Guidelines for the Use of Platelet Rich Plasma

Presented by

The International Cellular Medical Society

DRAFT

Version 1.0

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The International Cellular Medical Society (ICMS) asserts that a need exists to create standards for platelet rich plasma (PRP) protocols, preparations, techniques and tracking. We believe that physician-led organizations will serve the needs and interests of both patients and physicians toward achieving the best outcomes. In order to advance PRP in particular (and autologous cellular medicine therapies in general), we have developed these guidelines to assist physicians in performing safe therapies, promote patient education, encourage robust clinical research and begin to define the scope and anticipated effects of these procedures.

Platelet Rich Plasma: Historical Perspective

The application of PRP has been documented in many fields. First promoted by M. Ferrari in 1987 (1) as an autologous transfusion component after an open heart operation to avoid homologous blood product transfusion, there are now over 5200 entries in the NCBI for PRP ranging in fields from orthopedics, sports medicine, dentistry, otolaryngology, neurosurgery, ophthalmology, urology, wound healing, cosmetic, cardiothoracic and maxillofacial surgery.

The initial popularity of PRP grew from its promise as a safe and natural alternative to surgery. PRP advocates promoted the procedure as an organically based therapy that enabled healing through the use of one's own natural growth factors. In recent years, scientific research and technology has provided a new perspective on platelets. Studies suggest that platelets contain an abundance of growth factors and cytokines that can affect inflammation, postoperative blood loss, infection, osteogenesis, wound, muscle tear and soft tissue healing. Research now shows that platelets also release many bioactive proteins responsible for attracting macrophages, mesenchymal stem cells and osteoblasts that not only promote removal of degenerated and necrotic tissue, but also enhance tissue regeneration and healing.

Musculoskeletal practitioners began using PRP for tendinopathy in the early 1990s. These early practitioners were primarily trained in the use of prolotherapy. The popularity of PRP grew as physicians began to see clinical results in concentrating a patient's own blood factors. The PRP procedure is significantly more complex and requires additional equipment to perform successfully, but many practitioners have seen a relatively more robust response, fewer treatments and improved tissue health compared to prolotherapy.

The growth of PRP therapy has relied primarily on anecdotal or case reports. Historically, there have been few controlled trials to prove the efficacy of PRP. Of these existing trials, the sample sizes tended to be too small to allow for generalization of findings. Moreover, lack of concensus on technique, number of injections, spacing of injections, number of platelets, concentration of platelets over baseline, with or without leukocytes in the injection, exogenous activation of injected platelets and even a definition of appropriate candidates for the procedure are lacking and in need of further definition and evaluation. Recently, however, there has been an emerging literature on the beneficial effects of PRP for chronic non-healing tendon injuries including lateral epicondylosis, plantar fasciopathy and cartilage degeneration (2,3).

The current guidelines will focus on general principles of PRP use and its applications specifically to musculoskeletal care. Future sections will be added in reference to other fields such as dentistry, cosmetics, wound healing, etc.

Platelet Rich Plasma: Definition and Preparation Considerations

By definition, PRP must contain a higher concentration of platelets than baseline, however an increase in platelets is a very gross description of PRP and does not accurately describe the variability among different types of PRP. There are several parameters that need to be taken into account when considering PRP, including: platelet concentration above baseline, whether or not leucocytes are included, whether or not the PRP has been anticoagulated and whether it requires exogenous activation.

Platelet count is the first variable to consider. Absolute platelet count varies depending on the platelet concentration in the subjects' peripheral blood. PRP devices can be usually divided into lower (2.5 - 3 times baseline concentration) and higher (5 – 9 times baseline concentration) systems. It would seem intuitive that a higher platelet count would yield more growth factors and better clinical results, however, this has not yet been determined. Graziani et al suggested that the optimal concentration of PRP was 2.5x baseline and above this there may be an inhibitory effect (4). More research is needed.

PRP containing white blood cells will have different biologic activity than PRP in which they are absent. The lower platelet count systems separate the whole blood into two components: one with the cellular components and the other consists of serum in which the platelets are suspended. The higher platelet count systems separate the whole blood into three fractions: the red cells, serum and buffy coat. The buffy coat contains both platelets and white blood cells (WBCs).

