

Using Platelet-Rich Plasma to Reverse the Effects of Tendinopathy and Prevent Tendon Re-rupture after Surgery in Athletes: The Search for a Standardized Protocol

By Shiv Patel

Abstract

30-50% of all lesions amateur and professional sports players will experience during activity are related to the tendon. Moreover, the incidence of tendinopathy, a precursor to tendon rupture, is much higher in both of these groups due to excessive loading of tendons during physical activity, insufficient rest afterwards and certain antibiotic use. The tendon anatomically has both a low blood supply and a low cell turnover rate, which contribute to the relative ease by which an athlete can develop tendinopathy. Chronic tendinopathy has very few high-success treatments but in recent years, platelet-rich plasma (PRP), a treatment in which platelets are isolated from the patient's blood and injected back into the diseased tendon, has seen promising results. Prior research has focused on assessing the viability of PRP as a treatment but failed to come up with a standard and procedure protocol for its administration. In this study, PRP is evaluated in terms of success rate, concentration of cells other than platelets, concentration of growth factors, life of growth factors, and size and cross section of the tendon to develop a formulation standard, injection plan, and procedure protocol for different tendinopathies. Furthermore, a rehabilitation program that takes into account both the treatment and natural healing process of the tendon to shorten the time the athlete spends off the field is outlined.

Introduction

Tendinopathy, a precursor to tendon rupture, "is used to describe the clinical picture of pain and swelling of a tendon that exhibits no inflammation" (Andia, Sanchez, and Maffulli 1420). The symptoms of tendinopathy manifest when the rate of healthy tenocyte proliferation and extracellular matrix (ECM) formation is surpassed by the rate of tissue degeneration. Because of its prevalence among athletes and non-athletes alike, tendinopathy is widely studied with the leading research focused on cell apoptosis, angiogenesis that has escaped cell control, or inflammation and pain, according to Andia, Sanchez, and Maffulli (1420). Despite these efforts, not many treatments exist for tendinopathy.

In recent years, however, a new therapy by the name of platelet-rich plasma (PRP) has demonstrated promising results for treating tendinopathy and preventing rerupture in tendons after surgical repair. PRP is, in simple terms, a therapy in which platelets are isolated from the patient's blood and injected back into the affected tendon. The platelets release growth factors, which can quicken the healing process and promote organized growth of structural proteins such as collagen. Prior research has focused on assessing the viability of PRP as a treatment, but all have failed to come up with a standardized procedure protocol for its administration.

In this study, PRP is evaluated in terms of success rate, concentration of platelets, concentration of cells other than platelets, life of growth factors, and size and cross section of the tendon to develop a formulation standard, injection plan, and procedure protocol for treating different tendinous lesions and tendinopathy. Furthermore, a rehabilitation program that takes into account both the treatment and natural healing process of the tendon to shorten the time the athlete spends off the court or field is outlined.

Especially in athletes, tendinopathy, if not properly treated, can progress into tendon rupture. Joseph et al. in “Achilles Tendon Biomechanics in Response to Acute Intense Exercise” contend that the most common tendon ruptured is the Achilles, attributed to the extreme direct and repeated forces and intensive flexing that it experiences during activity, experiencing loads up to 12 times the body weight (11). Sports cannot change to utilize the Achilles less, so a preventative treatment needs to be employed to protect athletes. According to various studies, including those conducted by Rosengarten et al. and Amin et al., normal tendon structure can be lost just after 2 days of intense exercise, showing that tendon rupture among athletes is a very real risk (Figure 1).

