Platelet-Rich Plasma Augmentation for Hip Arthroscopy

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Abstract: Biological augmentation and therapeutics are being increasingly used in musculoskeletal and orthopaedic care. Platelet-rich plasma (PRP) is produced from centrifugation of peripheral blood, a process that concentrates platelets within autologous plasma. The process of PRP preparation is fundamental in controlling the contents, and it influences its therapeutic potential. Platelets contain alpha granules that store and release a variety of growth factors and other proteins that may augment the healing environment; PRP also has the added benefit of promoting postsurgical hemostasis. The purpose of this report was to detail our institutional preparation protocol and method of administration of PRP during hip arthroscopy.

Biological augmentation and therapeutics are being increasingly used in musculoskeletal and orthopaedic care.[1-3] Platelet-rich plasma (PRP) has been used by orthopaedic surgeons and other health practitioners to enhance healing and modulate the environment of tendinopathy, surgical repair of tendons, ligament reconstruction, diffuse arthritis, and focal chondral defects.[4-6]

PRP is produced from centrifugation of peripheral blood, a process that concentrates platelets within autologous plasma.[2] Platelets contain alpha granules that store and release a variety of growth factors, such as cytokines and chemokines, as well as other proteins including insulin growth factor 1, transforming growth factor β1, and vascular endothelial growth factor.[1] These biological factors can be modulated by the preparation method used to produce the PRP. For example, leukocyte-rich PRP may increase inflammation and catabolic pathways, whereas leukocyte-poor PRP may decrease inflammation and anabolic pathways.[7,8] In addition, PRP can be prepared as a fibrinous product with adhesive hemostatic properties through endogenous or exogenous activation. The PRP-fibrin preparation can enhance endothelial, epithelial, and epidermal regeneration, by stimulating angiogenesis, improving collagen synthesis, and decreasing scarring.[9]

PRP has been historically described as "a volume of plasma with a platelet count above baseline,"[10] but a more recent quantitative definition requires PRP to contain more than 1 million platelets per milliliter of serum or a 5-fold increase from the baseline platelet concentration.[11] The preparation of PRP is essential in determining the therapeutic potential for biological augmentation. The purpose of this report was to detail our institutional PRP preparation protocol and method of administration during hip arthroscopy.

Procedural Steps

Peripheral Venous Blood Draw

In the preoperative holding room, a temporary elastic tourniquet is used to accentuate the peripheral veins;

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The authors report the following potential conflict of interest or source of funding: T.A.E. receives support from American Board of Medical Specialties (ABMS)—American Board of Orthopaedic Surgeons (ABOS) Visiting Scholars Grant. Patent number 08926626. Mannava S et al. to Wake Forest University Health Sciences. Tissue tensioning devices and related methods. Utility Patent Awarded January 6, 2015. United States Patent and Trademark Office. R.F.L. receives support from Arthrex; Smith & Nephew; Ossur; Health East, Norway; NIH R-13 grant for biologics. Institution provided support by Arthrex, Ossur, Siemens, Smith & Nephew. M.J.P. receives support from ISHA, Smith & Nephew, MIS, Ossur, Siemens, Vail Valley Medical Center, Arthrosurface, DonJoy, Slack, Elsevier, Linvatec, MJP Innovations.

Received November 4, 2016; accepted February 1, 2017.

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2212-6287/161082/$36.00

http://dx.doi.org/10.1016/j.eats.2017.02.001

this is further accomplished by asking the patient to make and release a fist. After preparation of the site with alcohol, peripheral intravenous access is established with insertion of an 18-gauge, 1.5-inch needle. For PRP to be augmented in a unilateral procedure, 60 mL of blood is drawn from a peripheral vein in the arm (Fig 1).