WBC can be further classified into different types. These include neutrophils, monocytes/macrophages, and lymphocytes. Their roles in tissue healing are different. Neutrophils are phagocytic and contain over 40 hydrolytic enzymes. Their activation leads to phagocytosis of debris and the release of oxygen free radicals and proteases. This release of toxic molecules from the neutrophils can lead to secondary damage to the muscle (5,6). Whether or not neutrophils have a negative or positive effect on acute or chronically injured soft tissue is unknown.

Macrophages are the tissue form of the circulating monocytes. Their role is the removal of debris and they are primarily phagocytic. They also have a role in balancing the pro-inflammatory and anti-inflammatory aspects of healing (5,7) Since it is not possible to fractionate different types of white blood cells out of PRP, it may be that the absence of macrophages is more detrimental to healing than any secondary damage inflicted by neutrophils. More study is also needed in this area.

When whole blood is drawn, many PRP kits will use an anti-coagulant to prevent it from clotting. Most kits use anticoagulant citrate dextrose (ACD) to inhibit clotting. ACD binds calcium and prevents the coagulation proteins from initiating the clotting cascade. The addition of citrate to the blood also makes it more acidic than is physiologic. Since some growth factors are influenced by the pH of the tissue, some protocols recommend buffering the PRP back to a physiologic range prior to injection.

PRP is activated prior to injection is another parameter that requires further discussion. PRP can be activated exogenously by thrombin, calcium chloride or mechanical trauma. Once PRP is activated, a fibrin network begins to form, solidifying the plasma and creating a fibrin clot or membrane. If PRP is activated too strongly, the fibrin network will be a bivalent, unstable network. If it is activated in a more physiologic manner, a tetramolecular stable network forms that enhances enmeshment of cells and growth factors. (8) Although this can be useful for surgical procedures, it is undesirable to have the PRP overly viscous when injecting into soft tissue.

Activation results in rapid growth factor release, with 90% of the prefabricated factors released in ten minutes. Many growth factors have short half-lives, so greatest effectiveness may result if they are activated at or just prior to injection. Variable half-lives of growth factors also creates a differential PRP make-up depending on how quickly after activation it is used. Most commercial PRP kits do not activate PRP. Some replace calcium that was bound by ACD to create a more physiological state. Employing unactivated PRP may result in a more normal physiologic activation by the injected tissue.

To avoid unintentional activation of platelets, most protocols use large bore needles (>22) to draw the blood and re-inject PRP. In addition, there are different centrifuge protocols with different spin speeds and times. Some centrifuges offer special braking mechanisms to prevent unintentional activation. The optimal regimen to prevent unintentional activation is unclear. Collagen is a natural activator of PRP, thus when PRP is used in soft tissue, it does not need to be exogenously activated.

Once activation has occurred at the injection site, release of growth factors initiates an inflammatory response that lasts approximately 3 days (9). Fibroblasts accumulate at the site of injection, which marks the beginning of the proliferative phase of healing that lasts several weeks. After that, remodeling occurs to the collagen matrix that was laid down by the fibroblasts. This remodeling phase that leads to the formation of mature tissue lasts about 6 months. It takes all three phases for new tissue to form and provide long-term stability to tissue (10).

Platelet Rich Plasma: Physician Accreditation

PRP injections should be performed by allopathic or osteopathic physicians with a valid license to practice medicine and surgery in the State in which they practice. Successfully and safely performing PRP procedures requires knowledge of the diagnosis, standard treatments, benefits, risks, contraindications, methods of preparation and delivering it to the appropriate patient in the appropriate situation.

Physicians who perform PRP injections should be board certified according to ACGME standards and maintain appropriate overall and musculoskeletal specific continuing medical education credits. Topics specifically encouraged include diagnosis, treatment, and anatomical and radiological findings in chronic pain or acute neuromusculoskeletal injuries. Specific CME accredited courses with updated findings, procedures and uses of PRP are strongly encouraged.

Physicians who perform PRP injections should be familiar with the peer reviewed literature and the usual treatment of the diagnoses they are considering for PRP treatment and the benefits, risks, and methods of PRP injections, and adhere to ICMS/AMSSM guidelines for the handling and delivery of PRP.