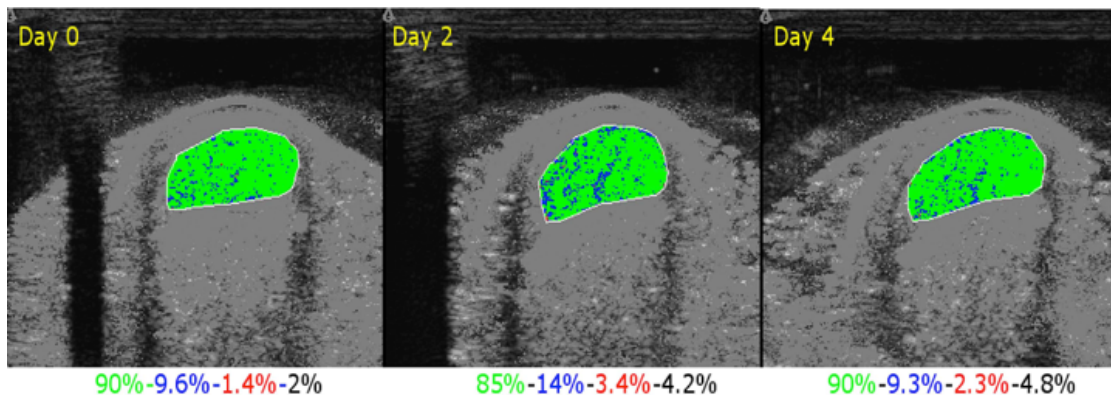


Figure 1. Image of an ultrasound tissue characterization of the Achilles tendon showing structural changes due to loading over a period of 4 days. Day 0: Prior to exercise; Days 2 & 4: After exercise. Green regions show uniform, intact tendon fibers and blue regions show less uniform, broken/wavy tendon fibers (Rosengarten et al. 185).

Combined with daily practices and long seasons, transient loss of tendon structure in athletes can cross the point of no homeostatic return according to Dr. Samuel Rosengarten and his co-authors in “Australian Football Players’ Achilles Tendons Respond to Game Loads Within 2 Days: An Ultrasound Tissue Characterization (UTC) Study” (185). Due to the nature of sporting, this can lead to permanent damage of the tendon and a decreased sporting ability for the athlete. Constant loading and unloading of the tendon makes it more malleable, reducing its cross-section. As there is less area for the tendon to absorb impact and more load per square inch, the tendon damage is more likely during physical activity. Rosengarten et al., in accordance with Dr. Christophe Charouset et al., assert that the tendon is a connective tissue that responds to mechanical loading in the short-term, defined as 24-72 hours, and the long-term, defined as 12 weeks-years. Rosengarten et al. also believe that that intense exercise results in changes of catabolic and anabolic activity within the tenocytes that tend to normalize after 4 days or longer and that if enough time is not given in between games and/or practices, these microtrauma can transform into disorganization of the collagen and ECM of the tendon (183). In all professional sports there is a rigorous practice schedule, and in some, such as baseball and basketball, athletes have to play back-to-back games. This does not allow the athlete enough time to rest tendons and give them the 4 days required to normalize the structure.

The tenocyte, the cell tasked with restoring the tendon ECM, is prompted by mechanical stimuli to begin reconstruction. Furthermore, the overloading of the tendon prompts the tenocyte to upregulate large proteoglycan expression, leading to an “increase in bound water within the ground substance, which may lead to matrix disorganization” (Rosengarten et al. 186). Thus, the tenocyte will repair the tendon to baseline strength during normal impact and use, but hyperloading will disrupt the structure of the tendon and make it much more difficult

for the tenocyte to repair it. Andia, Sanchez, and Maffulli add that the tenocyte responds to continual mechanical loading with the release of cytokines IL-1 β and tumor necrosis factor- α (TNF- α), prostaglandin E2 (PGE2), and neuropeptides P, and connective tissue growth factor (CTGF), all of which are pro-inflammatory agents (1422). Fox et al. agree, discussing that tendinopathy is correlated with an increase in inflammatory cytokines TNF- α and IL-1 β (2855). PRP is usually prepared free with few leukocytes and other inflammatory cells and many its growth factors. When injected into the tendon, the growth factors overtake the inflammatory cytokines already present in the injured tendon in concentration levels and promote faster healing.