Inactive PRP Preparation

The aforementioned 60 mL of peripheral blood is taken to the laboratory for PRP processing (Fig 2). Thirty-milliliter syringes are taken under a biosafety hood, and 1.0 mL of whole blood is added to a 2.0-mL microcentrifuge tube to conduct automated hematology analysis with the Cell-Dyn 3700 (Abbott Laboratories [Diagnostics Division], Abbott Park, IL). A complete blood count is quantified and recorded using the automated system; this includes platelets; erythrocytes; leukocytes; and 5 differentials including neutrophils, lymphocytes, monocytes, eosinophils, and basophils with counts of 1,000/μL. In addition to the quantification of cells and megakaryocytes, levels of hematocrit (as a percentage) and hemoglobin (in grams per deciliter) are quantified and recorded. PRP is prepared with a commercially available bench-top centrifuge, IEC Centra-CL2 (Thermo IEC, Needham Heights, MD). The blood is transferred into 2 sterile 50-mL conical tubes and placed in the centrifuge (Fig 3). The first centrifugation parameter is set to 2,600 rotations per minute for 10 minutes. After the completion of the first centrifugation, the 2 conical tubes are taken under a biosafety hood for manual extraction of separated blood components. The top fraction of platelet-poor plasma (PPP) is extracted with a pipette and added to a separate 50-mL conical tube (Fig 4). Then, the remaining buffy layer, also known as the *white blood cell layer*, and red blood cell layer are consolidated into one 50-mL conical tube. The second centrifugation is commenced at 3,400 rotations per minute for 6 minutes, followed by the final extraction of the PPP layer. The remaining 5 to 6 mL of the red blood cell and buffy layer is resuspended in the conical tube. Last, 1.0 mL of the PRP product is pipetted into a 2.0-mL microcentrifuge tube to use the hematology analyzer to quantify the final complete blood count (Fig 5). The

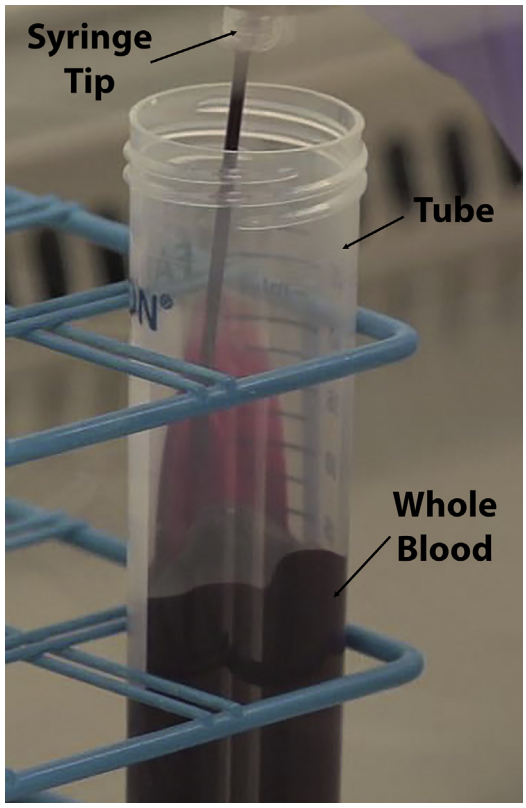

Fig 2. Whole blood from patient placed into centrifuge tube. Care must be taken to not contaminate the sample during transfer from the 60-mL syringe to the centrifuge tube. For platelet-rich plasma to be augmented in a unilateral procedure, 60 mL of blood is drawn from a peripheral vein in the arm.

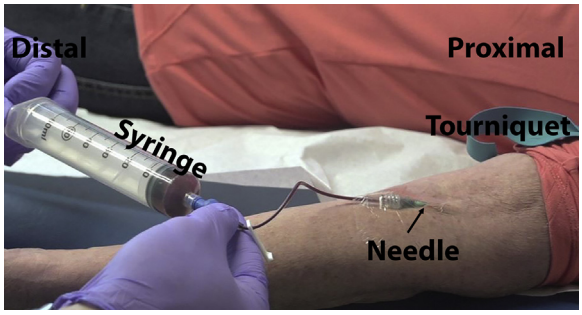

Fig 1. Patient blood drawn from antecubital fossa with 60-mL syringe. A rubber tourniquet should be applied proximal to the antecubital fossa to facilitate vasculature visualization. After preparation of the site with alcohol, peripheral intravenous access is established with insertion of an 18-gauge, 1.5-inch needle.

