circulates in a free plasmatic form, demonstrating higher concentration in serum than plasma samples. It is widely distributed in the central nervous system.
Epidermal growth factor (EGF): a protein that stimulates mitogenesis, proliferation, and differentiation.
Fetal bovine serum (FBS): originating from blood drawn from a bovine fetus, under a closed system collection, at the slaughterhouse. Is widely used as *in vitro* cell culture supplement.
Fibroblast growth factor (FGF): involved in metabolic functions, tissue repair, angiogenesis, and regeneration.
Hepatocyte growth factor (HGF): a protein secreted by stromal cells with mitogen capacity over hepatocytes.
Heterologous: transfer of cells, tissues, or organs between different species, from one species to another.
Insulin-like growth factor (IGF-1): insulin-like growth factor-1, a growth hormone, with neuroprotective and neuroproliferative effects, also regulating apoptosis in multiple organ systems.
Keratomalacia: an eye pathological disorder, where a mucosa atrophic alteration occurs and corneal stroma acquires a 'melting' aspect, associated with stromal loss. It results from an enzymatic breakdown of corneal collagen by collagenases, due to bacterial and even from WBC activity.
Open canalicular system: a system present throughout the platelet cell, allowing the entry of external elements into the platelets as well as the release of its granule contents to the exterior environment.
Platelet-derived growth factor (PDGF): a potent activator for cells of mesenchymal origin. PDGF has significant proangiogenic effect in tissue repair.

PRP-related are, in fact, 'PCs', independent of activation state and leukocyte content, in a manner that they are concentrated suspensions of thrombocytes. Other authors extended this designation, proposing the PAW system, in which 'P' refers to the absolute platelet number, 'A' the activation method, and 'W' the presence/ absence of WBCs [36]. Curiously, the designation PC obtained by plateletpheresis has been unanimously used in hemotherapy descriptions. More recently, different biotechnological companies have developed commercial kits for the production of standardized platelet-enriched suspensions [37]. In the veterinary context, ready to use kits are being introduced for horses and dogs, composed of collection tubes that, in specific centrifugation conditions, are expected to yield a specific platelet concentration, suspended in plasma. Nevertheless, the variations between kits in terms of the required volume of blood, anticoagulant type and quantity, together with the actual fairly variable final cellular and GF composition, overshadow the therapeutic predictability of these kits [6,38].

Pseudothrombocytopenia: false low platelet counts, occurring in *in vitro* blood sampling, generally associated with anticoagulant platelet aggregation.
Scaffolds: in tissue engineering, a structural template produced from a specific biomaterial aiming at providing the needed stability and architecture for new tissue formation.
Sonication: methodology in which high frequency and energy sound waves are used.
Thrombin: formed when the zymogen prothrombin is cleaved by a prothrombinase complex that forms in the presence of calcium from platelet phospholipids. Among other molecules, it converts fibrinogen to fibrin and induces platelet activation and aggregation.
Vascular endothelial growth factor (VEGF): a highly specific mitogen for vascular endothelial cells.

The Versatile PRF

PRF was firstly advanced in 2001 for human oral and maxillofacial surgery, to enhance bone regeneration [39]. During a single centrifugation process of nonanticoagulated whole blood, platelets become activated by the endogenous coagulation cascade. The autologous thrombin contributes to the direct polymerization of fibrinogen into fibrin, resulting in a 3D flexible fibrin network, in which platelets and WBC are entrapped [40]. The result is a PRF clot constituted of three layers: the top nonclotted layer is the acellular serum constituted by circulating plasmatic molecules; the clot, in the bottom of the tube, comprises a translucent upper layer and a lower red (RBC-rich) layer. In humans, PRF has been used as a membrane, reducing patient discomfort during the wound-healing period [41]. PRF membranes can release a high quantity of GFs for a prolonged period, for up to 2 weeks, acting as a space filler due to its scaffold-like function and GF temporal release [2,42].

PRF is probably the most versatile and accessible platelet-enriched product available for veterinary regenerative treatments that can be used in several clinical applications. It is a naturally biocompatible healing material, accelerating wound closure/soft skin healing. It can be used as a bone graft material, or as a biological graft for synthetic osteogenic materials (e.g., hydroxyapatite, bone grafts, etc.). Additionally, this natural scaffold is easily prepared in the veterinary clinic, having minimal contamination risks when prepared with aseptic technique, due to its minimal manipulation. PRFs have been also advocated to have bactericidal proprieties [43].

The Newcomer PL

PL is, by definition, an acellular plasmatic solution obtained by the disruption of the platelets produced from PC [44,45]. During PL production platelets are lysed, releasing the GFs, cytokines, and other related proteins, in an easily standardized protocol, contributing to the lower batch-to-batch variation [46].

The most common protocols for obtaining PL are based on the cryogenic disruption of the platelets in pooled PC batches by several freezing and thawing cycles and subsequent removal of cellular debris. Other physical treatments, such as **sonication**, have also been proposed for the disruption of platelets [44].

The human PL was primarily proposed as a prevailing alternative to **fetal bovine serum (FBS)**, avoiding the inherent animal suffering during FBS production, constituting a supplement for xenogenic-free cell culture, and reducing the occurrence of graft-versus-host rejection and zoonotic infections risk, once no antibodies are produced against PL [2,18]. Nowadays, equine, canine, and feline PL is encouraged for the corresponding cell culture production.