Most sports require frequent extension of one or more tendons, leading to tendinopathy and in some cases, tendon rupture. Hyperloading of the tendon in a short bout of physical activity causes the tendon to experience microtrauma, weakening its overall structure, Amin et al. write in “Performance Outcomes after Repair of Complete Achilles Tendon Ruptures in National Basketball Association Players” (1868). Charousset et al. concur, finding that tendinopathy is a result of exercise-induced microruptures within the tendon. Professional athletes cannot avoid exercise, as it is the nature of their career so a proper preventative treatment needs to be developed. Charousset et al. also attribute tendinopathy to “microruptures of the patellar tendon, incomplete healing, and extensive neovascularity that may lead to a chronic degenerative process” (907). Over time, microruptures can build up and ultimately cause a rupture in one or more tendons. If PRP is administered to the tendons that the athlete loads and unloads the most, the microruptures within those tendons can be healed faster, which allows the athlete to play back-to-back games and conform to intense practice schedules without fear of long-term tendon damage. PRP injections at the first signs of tendinopathy can introduce vital healing growth factors such as PDGF, which promotes type I collagen growth, and vascular endothelial growth factor (VEGF) and platelet derived angiogenesis factor (PDAF), which increase blood flow to the inside of the tendon according to the review of research conducted by Andia, Sanchez and Maffulli. Collagen is important to tendon healing and described by Mark D. Rekhter as “proteins that consist of a triple helix of polypeptide chains and globular domains,” in his article “Collagen Synthesis in Atherosclerosis: Too Much and Not Enough” (376). The tough buildup of collagen insures protection from mechanical stress and impact from sports-related activities.

Tendinopathy is extremely prevalent in sports as indicated in numbers by Charousset et al., which is why a high-success treatment such as PRP needs to be perfected. PRP treatment may not only save the careers of athletes in sports with high incidences of tendinopathy, such as volleyball and basketball, but also promote more competition by retaining elite athletes for a longer period of time. In the study by Amin et al., it is shown that even with PRP treatment, an athlete’s game time is reduced after an Achilles tendon rupture, but specific performance factors such as rebounds, steals, and blocks normalized when the time played was used as a reference frame against the control instead of a per-game reference frame (1865). This data shows that although PRP may not allow an athlete to fully regain their stamina, it will enable him or her to retain their capabilities as an athlete.

Platelet-Rich Plasma as a Treatment

According to Sánchez et al., tendon healing is a complex process that starts with the secretion of cytokines from broken alpha granules in platelets, which signal cells within the tendinous tissue to proliferate and construct the ECM (246). Despite the many growth factors that aid tendon healing, the tendon healing process is a slow one due to its insufficient blood supply

and low rates of cell division (Sánchez et al. 245). Blood supply to an injury site is vital as the circulatory system is the path through which platelets, growth factors, and other reparative molecules are delivered. The healing process of the tendon, according to Sánchez et al. “involves several stages, including angiogenesis, cell proliferation, and the deposition of extracellular matrix. These stages are followed by remodeling and maturation, during which the healing tendon should ultimately regain its mechanical strength” (245). Andia, Sanchez, and Maffulli disagree slightly and propose that the three stages to the healing process of tendons after trauma are, in order, “inflammation, angiogenesis, and proliferation” (1418). Although the healing process of the tendon is not completely agreed upon, the important fact is that the tendon requires a continuous blood supply, growth factors to signal proliferation, and growth factors to signal the secretion of a well-structured ECM to heal itself.

The plethora of growth factors and cytokines secreted by platelets help spur cell growth and promote quick repair of the damaged region. Platelets are produced by bone marrow cells called megakaryocytes, that make up approximately $1.4 - 4 \times 10^{11}$ cells/liter of the blood, and circulate for approximately 10 days according to Abrams and Dugdale. Platelets are short-lived, so once PRP is prepared, it needs to be injected immediately. Andia, Sanchez, and Maffulli further discuss that growth factors such as TGF- β , PRGF-BB, and angiopoietin-1 secreted by the platelets aid in stabilizing blood vessels, which move along the healing process (1417). Stabilizing blood vessels is of paramount importance, as unstable blood vessels can do more harm than good and cause uncontrolled growth of inflammatory tissue.