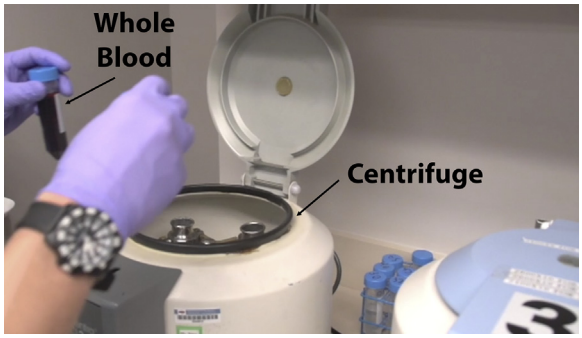

Fig 3. The whole blood sample is taken to the centrifuge and centrifuged for 2 cycles. The first centrifugation is performed at 2,600 rotations per minute for 10 minutes. The second centrifugation is commenced at 3,400 rotations per minute for 6 minutes.

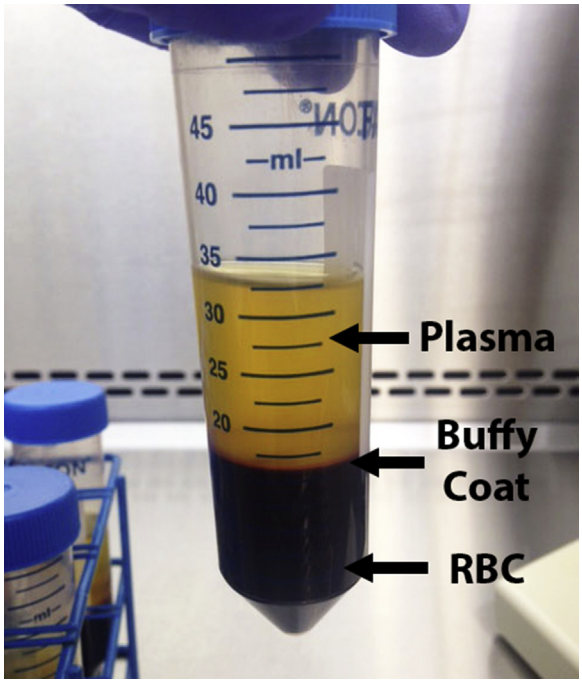

Fig 4. Centrifugation tube and contents after first centrifugation. After the completion of the first centrifugation, the 2 conical tubes are taken under a biosafety hood for manual extraction of separated blood components. The top fraction of platelet-poor plasma is extracted with a pipette and added to a separate 50-mL conical tube. Then, the remaining buffy layer, also known as the *white blood cell layer*, and red blood cell (RBC) layer are consolidated into one 50-mL conical tube.

desired PRP product used for intraoperative augmentation is to have a platelet count greater than or equal to 1,000/μL and an overall leukocyte count less than or equal to 1,000/μL. If the final PRP product is above the desired leukocyte count, the PRP is diluted with a minimal fraction of PPP (Fig 6). The final preparation of diluted PRP is then loaded into a sterile dual-syringe system with the 5 to 6 mL of inactivated PRP.

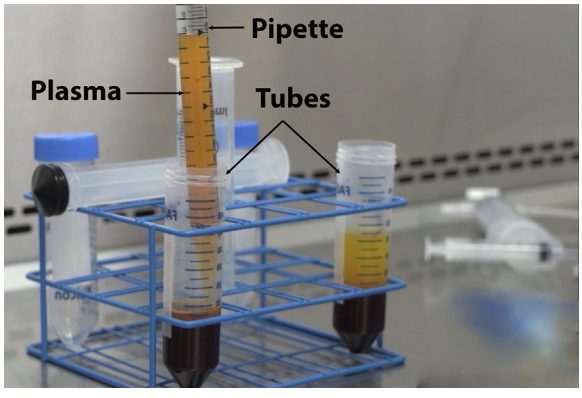

Fig 5. The remaining 5 to 6 mL of the red blood cell and buffy layer is resuspended in the conical tube, and 1.0 mL of the platelet-rich plasma product is pipetted into a 2.0-mL microcentrifuge tube for hematologic analysis.