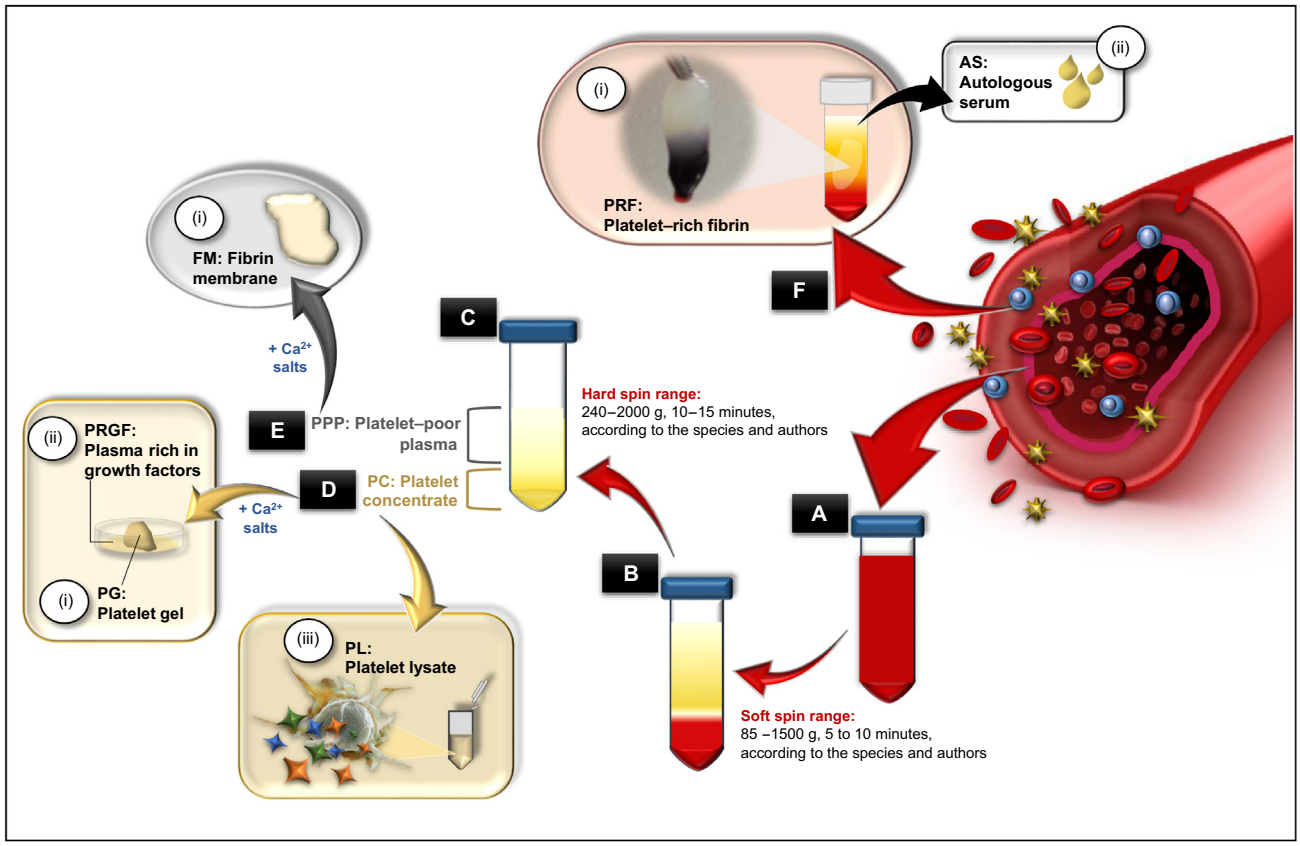

Figure 1. Flow Chart of Platelet-Derived Product Production from Peripheral Whole Blood. After the (A) blood collection with anticoagulant, the (B) first centrifugation is followed by (C) a second centrifugation, producing: (D) platelet concentrate (PC), also cited as platelet-rich plasma (PRP) and (E) platelet-poor plasma (PPP). The induction of the fibrinogenesis in PPP by addition of Ca^{2+} and/or thrombin produces fibrin membrane (E,i). When activated, PC will produce a platelet-rich clot, the platelet gel (D,ii), and solution rich in growth factors, the plasma-rich in growth factors (D,i). PC/PRP may be processed, inducing the platelet disruption and the release of their content, producing the acellular platelet lysate (D,iii). Blood collected without anticoagulant (F) enables the preparation of two hemoderivatives: when immediately submitted to a single centrifugation this allows the preparation of platelet-rich fibrin (PRF) (F,i); the serum resulting from the centrifugation of naturally coagulated blood produces the autologous serum (AS) (F,ii). Abbreviations: FM, fibrin membrane; PG, plasma gel; PL, platelet lysate; PRGF, plasma rich in growth factors.

PL has high potential to overcome the issues related to allogeneic administration of PDPs. Theoretically, all blood cell membranes would be depleted during PL preparation protocol, therefore minimizing the immunogenic reactions. Considering the inexistence of a complete blood typing in veterinary patients, during the use of single allogeneic donor platelet formulations, the recipient animal is only likely to be exposed to the formation of isoagglutinin immunoglobulins, a limitation theoretically absent in PL administration. However, pooled platelet concentrated units allow the production of larger PL batches and allow higher reproducibility when compared with individual lots [47,48]. Moreover, the allogeneic administration is advantageous over autologous methodology, in which the animal suffers from a medical condition for which blood collection is not advised, or simply are more difficult to obtain.

Recent studies have shown the great potential of PL, not only as a source of GFs to ameliorate the performance of biomaterials for TE applications but also as being processable into free-standing biomaterials [44]. Additionally, PL-based materials showed methicillin-resistant *Staphylococcus aureus* bacteriostatic activity, which may contribute to avoid infection of the wound site and subsequent TE graft failure [49].

Box 1. Liquid Platelet Concentrates: Understanding the Preparation Methodologies

Platelet-rich plasma (PRP) was the pioneer of PDPs, first introduced in 1987 by Ferrari and colleagues [100]. It was described as an autologous transfusion component for cardiac surgery patients, to avoid homologous blood transfusion. In humans, PRP applications are mostly used in dermatologic and maxillofacial surgery, but currently their use is broadly in orthopedic and sports medicine and cosmetic and plastic procedures [25]. Several studies have proven PRP therapeutic efficacy as an anti-inflammatory and prohealing natural agent, with unquestionable potential for tissue regeneration, even with analgesic proprieties [77,78].

According to the literature, PRP/PC can be produced mainly by three techniques: it can be acquired from a plasmatic platelet-rich fraction (termed PRP-methodology, contributing to the terminology confusion), used in North America; buffy coat (BC-method), used in Europe; and apheresis [10,44].

Considering the WBC influence in platelet-based formulations, the PC has been classified into pure-platelet-rich fraction if depleted of leukocytes (also documented as P-PRP) or leukocyte-platelet-rich fraction (L-PRP) [30].

With regard to preparation methodology, the BC method leads to young platelet recovery, but also higher WBC and RBC contamination [8,10], which is undesirable for allogeneic applications [6,8]. Instead, the platelet-rich method is associated with fewer leukocytes and erythrocyte contamination, with superior platelet counts [10]. The usage of membrane filters to separate WBC (leuko-reduction) has also been applied to produce purer human PCs [79].

The presence of WBC within PDPs remains unclear. *In vitro* works point to WBCs as being responsible for the release of proinflammatory cytokines [interleukin (IL)-1β, IL-6, IL-8 interferon (IFN)-γ, and tumor necrosis factor (TNF)-α] and metalloproteinases (MMP-3 and MMP-13) and their presence in PDPs is considered an inflammatory enhancer [80–82]. Other researchers have suggested that the presence of leukocyte-rich formulations does not produce a relevant upregulation in proinflammatory mediators in osteoarthritic knee studies [83,84].

Apheresis is a common procedure in human medicine for hemotherapy purposes. In the veterinary field, only a few institutions across the world have access to this technique, presenting considerable limitations for veterinary practice, namely: (i) considerable blood volume is needed from the patient, which is particularly hazardous for small animals; (ii) it leads to significant electrolyte unbalance, with hypoxia and cardiac alterations risk; (iii) procedure requires general anesthesia; and (iv) the collection needle of the equipment requires a considerable minimum caliber vein (usually jugular vein) [8,85].