The mechanism through which PRP works is simple: it provides an abundance of platelets that can secrete growth factors through alpha granules to synthesize collagen and promote organized angiogenesis (Charousset et al. 909, Sánchez et al. 246). PRP exploits the natural healing ability of platelets, providing a safe auto-derived treatment with minimal side effects. Hom-Lay Wang and Gustavo Avila simply describe PRP as “essentially an increased concentration of autologous platelets suspended in a small amount of plasma after centrifugation” (192). Centrifuging whole bloods draws platelets out of the blood and separates it from other components. PRP also includes structural proteins including fibrin, fibronectin, and vitronectin that help rebuild the tendon by “facilitat[ing] cell adhesion by forming three-dimensional scaffolds” (Andia, Sanchez, and Maffulli 1418). The fibrin matrix aids in building the scaffolding complex, a precursor to tenocyte proliferation (Sanchez et al. 246). If the fibrin matrix is synthesized outside of the cell, and injected, there would be a higher concentration of growth factors that are able to contribute to the healing process. Also, the fibrin matrix seems to be stimulating the growth of collagen I, the most uniform and structurally sound form of collagen, which means a faster and better overall recovery.

PRP is a viable treatment option for tendinopathy and tendon rupture (Figure 2), as it introduces platelets to the tendon, which stimulate the production of collagen type I. Sánchez et al. also concur adding that PRP contains “TGF- β 1, PRGF [PRP], VEGF, epithelial growth factor (EGF), hepatocyte growth factor (HGF), and insulin-like growth factor (IGF-I)” (246). These molecules produce vital signals that stimulate the synthesis of structural proteins. Furthermore, Andia, Sanchez, and Maffulli reason that the expression of genes IGF-I and TGF- β 1 are important to the healing process of tendons and control the type of collagen being synthesized and that platelets modulate inflammation by controlling a plethora of chemokines that, in turn, control the movement of leukocytes and monocytes at the injury site (1418). This creates a positive feedback loop in which growth factors flood the cells, drive out inflammatory cytokines (which upregulates growth factors), and so on.

Healthy vascularization is important in tendons so that growth factors can be circulated to the damaged area. Sánchez et al. report that the production of VEGF and HGF in tenocytes

helps vascularize the tendon (246). This gives the injured tendon the blood supply that it needs to heal quickly.

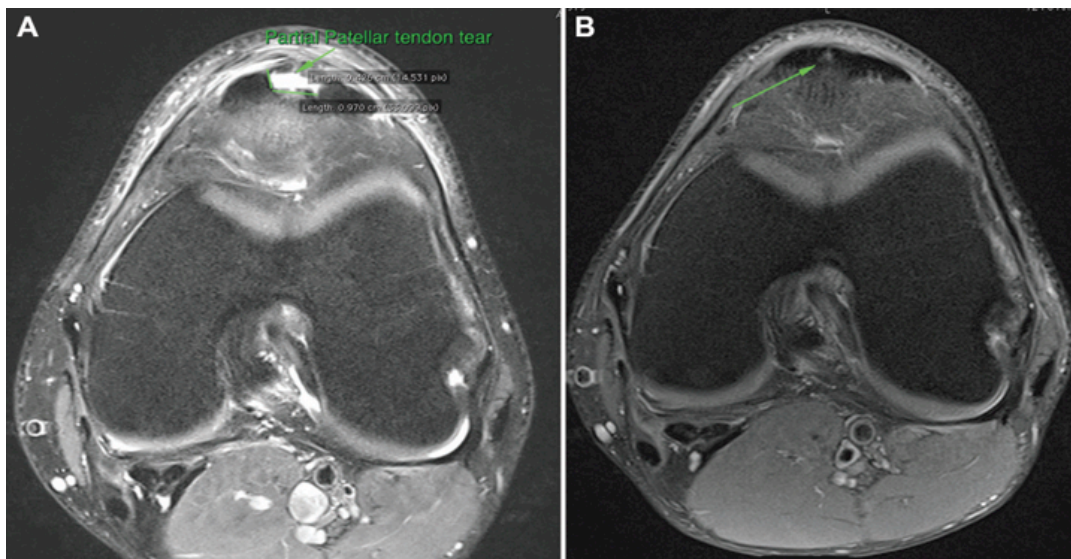


Figure 2. A) MRI of partial patellar tendon rupture before PRP treatment B) MRI of patellar tendon 3 months after 3 PRP injections (Charousset et al. 907)

Role of Fluoroquinolones in Tendon Rupture

Fox et al. in “Fluoroquinolones Impair Tendon Healing in a Rat Rotator Cuff Repair Model: A Preliminary Study” assert that there has been a recent trend in fluoroquinolone (FQ)-related tendon ruptures. The rate of incidence of FQ-induced tendon rupture is low 0.14-0.4% (it is much higher in transplant patients at 12.2-15.6%), Fox et al. claim, but has far-reaching consequences due to FQ’s status as the most prescribed family of antibiotics in the United States in 2011 according to Mercola.com (Fox et al. 2852). The side effects of FQ antibiotics may be especially stark for the transplant patients because they are already in a weakened state and there are few growth factors present in the tendons. Increasing the amount of growth factors in circulation may lower the chances for tendon rupture in transplant patients even without PRP treatment.