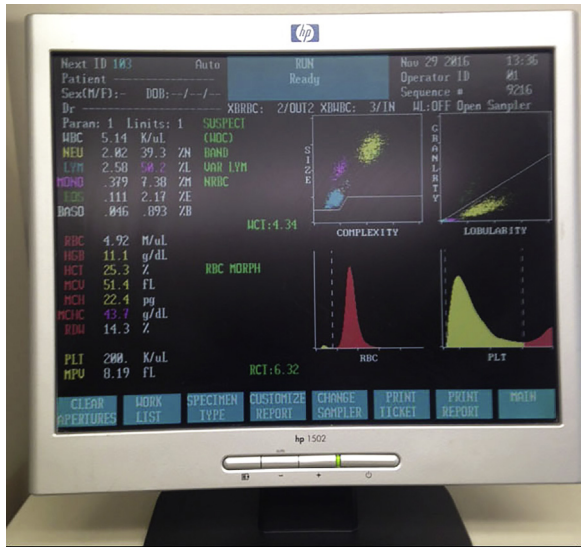

Fig 6. Hematology analyzer displaying the cell counts in the final platelet-rich plasma (PRP) product. If the final PRP product is above the desired leukocyte count, the PRP is diluted with a minimal fraction of platelet-poor plasma. The final preparation of diluted PRP is then loaded into a sterile dual-syringe system with the 5 to 6 mL of inactivated PRP.

Activated PRP Preparation

The previously described method for preparation of PRP can be activated by the following method: After the final centrifugation is completed, 2 increments of 1.5 mL of PPP and 2 increments of 4 mL of diluted PRP are added to two 10-mL Vacutainers (BD, Franklin Lakes, NJ) with 0.3 mL of calcium chloride and set aside for 30 minutes to form a fibrin clot from the platelet-rich plasma releasate (PRPr) supernatant. The PRPr is extracted and loaded into the dual-syringe system. The PRP fibrin clot is set aside to allow for retraction, and then the viscous PRP is loaded into a 60-mL syringe. A summary of both the inactivated and activated PRP preparation techniques is displayed in Table 1.

Hip Arthroscopy With PRP Application

Hip arthroscopy is performed at our institution through 2 portals—an anterolateral (AL) portal and a modified/mid-anterior portal (MAP).[12] Administration of PRP is performed after completion of the procedure, which may include the following components depending on the identified pathology: labral repair, resection of femoral-acetabular impingement lesion (cam and/or pincer), synovectomy, ligamentum teres debridement, labral debridement, acetabular and femoral chondroplasty, and capsular plication (thermal capsulorrhaphy; Smith & Nephew, Andover, MA). Additional procedures that may be performed during hip arthroscopy include debridement of greater trochanteric bursa with iliotibial band release, labral reconstruction, capsulolabral spacer insertion, capsule reconstruction, lysis of adhesions, removal of loose bodies, and iliopsoas release.

Table 1. Summaries Describing Inactivated and Activated PRP Protocols

	Inactive PRP Preparation	Activated PRP Preparation
Pre-processing preparation	Two sterile 30-mL syringes are prefilled with 5 mL of anticoagulant dextrose A.	Three sterile 30-mL syringes are prefilled with 5 mL of anticoagulant dextrose A.
Whole blood collection	60 mL of peripheral venous blood is drawn into two 30-mL syringes from an IV port before general anesthesia administration.	30 mL of peripheral venous blood is drawn into one 30-mL syringe from an IV port before general anesthesia administration.
	The samples are transferred to a separate laboratory for processing.	The sample are transferred to a separate laboratory for processing.
Analysis and processing	1.0 mL of peripheral blood is added to a microcentrifuge tube to quantify and record CBC using a hemoanalyzer.	1.0 mL of peripheral blood is added to a microcentrifuge tube to quantify and record CBC using a hemoanalyzer.
	The first spin is performed at 2,600 RPM for 10 min.	The first spin is performed at 2,600 RPM for 10 min.
	Manual extraction of PPP is performed, and the remaining contents (buffy layer, RBC layer) are spun a second time at 3,600 RPM for 6 min.	Manual extraction of PPP is performed, and the remaining contents (buffy layer, RBC layer) are spun a second time at 3,600 RPM for 6 min.
	A second manual extraction of the PPP layer is performed; the remaining 5-6 mL of the buffy layer and RBC layer is resuspended.	A second manual extraction of the PPP layer is performed; the remaining 2-3 mL of the buffy layer and RBC layer is resuspended.
	1.0 mL of the final PRP product is analyzed by CBC.	Two 1.5-mL increments of PPP and two 4-mL increments of diluted PRP are added to 0.3 mL of $CaCl_2$ (activator); 1.0 mL of the final PRP product is analyzed by CBC.
	PPP is used to dilute the final PRP to produce LP-PRP at a WBC count of 1,000/μL; diluted PRP is then loaded into a dual-syringe system.	PRP fibrin clot is set aside, and viscous PRP solution is loaded into a 60-mL syringe; the dual and 60-mL syringes are then delivered to the operating theater.