What about PDPs in Veterinary Medicine?

Regenerative veterinary medicine is following human progress in a translational trend (Figure 2) [3,8,50,51]. Regenerative medicine, more than helping damaged tissues to heal, aims at returning their original or near-original condition, often used as a complementary strategy in multimodal approaches [52].

The longer life expectancy of pets has resulted in natural occurring age-related diseases, resembling those diagnosed in human patients. Additionally, the conscience shift on the role of companion animals in our society has contributed to the increased investment in regenerative therapies.

Veterinary treatments using PDPs are framed in musculoskeletal conditions, wound defects, and articular lesions, mainly in equine, canine, and feline patients (Table 1) [3,32,50,53,54]. In particular, cellular treatments delivering cells derived from bone marrow or adipose tissues seem to benefit from combination with PDPs, displaying both enhanced regenerative and anti-inflammatory outcomes [11,55–60]. Clinical trials involving stem cells such as adult mesenchymal stem cells (MSCs), GFs/platelet-components, and/or TE scaffolds in animals are continuously increasing, most of them in association with platelet-based therapies [1,54,58,61].

Clinical Applications in Horses

Clinical studies have been developed to study the combination of both platelet and cellular formulation in the recovery of musculoskeletal injuries of equine patients. However, the results are quite ambiguous (for further reading see [1]). While in some studies PC administration, as a sole

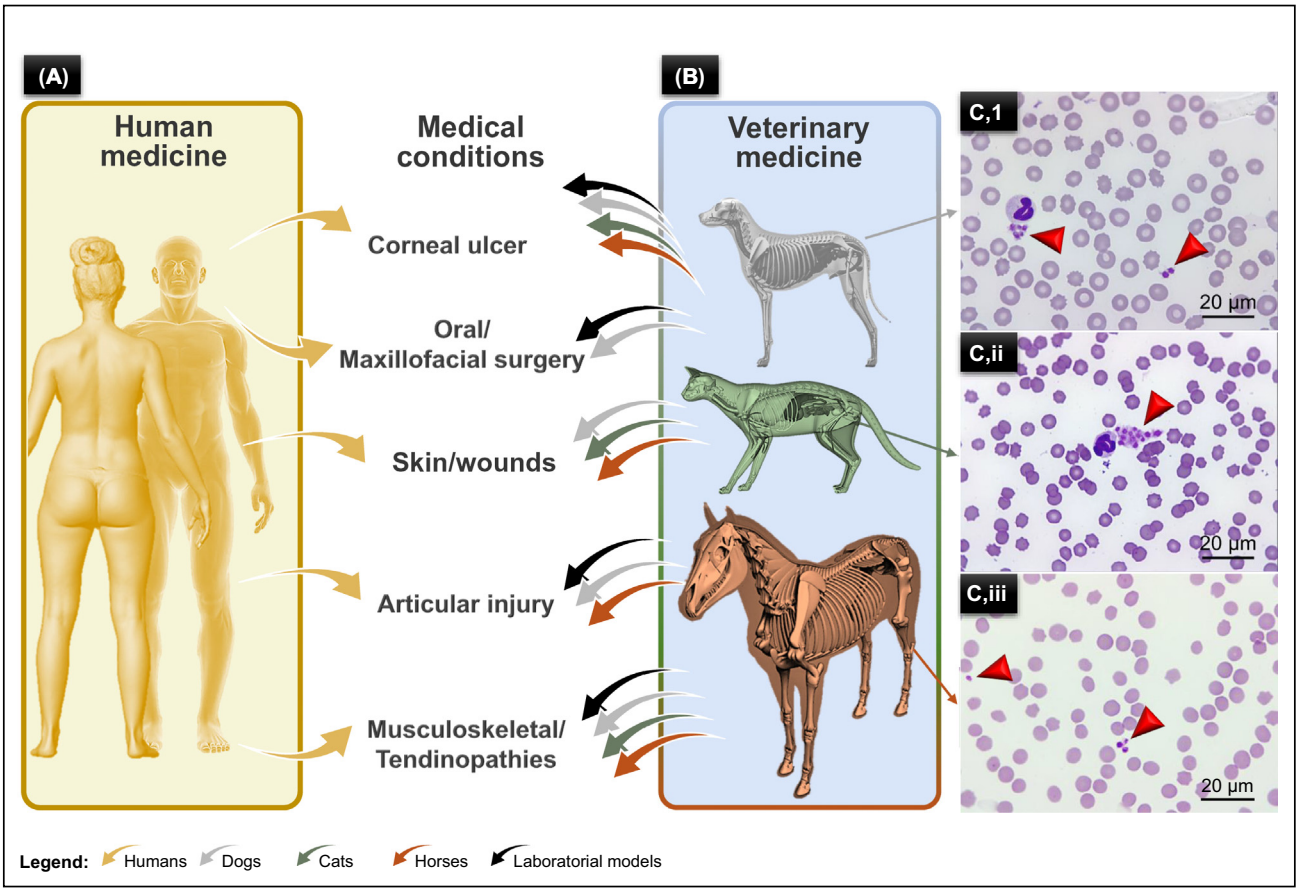

Figure 2. Veterinary Platelet-Derived Preparations Have Benefited from Advances in the Application of Platelet-Derived Product (PDP) Achievements in Humans, in a Translational Approach. (A) Schematic representation of clinical application of PDPs in both human and (B) veterinary pathological conditions, for which platelet formulations have been administrated. (C) Representative peripheral blood smears from domestic species, with platelets in evidence (red arrowheads), with felines exhibiting large feline platelet size when compared with equine and canine species [5,51]: (C,i) canine platelets, measuring 1–3 µm in diameter [5]; (C,ii) feline platelet clump often results in **pseudothrombocytopenia** in regular hematological analysis performed in this species, due to platelet aggregation, where thrombocytes are 2–6 µm diameter each [72]; (C,iii) equine species platelets, 2.5–3.5 µm in diameter [75].

therapy, demonstrated clinical improvement, others comparing PC alone versus the placebo group (saline injection), in the treatment of flexor tendon injuries, did not observe any beneficial effect of PC [62]. Likewise, intravenous allogeneic PC in combination with MSCs was successfully administrated to equine patients for ligament and tendon trauma treatment, registering improvements in the degree of claudication of the animals without adverse reactions [60]. In another study, treatments using intra-articular PL injections were cited for the treatment of osteoarthritis in horses, where nine of the ten affected animals improved on the lameness grade [50]. However, 1 year following the treatment, the horses relapsed. Other works did not report significant differences in the administration of PC alone versus PC combined with MSCs, or even MSCs alone, in the treatment of joint disorders [57].