At this time, there are no governing bodies to accredit the application or performance of PRP. This is considered the practice of medicine.

Platelet Rich Plasma: Physician Training

1. *Basic Physician Training:*
   The Physician should have completed a residency or fellowship detailed in training of the musculoskeletal system. This will assure the physician was instructed in obtaining a proper history, performs a detailed neuromusculoskeletal examination, considers appropriate differential diagnoses, understands the usual treatments for the common diagnoses and has the ability to consider alternative, complimentary or advanced treatment options.
2. *PRP Training*
   We suggest attendance at training courses on the preparation and use of PRP or extensive peer instruction on the use of PRP grafts by a physician accustomed to PRP therapies. There are currently no ICMS/AMSSM approved courses, however, suggestions will be forthcoming.
   It is suggested that appropriate conservative measures have been exhausted to both the physicians' and patient's satisfaction prior to PRP use.
   The physician must be able to determine the appropriate indication and contraindications for PRP use, consistent with ICMS/AMSSM standards.
   The physician must articulate the appropriate risks, benefits, expected course, follow-up and acceptable activities to the patient.
   The physician must have training and the understanding of appropriate graft selection and preparation of such a graft with or without additive supports (calcium, thrombin, etc).
   The physician must be trained in the recognition and management of any complications.
   The physician must have the training and comfort with use of proper pain management strategies for post-procedure pain control.
   Optimizing patient outcome by use of adjunctive bracing, physical therapy, medications and other strategies is strongly encouraged.
3. *Injection Guidance Training*
   In light of safety and efficacy differences between guided and blind injections, we adhere to guided injections as the general standard with PRP injections. Individual physician ability and patient preference may alter this choice on a case-by-case basis.
   The physician should have training and expertise in the appropriate choice and use of guidance technology (ie CT, fluoroscopic, ultrasound, etc.) either through residency, fellowship, sufficient post-graduate continuing medical education, or clinical proctoring (peer-to-peer training). A weekend course in isolation is not sufficient.
   a) Established CT or fluoroscopic privileges will suffice for use with PRP.
   b) American Institute of Ultrasound Medicine (AIUM) accredidation for ultrasound guidance will suffice (when instituted).
      1) In the absence of AIUM accredidation, we believe that a MINIMUM of one high quality basic ultrasound course and one advanced course with particular training in procedures and

needle guidance should be the standard coursework. Moreover, an appropriate amount of completed injections such that the physician consistently, safely and effectively places a needle in the correct structure without damage to vital surrounding structures is required.

Platelet Rich Plasma: Indications

As with any musculoskeletal complaint, a thorough history and examination is required to reach a differential diagnosis. Additional diagnostic studies may be warranted and review of the prior "failed" treatments. PRP is generally considered an elective treatment for subacute and chronic conditions. Generally, healing slows or stops 6-12 weeks after an acute injury. If a patient has had no improvement for over six weeks, it is possible their healing phase has arrested. In overuse or repetitive conditions, it can be more challenging to isolate the transition from an acute phase.

We present some of the common orthopedic indications for which PRP is used along with a partial-not circumspect-review of current evidence for each indication. We believe that evidence based medicine is a helpful tool, but a great deal of the art of treating a patient with concurrent modalities is very difficult to quantify in the RDBCT trial.

Tendinopathies

Tendinopathy refers to a degenerative condition of tendons marked by the chronic loss of collagen, tissue integrity, stability and strength. Tendinopathy is not an inflammatory condition, as histologic specimens lack inflammatory cells (11). Causes are multifactorial, but natural aging, injury, repetitive stress, neural, vascular and hormonal inputs all likely contribute. Whereas tendinopathy is nearly ubiquitous as we age, pain and dysfunction generally occur only when sufficient stresses are applied to the degenerated tendon.

Basic science and animal studies are supportive for the use of PRP in tendinopathy. Lab studies have shown improved tenocyte proliferation, collagen deposition and endogenous growth factors (12). Animal models with surgically induced lesions are common and show good results (13). However, clearly positive high-level human evidence is still lacking in RDBPCT trials (14), and others have shown negative results (15). Mischra & Barnett have separately produced convincing case series' on recalcitrant tennis elbow (2) and plantar fasciosis (3).