Due to their effectiveness in eliminating both gram-positive and gram-negative bacteria, FQs are the best treatment currently for treating tough bacterial infections. Simonin et al. argue that FQs are not dangerous most of the time to most of the population, generally speaking (869). However, if a professional athlete acquires an infection that requires the use of FQ antibiotics, they may experience tendon rupture due to extensive use. Constant loading of the tendon after FQ has already mutated tenocytes within it may cause the tendon to overstress and rupture. Thus, it is important to administer PRP to high-impact tendons such as the Achilles in athletes to help reverse the side effects of FQs and serve as a layer of protection while they are taking the antibiotics.

It is not fully known how FQs affect the body, but Simonin et al. suggest FQ antibiotics’ high tissue penetration to be a possible reason for inducing tendon rupture (867). This allows the antibiotics to cause mutations in tenocytes for a longer period of time. Structural proteins are important, as their name implies, in maintaining tendon conformity, but FQs reduce their expression in the extracellular matrix, including those of “type I collagen, elastin, fibronectin, and b1 integrin” and also “reduced mitochondrial activity, [caused] direct toxicity on collagen, elevated levels of activated caspase 3 (an apoptosis marker), and non-cytotoxic inhibition of canine tendon cell proliferation” (Fox et al. 2858). Combined with their high tissue penetration

and their ability to induce apoptosis, FQs can wreak havoc inside the tendon and disrupt tendon structure.

In addition to repressing and activating certain genes, FQs cause an influx in reactive oxygen species (ROS) in cells, resulting in a decrease of type I collagen synthesis, according to Baboldashti et al. (1935). Baboldashti et al. further explain that the ROS decrease collagen synthesis because they activate the transcription factor FOXO1, which is a repressor for type I collagen synthesis (1935). Type I collagen is important in maintaining tendon structure and if the FQ is targeting it, this can be the cause of tendon rupture in patients treated with FQ. The oxidative changes induced by pefloxacin manifested a few days after treatment, supporting the findings of other studies. FQ seems to be deeply penetrating tissues and affecting them slowly. For example, Simonin et al. add that the ROS modulation of collagen synthesis was seen at least 5 days after FQ administration (871). In the study conducted by Fox et al., the tendons of the test subjects treated with FQs failed structurally.

Fox et al. discovered that the FQs inflicted microtrauma upon the healing enthesis, weakening the entire tendon (2858). This demonstrates that FQ exposure directly affects the weakening of tendon structure. Less fiber at the healing enthesis not only means compromised structural integrity but also a lowered ability to heal in the long term.

Similarly, structurally weak collagen synthesis lowers the ability of the tendon to securely bind to the bone (Joseph et al. 1181). With less calcified fibrocartilage to shield the bone, the tendon has a higher chance of rupturing. For this reason, the body goes into a “fibroproliferative response, presumably in an attempt to repair the tendon defect resulting from the depletion phase” (Simonin et al. 870). The presence of a natural repair mechanism indicates that the body is trying to reverse the adverse effects of the FQ but is not necessarily effective in doing so. As shown in the study by Rosengarten et al. and Fox et al., the body may be promoting the growth of collagen types that reduce the structural integrity of the tendon. Also, the tendon fibers may not be growing uniformly, resulting in disorganization and again, a compromised structure. PRP injections can be given at the healing enthesis and spread from there to mitigate the chance of rupture. It also means that tendon integrity at the organizational level has been damaged in some way to yield these results.