Clinical Applications in Dogs

Surprisingly, studies focused on PC protocol standardization emerged only after its clinical application [8,63]. Similar platelet-based formulations attained by different preparation methods started to be applied in veterinary pathologic conditions, without detailed characterization/

Table 1. Studies of Platelet-Derived Products in Veterinary Medicine[a,b]

Species	PDP: type and characteristics	Study type	*n*	Pathology/lesion	Protocol	PLT, RBC, WBC, and GF content	Treatment outcomes	Follow-up	Refs
Equine	PC gel + BMMNCs, both autologous. Double centrifugation. Activation: 10% $CaCl_2$.	Clinical research	13 (9 + 4)	Suspensory ligament desmopathy (n = 9) and superficial flexor tendinopathy (n = 4), both refractory to other therapies.	Intra-articular. Ultrasound guided injection of BMMNCs suspended in PC, in the lesion core.	PLT: 751 ± 185 × 10^3/µl RBC: N/A WBC: N/A TGF-β1: 3055.0 ± 568.2 pg/ml PDGF-AB: 357.1 ± 102.2 pg/ml VEGF: 169.1 ± 17.1 pg/ml IGF: 289.2 ± 83.0 ng/ml EGF: 4.6 ± 2.8 pg/ml IL-β1: 3.9 ± 2.1pg/ml	Competition horses showed a marked improvement in their degree of lameness and 84.6% were able to return to competition.	12 months	[55]
Equine	PC + peripheral blood as the source of MSCs, both allogeneic. Double centrifugation. No activation before use.	Clinical research	20	Degenerative joint disease	Intra-articular. Horses were divided into four groups and injected with: (i) PC; (ii) MSCs; (iii) MSCs and PC; or (iv) chondrogenesis--induced MSCs and PC. The horses were evaluated by means of a clinical scoring system after 6 weeks, 12 weeks, 6 months, and 12 months after injection.	PLT: 200 × 10^3/µl RBC: N/A WBC: N/A GF/CYs: N/A	The combined treatments were significantly better than the PC treatment alone. No significant differences between the combined treatment and the MSC treatment alone. The combined use of chondrogenic-induced MSCs and PC generated better results.	12 months	[57]
Equine	Autologous PL. Double centrifugation. Activation: freeze-and-thaw cycles.	Clinical research	15	Osteoarthritis	Intra-articular. Horses were randomly allocated into two groups: ten horses received intra-articular injections of the PL and five horses received normal saline (control group, injected in distal interphalangeal joint. Two administrations, separated by 3 weeks.	PLT: 338.7 ± 76.2 x10^3/µl RBC: N/A WBC: 0.4 ± 0.2 x10^3/µl PDGF-AB: 258.0 ± 52.3 pg/ml	Most of horses (9/10) responded positively to the treatment (lower lameness grades compared with controls 10 days after the second injection) and return to normal athletic activity. One year after the treatment all horses relapsed to initial lameness score.	12 months	[50]

Equine	PC combined with MSCs tenogenically induced, both allogeneic. Double centrifugation. No activation before use.	Clinical research	104	Tendon or ligament injury: (two groups: SDFT or SL injuries)	Intralesional ultrasound-guided injection. One administration.	PLT: 100 × 10^3/μl RBC: N/A WBC: <100/μl GF/CYs: N/A	No adverse reaction. After 24 months, 82.4% (*n* = 56) of the horses with SL lesions were competing at their initial level. In the SDFT group (*n* = 21), 85.7% (n = *18*) of the horses were competing at their initial level after 24 months.	24 months	[60]
Canine	Autologous PC, using a commercial platelet filtration system (C-PET®, Pall Corporation). Number of centrifugations: according to manufacturers. No activation before use.	Clinical research	20	Osteoarthritis involving a single joint	Intra-articular. Dogs were randomly assigned to treatment or control groups. Control dogs received intra-articular injection of saline solution and treated dogs received PC injection.	PLT: 739 ± 365 × 10^3/μl RBC: N/A WBC: 15.1 ± 7.80 × 10^3/μl GF/CYs: N/A	In control dogs, lameness and pain scores at week 12 were not significantly different from pretreatment values. In treated dog's lameness scores (55% decrease in median score) and pain scores (53% decrease in median score) were significantly improved after 12 weeks.	12 weeks	[53]
Canine	ACP + hyaluronan versus ACP + methylprednisolone, autologous, using a commercial system (Arthrex®). Number of centrifugations: according to manufacturers. No activation before use.	Clinical trial (double-blinded)	10	Bilateral elbow osteoarthritis	Intra-articular. Dogs with bilateral elbow osteoarthritis were treated with hyaluronan plus methylprednisolone or ACP. The evaluation was performed before and 6 months after a single injection with one of the two treatments, before and 1, 6, 12, and 24 weeks after injection.	PLT: N/A RBC: N/A WBC: N/A GF/CYs: N/A	Both treatments have beneficial effects for dogs with bilateral elbow osteoarthritis and ACP registered a significant improvement, but without statistical differences between groups.	24 weeks	[54]
Canine	ASCS versus PRGF, both autologous. Commercial kit (PRGF-Endoret®). Single centrifugation. Number of centrifugations: according to manufacturers. Activation: with 5% of its volume with 10% $CaCl_2$.	Clinical research	53	Hip osteoarthritis	Single intra-articular injection for each group.	PLT: N/A RBC: N/A WBC: N/A GF/CYs: N/A	The ASC group obtained better results at 6 months. The study shows that ASCs and PRGF are safe and effective in the functional analysis at 1, 3, and 6 months, providing a significant improvement in the canine's life quality.	6 months	[11]

(continued on next page)

Table 1. (continued)