We encourage further research on technique, number of injections, spacing of injections, number of platelets, concentration of platelets over baseline, with or without leukocytes in the injection, exogenous activation of injected platelets, use of common and validated outcome measures (i.e. VISA scores) in studies and patient selection.

Ligament Sprains

Most ligament studies on humans to date have been in combination with surgical anterior cruciate ligament reconstruction. Overall, the evidence suggests improved pain, healing and graft quality (16). The sports medicine literature with regard to non-surgical care is lacking at this point in time. Anecdotal evidence from expert sources indicate improved time of healing, reduced pain and reduced time to return to sport. We encourage more research in this area.

Muscle Strains

Muscle strains are a very common source of pain in dysfunction, particularly in the athlete. Muscles are rich in blood supply and generally heal with usual care, approximately 8 times faster than ligaments. If a subacute or chronic condition developed, consideration for PRP treatment would be acceptable. In rare situations, the delivery to an acute injury in an attempt to facilitate function could be considered, but there is insufficient evidence to endorse this currently. A study by Sanchez has shown faster recovery of acute muscle tears with PRP injection (17). If applicable to larger scale human use, it is unknown if this is relevant clinically, functionally or for return to sport. Treatment of myositis ossificans with barbotage, aspiration and PRP injection is an area of specific interest to consider.

Joints

Osteoarthrosis (OA) is a chronic degenerative condition of hyaline cartilage. OA accounts for profound morbidity, pain and health care expenses. The consequences to the individual and to the population as a whole are very significant, particularly with our aging population. There are few validated interventions that improve the clinical condition of a patient once the degenerative process becomes symptomatic. Given the lack of response of the body's healing mechanisms to degenerative conditions generally, injection of growth factors and cytokines is sensible. Lab and animal models exist for using PRP in OA with generally favorable results (18). A recent article by Kon et al (19) indicates improved functional outcomes. It is unknown whether PRP acts by local paracrine factors to alter pain, by new hyaline or fibrocartilage formation or a combination of both or neither. We encourage further high quality studies, pre/post imaging and joint fluid analysis to help elucidate the effects.

Animal models describe improved healing in meniscus, glenohumeral labrum and OCD with induced defects, but human studies are currently lacking in these areas.

Intervertebral Discs

Animal models using various preparations and matrices show encouraging results, however, no human studies exist. Placing PRP in a disc will necessitate damage to the disc, which has been shown to be potentially permanent after discography. Due to the close proximity of critical neurologic structures to the posterior annulus, CT or fluoroscopic guidance would be the preferred methods of regenerative factor placement in discs.

Nerves

Entrapment neuropathies that have failed "conservative management" have traditionally been treated with surgical release/decompression (neurolysis). With the advancement of musculoskeletal ultrasound, peripheral nerves and their adjacent structures can now be clearly visualized. There is growing experience in performing percutaneous release of nerves using different solutions (termed hydrodissection or hydroneurolysis) (20). There is insufficient information to endorse PRP treatment for this use, however, in cases of ischemic damage to a nerve due to scar tissue banding, there is theoretically a role for PRP during percutaneous procedures and we encourage further investigation.

Fracture Non-Union

Fracture non-union is a debilitating, albeit fortunately rare complication in the care of fractures. PRP has been shown to be inferior to recombinant BMP-7 to speed non-union healing in one randomized human study (21). The role in acute fractures in humans has not been well evaluated and seems impractical given the rate of successful healing without intervention. PRP has been used in spinal and joint fusion

surgeries with success. Further study of fracture non-unions would be of value to the literature base and may hold significant benefit for patients if extended courses of healing or bone stimulator use could be avoided.

Platelet Rich Plasma: Contraindications

Absolute Contraindications:

- Platelet dysfunction syndrome
- Critical thrombocytopenia
- Hemodynamic instability
- Septicemia
- Local infection at the site of the procedure
- Patient unwilling to accept risks

Relative Contraindications:

- Consistent use of NSAIDs within 48 hours of procedure
- Corticosteroid injection at treatment site within 1 month
- Systemic use of coriticosteroids within 2 weeks
- Tobacco use
- Recent fever or illness
- Cancer- especially hematopoetic or of bone
- HGB < 10 g/dl
- Platelet count < 105/ul

Platelet Rich Plasma: Protocols, Technique and Safety Recommendations

Protocol/Technique

Generally speaking, the procedure only requires the physician and an assistant to aid in preparation of a PRP graft, maintenance of aseptic technique and saving images on ultrasound (if applicable).