$CaCl_2$, calcium chloride; CBC, complete blood count; IV, intravenous; LP-PRP, leukocyte-poor platelet-rich plasma; PPP, platelet-poor plasma; PRP, platelet-rich plasma; RBC, red blood cell; RPM, rotations per minute; WBC, white blood cell.

After completion of the procedures, the central and peripheral compartments are thoroughly lavaged with arthroscopic fluid. The knee is flexed to 45° for relaxation of the capsule. The arthroscope is placed in the MAP, and a disposable cannula is placed in the AL portal. The capsule is closed with No. 2 Vicryl (Ethicon, Somerville, NJ) passed through the capsular leaflets in a double-limb fashion with a suture passer and secured with racking half-hitch knots by use of the "Quebec City slider knot."[13] Usually, 2 or 3 such suture configurations are satisfactory for capsular approximation, without resulting in excessive tightening and constraint. The 2 types of PRP used in hip arthroscopy are outlined in Table 2.

Next, the hip is lavaged with arthroscopic fluid and is then drained. A cannula is placed in an intracapsular manner (between the No. 2 Vicryl capsular repair sutures), and 10 to 15 mL of PRP is injected into the osteoplasty site. This is followed by an injection of Supartz (Bioventus, Durham, NC). The dual-syringe injection (Video 1) of the diluted PRP and PRPr is performed under direct visualization, and approximately 4 mL of diluted PRP and 0.5 mL of PRPr is injected into the repaired hip joint capsule in the peripheral compartment through the arthroscopic cannula with traction removed from the joint. The arthroscopic instruments and cannula are then removed. The portal sites are closed with No. 3-0 nylon suture. The remaining viscous PRP, approximately 10 mL, is then injected into the soft tissues deep to the MAP and AL portal. As the injection is being delivered, the needle is pulled out through the portal tract from deep to superficial, thereby delivering the PRP to the hip arthroscopy portal track. A sterile dressing is then applied. Images depicting the process of PRP injection into the hip are shown in Figure 7.

The advantages and disadvantages of PRP injection during hip arthroscopy are displayed in Table 3. Video 1 presents a complete overview of the application of PRP during hip arthroscopy.

Discussion

The delivery of PRP for various musculoskeletal and non-musculoskeletal uses in medicine has been shown to be safe, with more than 50 years of experience in dermatologic and maxillofacial conditions.[5] The delivery of PRP for various musculoskeletal conditions can be performed simply by a "blind" injection, with localization using radiographs or ultrasound, or under direct

Table 2. PRP Classifications and Biological Profile Description for PRP Preparation in Augmented Hip Arthroscopy

Type of PRP	Biological Description or Activation Mechanism
Leukocyte-poor PRP	<1,000,000/μL of platelets, <1,000/μL of WBC
Leukocyte-rich PRP	>1,000,000/μL of platelets, >1,000/μL of WBC
PRP fibrin clot	$CaCl_2$
PRP releasate	$CaCl_2$

$CaCl_2$, calcium chloride; PRP, platelet-rich plasma; WBC, white blood cell.