Species	PDP: type and characteristics	Study type	*n*	Pathology/lesion	Protocol	PLT, RBC, WBC, and GF content	Treatment outcomes	Follow-up	Refs
Canine	*Allogeneic* PC. Double centrifugation. Activation: equal volume of 10% $CaCl_2$.	Case report	1	Massive cutaneous hip lesion, induced by CID	Topical. Activated PC mixed with an equal volume of sterile saline solution, erythropoietin, and granulocyte colony-stimulating factor, applied directly in the wound.	PLT: 1000–1200 × 10^3/μl RBC: N/A WBC: N/A GF/CYs: N/A	Gradual and complete enhancement of the wound lesions 28 days after applying allogeneic topical PC.	28 days	[7]
Canine	Lysated PC (PL) + ASCs, both autologous. PC by double centrifugation. Activation: freeze-and-thaw process (on cycle).	Case report	1	Large open skin wound and bilateral lesions of the Achilles tendons	Topical and intra-lesion. PL: ASCs applied by dripping and/or spraying over the wound surface (every 48–72 h), as well as by injecting the platelet concentrate along the attached wound edge. Five applications at day 11, 17, 23, 31, 41.	PLT: 1000 × 10^3 μl RBC: N/A WBC: N/A GF/CYs: N/A	Complete wound healing in 3 months. Lameness of the hind limbs and the edema of the Achilles's tendon disappeared 20 days after intra-lesion application of ASCs and PC. Pigmentation of the skin appeared after about 2 months.	480 days	[59]
Canine	PC + BMAC versus PC + ASCs, both autologous. All using commercial systems (BMC Stem Cell Kit; PurePRP® kit). Double centrifugation. No activation before use.	Clinical research	36	Partial cranial cruciate ligament tear	Intra-articular. Retrospective study: medical records of client-owned dogs were reviewed for cases diagnosed arthroscopically with an early partial (≤50%) tear of the craniomedial band of the CCL that was treated with BMSC-PC or ASC-PC. Dogs were assessed at baseline and 30, 60, and 90 days post-treatment with an orthopedic and neurologic examination. The exact stem cell numbers injected are	PLT: 6–7-fold increased over whole blood RBC: 95% reduction WBC: 85% neutrophil reduction GF/CYs: N/A	Stifle arthroscopy at 90 days post-treatment were found on 13 of the 36 dogs. Significant difference was found between the treated limb total pressure index percent at day 0 and day 90. Treatment with BMAC-PC and ASCs-PC produced satisfactory results in early partial cranial cruciate ligament tears in dogs.	90 days	[56]

					unknown.				
Canine	Treatment with ASCs and PC, both autologous. Double centrifugation. No activation before use.	Clinical research	55	Supraspinatus tendinopathy	Intra-tendon. Ultrasound-guided injection of ASC-PC, into the tendon lesion. All patients were enrolled in rehabilitation therapy following ASC-PC therapy.	PLT: 3–4-fold increase over whole blood RBC: N/A WBC: 80–90% reduction GF/CYs: N/A	Following treatment, a significant reduction in tendon size was visible in the treated tendon, at 45 and 90 days after the injection. Supraspinatus size reduction in all cases and improvement in fiber pattern.	90 days	[58]
Canine	Autologous PC (Angel® System kit). Number of centrifugations: according to manufacturers. Activation: $CaCl_2$ + thrombin.	Clinical research	60	Anterior cruciate ligament rupture	Intra-articular. After surgical treatment, activated PC was placed at the osteotomy site of treated group. Control group received saline lavage.	PLT: 1370 ± 489 × 10^3/μl RBC: negligible WBC: 5.45 ± 3.5 × 10^3/μl GF/CYs: N/A	No significant effect of PC on osseous union promotion, as assessed with radiology, ultrasound, or with MRI.	70 days	[76]
Feline	Heterologous PC (from a donor dog). Double centrifugation. No activation before use.	Case report	1	Large contaminated skin defect (dog bite)	Topical, one single application directly to the surface of the lesion.	PLT: 1513 × 10^3/μl RBC: 0.20 × 10^6/μl WBC: 7.14 × 10^3/μl GFs: N/A	Heterologous PC with no local or systemic adverse effects. Complete wound healing within 20 days.	3 months	[65]
Feline	PRF combined with BM aspirate as the source of MSCs; both autologous. Supported by a 3D-printed polycaprolactone implant.	Case report	1	Recurrent chronic oronasal fistula (previous surgeries failed)	Whole blood in a polypropylene tube, without anticoagulant, was centrifuged for 10 minutes at 3000 rpm. Bone marrow aspirate was seeded in the PRF and a customized polycaprolactone 3D-printed implant supported the system. PRF + bone marrow placed in the defect, after fibrotic tissue removal. Surgical closure of the ONF.	PLT: N/A RBC: N/A WBC: N/A GF/CYs: N/A	A CT scan at day 75 after surgery showed uneventful closure of ONF. The patient regained its olfactory sense, food interest, and body condition. No osteomyelitis recurrence or dehiscence revealed.	10 months	[66]
Bovine	Allogeneic PC. Double centrifugation. No activation before use.	Clinical research	229	Mastitis quarters	Intramammary administration: 5 ml of PC alone or associated with antibiotic or antibiotic alone were administrated for 3 consecutive days, depending on the treatment group	PLT: 1000 × 10^3/μl RBC: N/A WBC: N/A GF/CYs: N/A	Association of antibiotic and PC performed significantly better than the antibiotic alone, either for the recovery of the affected mammary quarters or for somatic cell count reduction.	30 days	[67]

(Table legend on the bottom of the next page)

reference about its composition, just assuming the existence of platelets would produce positive therapeutic effects.

For the treatment of soft tissue injuries, autologous or allogeneic PCs have been topically administrated as wound-healing promoters [7,59]. Furthermore, PL was described for the successful treatment of a large open skin wound, combined with MSCs, for the treatment of bilateral injuries of the Achilles tendons caused by a train accident [59].

Applied research for the treatment of bone and articular injuries using platelet treatments has shown improvement on the lameness score after the application [53,54]. For instance, for ridge augmentation after tooth extraction in beagle dogs, PRP and PRF promoted bone maturation, with corticalization and lamellar bone formation, and PPP promoted a higher volume of new bone augmentation [3].

The efficacy of a single intra-articular injection of adipose MSCs or PRGF was assessed in a randomized study, in which cell therapy showed better results at 6 months after treatment than PRGF-Endoret® alone [11]. Likewise, a single intra-articular administration of adipose-derived stem cells, combined with PRGF-Endoret®, reduced pain and lameness in dogs with osteoarthritis over at least 6 months [64]. Considering cell-origin, research focused on the treatment of cranial cruciate ligament injury with autologous bone marrow aspirate concentrate or adipose-derived mesenchymal cells, both combined with PC, producing comparable amelioration of the symptoms [56].

Clinical Applications in Cats

Cats are naturally limited as low blood-volume donors, limiting the autologous administration of platelet-based formulation. Conversely, **heterologous** PC (from a canine donor) was described as an effective treatment of a feline's infected skin wound, caused by a dog bite, with no local or systemic adverse effects, based on a single administration without platelet activation [65]. Complete wound healing occurred within 20 days.

PRF was documented as a novel TE approach for the surgical repair of a recurrent and large chronic oronasal fistula in a senior cat, combined with bone marrow aspirate (demonstrated by the authors to have MSCs), supported by a tailored 3D-printed implant [66]. Soares and colleagues produced autologous PRF using a protocol modified from human literature, operating tubes deprived of clotting promoters [66].

Clinical Applications in Bovines

In bovine species, PC with 1000×10^3 platelets/μl was found to be beneficial in the treatment of mastitis, associated with antibiotics. PC performed significantly better than the antibiotic alone, as determined by the recovery of the affected mammary quarters and somatic cell count reduction in a 30 day follow-up period [67].