Pre-procedure Considerations

1) There should be a specific indication correlated with physical exam and confirmed with imaging studies such as x-ray, ultrasound, MRI, or CT scan prior to treatment.
2) Appropriate patient education and discussion has occurred with an informed consent signed prior to the initiation of the procedure.
3) Contraindications to the procedure are reviewed prior to initiation (see above).
4) Analgesics (no NSAIDs) or anxiolytics have been administered, if applicable

Graft Preparation

1) The patient is placed in a comfortable seated or recumbent position.
2) Sterile single use needles and syringes should be used with appropriate handling and disposal.
3) Using aseptic technique (see below), an appropriate amount of venous blood is obtained for the given procedure.
    a. Single-stick draws are preferred to decrease chances of activation.
    b. If a vein is passed through completely, blood flow is not smooth, needle comes out of vein or multiple attempts at a single site occur, consideration of a second site should be given.
    c. If the patient is a difficult draw, consider using ultrasound to guide the drawing needle.
4) Using sterile technique, the venous blood is transferred to the centrifuge. PRP should be obtained using a separating device designed for autologous blood. Preference is given to a closed system that prevents exposure of the blood and cellular components to the open air in the room and allows for minimal manipulation of the tissue.
5) If multiple patient grafts are prepared concurrently, proper labeling of each graft should be completed to ensure no cross contamination or the graft being used on the wrong patient.

Image Guidance

1) Real-time image guidance using CT, fluoroscopy or ultrasound should be used when injecting PRP.
2) If ultrasound is used, the following considerations should be decided upon in advance:
    a. Sterile gel. We recommend this for longer procedures, intra-articular injections, and any injections around the spine. Universal use has not been shown to improve infection rates, and in the setting of simple soft tissue injections, judicious use of aseptic technique is sufficient.
    b. Sterile probe covers. We recommend probe cover use with longer procedures (percutaneous needle tenotomy, etc). Cleansing of the probe before and after procedures and adherence to aseptic technique is sufficient. Covering the probe with sterile wound products (tegaderm) or using sterile gloves are other options that have been used in the community with success.
    c. Scout images and indelible markings of the site of probe position and needle entry should be made prior to final cleansing of the skin.

PRP Injection

1) The patient is placed in an appropriate and comfortable position that allows for sterility and access to the site of injection.
2) All necessary materials for the injection (PRP, additives, 4X4s, needles, US gel) should have been planned and placed on a sterile table adjacent and easily accessible to the physician.
3) The patient's skin is cleansed appropriately and towels or drapes may be used to create an aseptic field.
4) If local anesthetic will be used, it is to be applied with aseptic technique. See above discussion on anesthetic effects on PRP; consider infiltrating only the local subcutaneous area with anesthetic. Consider nerve block for larger/longer procedures (tenotomies).
5) If ultrasound is used, apply gel consistent with markings made previously.
6) Complete the injection with real-time recording of images.
7) Apply a dressing or appropriate bandage to protect the needle entry site.

Post-Injection

1) Monitor for post-procedure complications (vaso-vagal most common)
2) Patients should be given post-procedure instructions, precautions, and emergency contact information.
3) Protocols for immobilization and post-procedure activity allowed/encouraged vary widely. Future recommendations will be forthcoming once protocols are more widely accepted +/- studied.
4) Post procedure analgesic prescription should be dispensed. Avoid NSAIDs until the patient has healed, is pain free, has full function or has reached a plateau.
5) Contaminated areas should be disinfected in between patients per OSHA guidelines.
6) The procedure should be recorded in detail with a procedure note including: date, pre/post-procedure diagnosis, procedure title, performing physician w/wo assistants, anesthesia, brief indication of procedure, description of graft preparation, description of procedure including guidance and instruments.

Follow-up

1) Patients are generally re-examined 2-6 weeks after the procedure to follow pain, function, injection site and to discuss concerns and future course.
2) Patient response should be recorded using validated outcome measures such Nirschl, VISA, etc.
3) Complications, response and all other pertinent data should be entered in the ICMS tracking system.
4) Consideration for re-injection should be a patient centered decision and made based on functional outcome. We do not endorse a specific number of injections at any site.