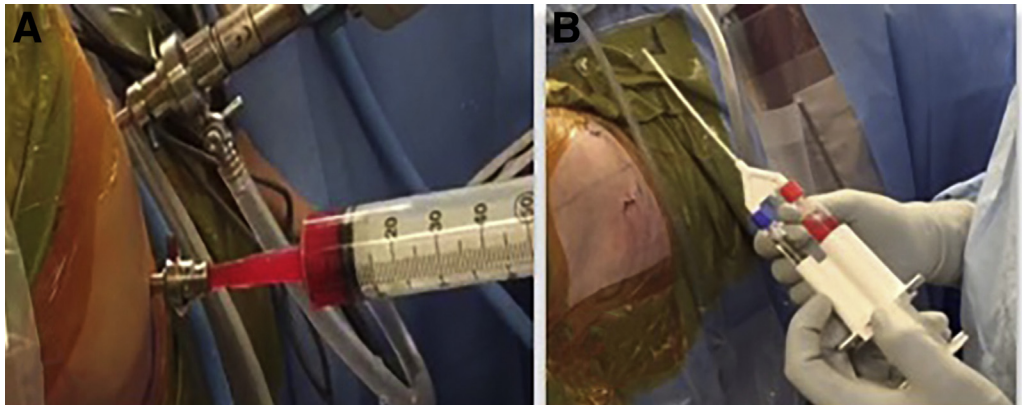

Fig 7. (A) Viscous platelet-rich plasma loaded into 30-mL syringe, being injected into arthroscopic cannula in left hip. (B) Inactivated platelet-rich plasma and platelet-rich plasma releasate loaded into dual-syringe system before injection into left hip.

visualization using fluoroscopy. Hip arthroscopy can be particularly challenging for the delivery of PRP because the joint is held distended with arthroscopic fluid, which can potentially dilute the PRP preparation or cause the delivery to wash away to another location. Furthermore, hip arthroscopy often requires a capsulotomy to gain access to various anatomic locations within the deep and constrained joint, which can also result in the delivered PRP flowing out of the joint.

This article describes our technique for ensuring that the PRP is optimally delivered to the hip joint and portal tracts are created to target the desired anatomic location and maximize the volume of PRP that is administered. More specifically, viscous PRP is only administered after copious lavage and drainage of the hip joint; then, the PRPr and inactivated PRP are administered by use of the dual-syringe system after the hip capsule is partially closed to restrict the PRP from flowing out of the joint.

Other aspects of PRP preparation that often require further clarification are the exact preparation and centrifugation methods. On the basis of the preparation method, the PRP can be customized to take advantage of the many biological properties of the augmentation.[2] For example, leukocyte-poor PRP may induce greater cell anabolism and cell growth, whereas leukocyte-rich PRP may promote catabolic pathways through cytokine induction.[7,8] Some examples of proteins that may be delivered by PRP augmentation include insulin growth factor 1, transforming growth factor β1, vascular endothelial growth factor, hepatocyte growth factor, platelet-derived growth factor, fibroblastic growth factor, platelet factor 4, fibronectin, vitronectin, fibrinogen, and prothrombin.[1] Cytokines and chemokines can be delivered as well.[14] Our institution's method for preparation of PRP for use during hip arthroscopy is provided in this article. It should be noted that in addition to delivery of growth factors, many of the bioactive molecules present in our PRP formulation aid in hemostasis postoperatively. With respect to the described preparation of PRP, it has been reported that leukocyte-rich PRP is effective at initiating a proinflammatory healing response to initiate the regrowth of vascular tissue.[15] However, for the treatment of avascular tissue, this may induce profibrotic tissue formation, which reduces the quality of the tissue.[16,17] In support of this concept, numerous clinical reports have found that leukocyte-poor PRP is most effective for intra-articular treatment.[18-21]

The study of PRP use within orthopaedic surgery applications continues to be investigated. The utility of PRP is

Table 3. Advantages and Disadvantages of Augmenting Activated and Inactivated PRP for Hip Arthroscopy

Inactivated PRP		Activated PRP	
Advantages	Disadvantages	Advantages	Disadvantages
1. Minimal resource consumption during preparation	1. Difficult to deliver to target tissue defect	1. Adhesive to tissues	1. Longer preparation time
2. Minimally invasive delivery	2. Platelet secretion can be disrupted by mechanical or chemical stress	2. Durable construct	2. Invasive application
3. Slow release of platelet-derived growth factors	3. May promote inflammatory response	3. Fast release of platelet-derived growth factors	3. Short growth factor half-life after activation
4. Growth factors are easily dispersed throughout intra-articular regions	4. Potential risk of platelet activation before delivery	4. Gradual excess secretion in vivo	4. Unknown duration of durability to mechanical stresses
		5. Direct application to tissue defect	5. Potential risk of clot retraction

PRP, platelet-rich plasma.

not well known, and investigation is under way to determine the exact mechanisms by which PRP augmentation may aid in healing of orthopaedic maladies. The delivery and localization of PRP to various orthopaedic sites can be challenging, especially during arthroscopy. This article describes our method for PRP preparation and delivery for augmentation of hip arthroscopic surgery.

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