Platelet-Therapy: Boundaries and Opportunities

The literature reflects a limited number of clinical studies about hemoderivatives administration with regenerative medicinal interest. The majority of the published data exposes isolated case

Notes to Table 1:
[a]The original PRP preparations are cited in this table as PC (platelet concentrate).
[b]Abbreviations: ACP, autologous conditioned plasma; ASCS, adipose-derived mesenchymal stem cells; BMAC, bone marrow aspirate concentrate; BMMNCs, bone marrow-derived mononuclear cells; $CACL_2$, calcium chloride; CID, disseminated intravascular coagulation; GF/CYS, growth factors and/ or cytokines; L-PRF, leukocyte-platelet-rich fibrin; L-PRP, leukocyte-platelet-rich plasma; Min, minimum; MSCs, mesenchymal stem cells; N/A, not applicable or not available; ONF, oronasal fistula; PL, platelet lysate; PLT, platelet; PRF, platelet-rich fibrin; PRGF, plasma-rich in growth factors; PRP/PC, platelet-rich plasma or platelet concentrate, the latter terminology being the most accepted; PPP, platelet-poor plasma; RBC, red blood cells; SDFT, superficial digital flexor tendon; SL, suspensory ligament; WBC, white blood cells.

reports, or clinical case studies with a small population number, uncontrolled clinical trials with insufficient data relative to both diagnostic and post-treatment monitoring, and frequently with inadequate criteria applied during the clinical research, resulting in a high outcome bias [68].

PC and PRF are the furthermost applied PDPs in the veterinary field. Bearing in mind the data available, even with the inconsistent results of PDP clinical administration, it is prudent to consider that PDPs hold promising regenerative biological effects. Nevertheless, the composition of the principal players of PDPs (platelets, GFs, and even WBCs) are regularly neglected in the veterinary descriptions, particularly when commercial kits are used to obtain liquid PCs. Consequently, in a considerable number of veterinary reported researches, the therapeutic dose and application regime cannot be studied and defined, due to the absence of complete characterization of the applied PDP. Degenerative joint disease is one of the most investigated disorders suitable for hemoderivative use, but even so, it is the one with more uncertainty due to the critical points previously stated.

However, numerous optimization protocols have been described for the attainment of PCs, reflecting a lack of consensus on PC production. Several protocols have been entitled as successful in their great final purpose (to obtain a liquid platelet-enriched formulation), accomplishing similar results, corroborated by final efficient platelet concentrations (Box 2).

Similar to the human field, veterinary published data should be addressed with pragmatism and the results should be called into question, starting with the restraints inherent to the manufacture procedure. Moreover, contemplating the diversity of optimization procedures described for PC preparation, considerable boundaries should be perceived: low donor variability among the populations used for the optimization works, reflected by low population stratification; low population number; ratio of anticoagulant volume and whole blood used; hematocrit level presented by the donor; inappropriate reference to gravitational forces used; and lack of information about the time elapsed between sampling and hematological analysis, particularly considering the occurrence of natural platelet aggregation in PC units [68–72]. For example, PDPs manufactured from a protocol attained from a low stratified donor population (from same age, breed) may contribute to inconsistent clinical performance of that platelet formulation [73].

Box 2. The Diversity of Optimization PRP/PC Protocols in Horses, Dogs, and Cats

Equine sports medicine was the first veterinary area to use PDP approaches, for the treatment of tendinopathies and joint injuries in competition horses, with the expectation of obtaining similar results to humans [50,55,57]. Curiously, platelet-derived preparations, namely PC, began to be frequently recommended in human professional sports activity for increasing physical capability. In 2010, PRP was included on the World Anti-Doping Agency (WADA) Forbidden Substances List, being latter readmitted. Nevertheless, individual GFs administrated separately are still prohibited in professional human competition [86].

Although clinical administration of PCs first began in equines, a scarcity on the optimization protocols is uncovered by the available published data. Of all the domestic species, canines have been extensively studied. The literature reflects different protocols for canine PC preparation, mainly, double centrifugation procedures. Herein, protocols range from 610 ×*g* for 10 minutes, followed by 1600 ×*g*/15 minutes [87], to 1000 ×*g* for 5 minutes, followed by 1500 ×*g* for 15 minutes [88].

There are insufficient descriptions regarding feline PC optimization protocols, with one study using a single centrifugation step, under low centrifugation forces (85 ×*g* for 6 minutes), with the collection of a plasma portion above the RBC-plasma fraction [89].

PC quality, recognized in platelet concentration present in autologous formulations, is straightforwardly related to both preparation protocol and methodology, adding a source of variation within treatment results.

Nevertheless, PRF is presented as a consistent PDP and, among all the hemoderivatives, its manufacture is the only one that is unanimously described. Autologous PRF efficacy as a biological tissue healing promoter has been validated in human treatments. The minimalistic PRF manufacture procedure (a safe, easy, and cost-effective strategy, with no requirement of advanced manufacturing skills), allied to inherent low variables in its preparation methodology, high level of reproducibility, and consistent clinical performance have determined the success of this biological product [39,74]. To date, PRF use in veterinary patients has been rarely described, despite the promising results accomplished in humans.

PRF should be observed as an organic and biodegradable scaffold, having the capacity to release important GFs and other structural proteins along time, at high concentrations, contributing to angiogenesis and, thereby, to the orchestration of tissue healing [42].

Reflecting on both clinical and laboratorial team expertise, and the attributes previously described, the authors believe PRF may become the most promising formulation for veterinary medicine, suitable for regenerative skin wound therapy and critical corneal defects, following human outcomes.

Concluding Remarks and Future Directions

Financial consultants claim that the market for age-related pathologies such as joint disease is exponentially growing (https://www.futuremarketinsights.com/reports/canine-arthritis-market), with major pharmaceutical investment in the discovery of new treatments. Platelet therapy offers an alternative to conventional pharmacologic treatments that are usually needed in chronic conditions; they stimulate the regeneration of injured tissues, for which the conventional medical or surgical approaches do not provide a satisfactory solution. These conventional treatments are usually expensive and present considerable side effects associated with prolonged use, especially in veterinary geriatric patients, who are frequently affected by other pathologic conditions. Skin wounds, tendinopathies, and joint degeneration usually require a long administrative course of nonsteroidal anti-inflammatory drugs, opioids, and nutraceutical supplements to attenuate pain. These treatments are frequently prolonged, registering relapse periods and with refractory responses, and aggravating the mobility state of the animal. Associated problems include the lack of owner compliance and caregiver exhaustion.