***Safety**:*

1) Universal precautions at all times during the procedure and immediately following the procedure.
2) Infection: PRP is antimicrobial and effective against most bacteria classes except Klebsiella, Enterococcus and Pseudomonas. Standard skin disinfection should be used before injection.
3) This is entirely an autologous graft making eliminating the concern for disease transmission unless the graft were contaminated.
4) Risks to patient from the procedure:
    a. Infection
    b. Bleeding
    c. Nerve damage
    d. Pain
    e. Lack of result
    f. Loss of limb and death are very rare but possible.

References:

1. M Ferrari et al. A new technique for hemodilution, preparation of autologous platelet-rich plasma and intraoperative blood salvage in cardiac surgery. *Int J Artif Organs.* 1987 Jan;10(1):47-50.
2. (Mishra and Pavelko. Treatment of chronic elbow tendinosis with buffered platelet-rich plasma. *Am J Sports Med.* 10(10):1–5, 2006
3. Barrett and Erredge. *Podiatry Today*. 17:37–42, 2004
4. Graziani et al. The in vitro effect of different PRP concentrations on osteoblasts and fibroblasts. *Clin Oral Implants Res*. 2006 Apr;17(2):212-9.
5. Tuomi H, Best TM. The inflammatory response: friend or enemy to muscle tissue*? Br J Sports Med.* 2003 Aug;37(4):284-6.
6. Smith C et al. The inflammatory response to skeletal muscle injury: illuminating complexities. *Sports Med*. 2008;38(11):947-69.
7. Tidball JG & Wehling-Henricks M. Macrophages promote muscle membrane repair and muscle fibre growth and regeneration during modified muscle loading in mice in vivo. *J Physiol. 2007.* Jan 1;578(Pt 1):327-36. Epub 2006 Oct 12
8. Dohan et al. Classification of platelet concentrates: from pure platelet-rich plasma (P-PRP) to leucocyte- and platelet-rich fibrin (L-PRF). 2009. *Trends in Biotechnology.* 27(3): 158–67.
9. Kumar et al. Pathologic basis of disease 7th ed. Chapter 2. Copyright 2005. Saunders.
10. Tate KS, Crane D. Platelet rich plasma grafts in musculoskeletal medicine. *Journal of Prolotherapy.* May 2010 2(2):371-376.
11. Almekinders LC, et al. Etiology, diagnosis and treatment of tendonitis: an analysis of the literature. *Med Sci Sports Exer.* 1998 Aug:30 (8):1183-90
12. De Mos M et al. Can platelet rich plasma enhance tendon repair? A Cell culture study. *Am J Sports Med* (2008) 36:1171-8.
13. Aspenberg P, Virchenko, O. Platelet concentrate injection improves Achilles tendon repair in rats. *Acta Othop Scand.* 2004; 75(1):93-99.
14. De Vos RJ et al. Autologous growth factor injections in chronic tendinopathy: a systematic review. *British Medical Bulletin.2010*. Doi:10.1093/bmb/ldq006.
15. De Vos RJ et al. Platelet rich plasma injection for chronic Achilles tendinopathy: a randomized controlled trial. *JAMA.* 2010;303(2):144-149.
16. Murray et al. *J Orthop Res.* 2007 Aug;25(8):1007-17
17. Sanchez M et al. Application of autologous growth factors on skeletal muscle healing. 2nd world congress on regenerative medicine. 2005.
18. Akeda K et al. Platelet rich plasma stimulates porcine articular chondrocyte proliferation and matrix biosynthesis. *Osteoarthritis Cartilage.* 2006;14(12):1272-1280.
19. Kon et al. Platelet rich plasma: intra-articular knee injections produced favorable results on degenerative cartilage lesions. *Knee Surg Sports Traumatol Arthrosc.* 2010 Apr;18(4):472-9
20. Mulvaney, S. Treatment of peripheral nerve entrapments with real time ultrasound guided percutaneous hydro-neurolysis. 2010. Presented at AMSSM annual meeting.
21. Calori et al. Injury. 2008 Dec;39(12):1391-4