Preparing PDPs is extremely challenging from a technical standpoint due to the delicate nature of platelets that become easily activated during processing. Therefore, special recommendations are necessary to maximize the clinical benefits of these hemoderivatives (Box 3), which cannot always be evaluated in the published literature.

So far, the therapeutic concentration of platelets in PDPs for veterinary applications, especially for individual species, is not clearly well-defined. Therefore, works in the veterinary field should more thoroughly characterize PDPs (see Outstanding Questions), with standardized protocols for clinical applications according to the species in question. Subsequently, more systematically describing the therapeutic outcomes of PDPs in future works on this subject will lead to more easily comparable data.

Currently, the different clinical cases and studies reported in the scientific literature are not integrally comparable with each other, which impairs the scientific discussion on platelet-based therapies. Substantial veterinary studies do not characterize the applied PDP, in particular regarding platelet, erythrocyte, leukocyte, GF, and cytokine content. Concurrently, when preparation protocols for PDP achievement are described, very often there are no indications of important

Outstanding Questions

What should the consensus and guidelines be in regulating the quality and safety of PDP bio-fabrication, especially when they are manufactured in clinical environments?

How feasible is it for all veterinary clinical trials or case reports involving PDPs to document the platelet, WBC, RBC, and even GF concentrations?

Is it important to describe both whole blood and final platelet-rich formulation volumes when different optimization or preparation protocols are discussed?

Could PRF become the most versatile and clinically accessible platelet-enriched product available for veterinary regenerative treatments that can be used in several medical or surgical applications?

Is platelet lysate a suitable biotechnological option for allogeneic delivery, as a vehicle with low immunologic reactivity in veterinary clinical context?

How can platelet lysate best reach its high potential to become a promising bio-fabricated platelet formulation for allogeneic veterinary use, contributing to reducing donor-to-donor variability observed in individual veterinary platelet-based formulations?

Box 3. A Call for Standardization: From Quality/Safety Concerns to Characterizing Veterinary PDPs

PDP manufacture presents some safety and quality concerns, especially when performed by in-clinic techniques (Figure I): risk of accidental contamination during preparation and absence of methods ensuring sterility at all stages; potential infectious agent transmission in allogeneic formulations, particularly in periods of subclinical disease; nonexistence of standard protocols in veterinary medicine, with qualitatively and quantitatively characterized PDPs.

For the definition of a standardized PC production, the centrifugation speed, time, and preparation temperature seem to be crucial, without underestimating the blood element concentration [41]. Citrate-related anticoagulants are recommended for PDP production in human and veterinary blood banks, allowing platelet survival, maintaining anticoagulation through reversal calcium binding, and cell membrane stabilization. Additionally, during the preparation of PDPs, the ratio between anticoagulant and blood volume should not be neglected, to avoid platelet agglutination [90].

The short life expectancy of platelets demands special requirements during storage and manipulation, considering platelet preservation time in whole blood units is 5 days, under constant/intermittent agitation [90,91]. With respect to PDP administration, autologous methodology is considered the safest [8]: it avoids antigeneic reactions [1,8], prevents transmittable diseases, involves a simple venous blood collection, and is easily preformed in-clinic. Nevertheless, some limitations have been pointed out for autologous delivery: patients should not have pre-existing metabolic, hematologic, and/or immunological disorders; small blood-volume donors may be not suitable; and donor-to-donor variability might result in variations in PDP composition, namely in platelet concentration, growth factors, or hormones [48].

Regarding allogeneic administration, one of the main risks is alloimmunization (host's immune system reacts against donor's antigens), which is minimized when pooled PC are grouped in accordance to blood type and compatibility testing [90,91]. Moreover, allogeneic therapies carry the risk of transmissible diseases. In these cases, donor animals should be rigorously and broadly screened for specific blood-borne pathogens in animal species (e.g., *Leishmania* sp., *Rickettsia* sp., etc.). Not less important, bacterial contamination is higher due to processing manipulation steps and all PDPs prepared for clinical use should be prepared under strict aseptic technique, preventing cross-contamination in scenarios potentiated by nosocomial infections, such as veterinary clinics/hospitals. Blood manipulation between the different production steps, without a laminar flow chamber to assure air septic conditions, may result in bacterial/fungal contamination occurrence, especially in clinical facilities.

Categorization of PDPs according to their features should be encouraged for a standardized recognition of veterinary platelet-inspired applications (Figure II).

The existing hemoderivatives have individual specificities, conferring demarcated characteristics for particular applications, namely in tissue engineering of clinical scenarios (Table I).

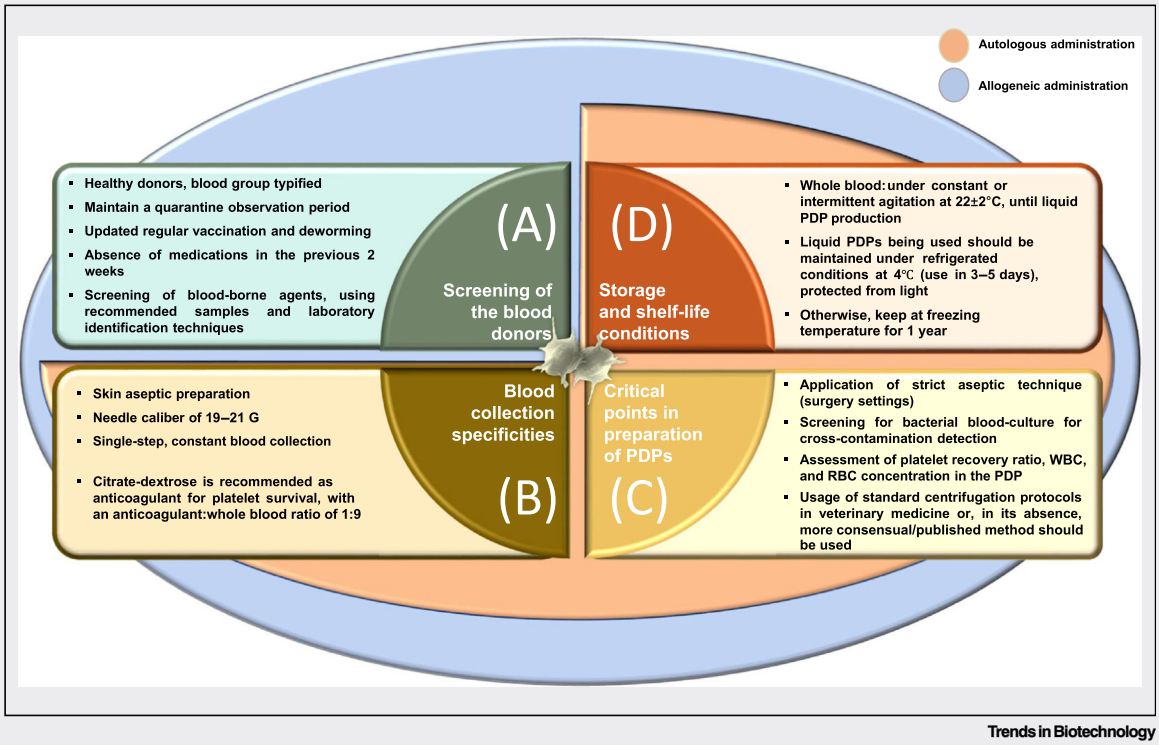

Figure I. Criteria Involved in the Obtainment of Platelet-Derived Products (PDPs) for Veterinary Clinical Applications. The production of PDPs should obey specific requirements, respecting donor selection (A), blood collection process (B), critical control points (C), and storage and shelf-life conditions of the formulations (D). The suggested criteria were extrapolated from human and veterinary consensus and guidelines about platelet-enriched product manipulation, regarding the PDP preparation, handling, and storage, for autologous or allogeneic administration [36,90,92–95]. Abbreviations: RBC, red blood cell; WBC, white blood cell.

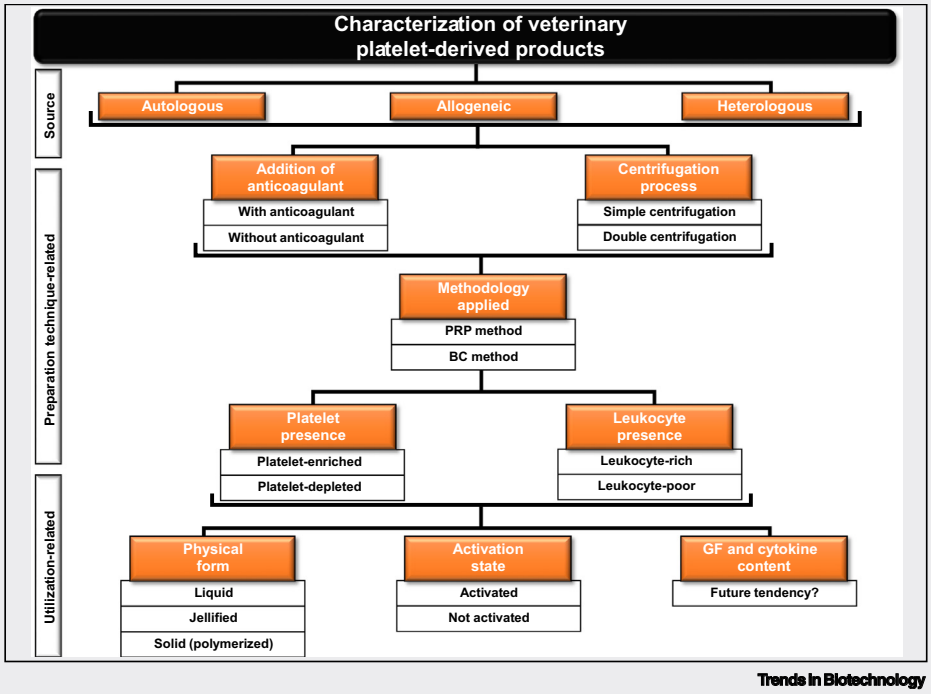

Figure II. Proposed Classification for Veterinary Platelet-Based Products. Abbreviations: BC, buffy coat; GF, growth factor; PRP, platelet-rich plasma.

Table I. Important PDP Features for Veterinary Applications

Features	PDP type[a]				
	PRP/ PC	PRGF	PPP	PRF	PL
Form	Liquid	Gel, after activation	Liquid	Solid	Liquid
Anticoagulant	Yes	Yes	Yes	No	Yes
Activation	Optional	Advised	Not necessary	Not necessary	Not necessary
Manipulation	Complex	Complex	Complex	Simple	Very complex
Time consumption	++	+	++	+	+++
Yield	Low	Low	High	Low	Low
Reproducibility	Possible bias	Possible bias	Possible bias	High	Moderate
Economic cost	Low	High	Low	Very low	Very high
Versatility	Moderate	Moderate	Low	High	Moderate
PLTs	+ + +	++	Vestigial	+++	Vestigial
WBCs	++	+	Vestigial	+++	Vestigial
RBCs	+	+	Vestigial	Vestigial	Vestigial
GFs/cytokines	++	+	+	+++	+++
Fibrinogen	+	–	+++	–	+
Clinical applications	Wound repair Articular disease Tendon injuries Corneal healing Cell culture supplement	Wound repair Corneal healing Tendon injuries Corneal healing Bone grafting materials	Corneal healing Biological glue	Wound repair Corneal healing Periodontal disease Bone repair Surgical tissue filler Bone grafting materials Tendon injuries	Wound repair Corneal healing Tendon injuries Corneal healing Periodontal disease Cell culture supplement
Refs	[3,4,14,18,44,66,96–99]				

[a]+++, high content; ++, median content; +, low content; –, no content.

parameters (e.g., volume of anticoagulant used, volume of both blood collected and final PDP achieved, working temperature, time elapsed between the harvesting/processing/analysis of the samples), which leads to debatable and noncomparable analysis. Furthermore, different methods can allegedly be used to produce similar PDPs, cited terminology is repeatedly different for similar PDPs and, often, proteins are not quantified.

At the same time, more clinical research is required, with precise case descriptions, using well-defined PDP compositions and validated scores and instruments to assess the pathologic conditions. Also, comparative research based on consistent and well-defined criteria, using representative numbers of participants and with long-term follow-up periods should be carried out to better understand the benefits of PDPs.

Consensual documents and regulatory guidelines have been proposed recently in human contexts. To the best of our knowledge, so far there are no equivalent guidelines with respect to veterinary settings. We recommend that clinicians and researchers use only well-characterized PDPs (Box 2) and report the administration route, frequency, and treatment period [36] (Box 3). Only then can therapeutic PDPs be properly used in a translational approach and their effectiveness for veterinary applications finally determined.

Disclaimer Statement

The authors declare the following competing financial interests: Pedro Carvalho is the CEO and Founder of Vetherapy, a biotechnology company that commercializes cellular therapeutic solutions and platelet-derived products for veterinary medicine. The other authors declare that there are no conflict of interests regarding the publication of this paper.

Acknowledgments

Authors thank VetLamações Small Animal Clinic (Braga, Portugal) for feline and canine blood samples and to Dr Ana Jacinta Ribeiro for equine blood samples. This work was supported by Fundação para a Ciência e Tecnologia, from Portugal SFRH/BD/118871/2016 PhD grant. The authors acknowledge the financial support from the European Union Framework Program for Research and Innovation HORIZON 2020, under the FOOD4CELLS project (PTDC/CTM-BIO/4706/2014 - POCI-01-0145-FEDER 016716).

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