FQs are known to promote the growth of collagens types II and III, according to Rosengarten et al., instead of the ones useful to tendon structure (collagen type I), causing a lapse in integrity (185). FQ-related tendinopathy has been correlated with a downregulation of tissue inhibitor of metalloproteinase (TIMP-2), an effect known to be associated with tendinopathy, and disorganization of collagen and fibrocartilage (Fox et al. 2855). TIMP is an especially important regulatory factor and, if not balanced correctly, can cause tendinopathy and other disorders of the tendon. TIMP expression, and thus collagen type II and III synthesis, can be reduced with the introduction of inhibitory molecules and flooding the system with collagen type I growth factor PDGF (Wang and Avila 192).

Finally, FQs are linked to gene expression in some way because of their effect on the production and inhibition of proteins and molecules. FQs upregulate factors that mark cells for apoptosis and prevent expression of those that are important to cellular structure. The ECM is extremely important to tendon structure and FQs are altering the concentration of apoptosis markers and other chemicals within the matrix. This imbalance is most likely the reason for tendon disorganization.

Application of Platelet-Rich Plasma in Sports

Andia, Sanchez, and Maffulli draw attention to the prevalence of tendon injuries, stating that “an estimated 30-50% of all sports lesions are painful tendon injuries that affect profes-

sional and recreational athletes in various anatomical locations” (1415). Christophe Charousset et al. break this statistic down, claiming that 40% of volleyball and 35% of basketball players are affected by tendinopathy (906). Rutland et al. concur, contending that, “overuse tendon injuries account for 7% of the injuries seen in United States physician offices and 40% of knee injuries in volleyball players” (166). The extremely high number of tendinopathy cases also shows a need for an effective preventative treatment to prevent tendon rupture. The rates of tendinopathy and tendon rupture are high, in part, because of the body’s inability to heal tendons effectively and because of the few treatment options available. Sánchez et al. in “Comparison of Surgically Repaired Achilles Tendon Tears Using Platelet-Rich Fibrin Matrices,” further argue that if tendinopathy progresses into tendon rupture, it would be especially devastating because the recovery time after operation is lengthy (246). These types of disorders can even drive athletes to abandon their careers and lead painful lives long after their sports careers are over.

Platelet-rich plasma is widely accepted as a treatment and is approved for use in sports. The World Anti-Doping Agency states on their website, “despite the presence of some growth factors, platelet-derived preparations were removed from the List as current studies on PRP do not demonstrate any potential for performance enhancement beyond a potential therapeutic effect.” Team physicians are afraid to approve this type of treatment for players, but science and those who have used PRP stand by it. It is so effective that the World Anti-Doping Agency had to make it clear that it is not an enhancement. Mikel Sánchez et al. in “Comparison of Surgically Repaired Achilles Tendon Tears Using Platelet-Rich Fibrin Matrices,” imply an agreement with the Agency’s ruling, claiming that PRP is the “safe strategy to accelerate tendon cell proliferation, stimulate the synthesis of type I collagen, and promote neovascularization both in vivo and in vitro” (246). PRP is an extremely effective treatment and reduces the time needed for an athlete to regain range of motion and to return to sporting (Sánchez et al. 250). Amin et al. agree and claim that although “professional athletes have essentially limitless means at their disposal to ensure an optimal recovery after an injury... only 60% to 70% can return to their sport, usually at a measurably lower level of performance” (1868). PRP is a relatively expensive treatment at its current stage, but professional sports teams have a plethora of financial resources to support the players. Furthermore, the cost of losing a player outweighs the cost of the PRP treatment.

Research both in vivo and in vitro has demonstrated the effectiveness of PRP, even if the results are not miraculous or a “cure-all” treatment. Especially for an injury such as an Achilles tendon rupture, coaches and team physicians should recommend PRP injections concurrent with and after surgery, as steroid injections and other treatments have not been very successful in the past.

Andia, Sanchez, and Maffulli claim that studies have indicated that PRP increases the blood supply and the amount of circulation-derived cells to the injured tendon (1418). Moreover, Baksh et al. assert in “Platelet-Rich Plasma in Tendon Models: A Systematic Review of Basic Science Literature” that most studies indicate higher vascularization of the injured tendon and increased tendon strength after PRP treatment. PRP has only shown positive results, minimal (if any) side effects, and no deterioration from the initial state. Even if the PRP treatment “fails,” the player can take comfort in the fact that they will not experience any rejection of or negative reaction to the treatment, as it is derived from their own body.

Charousset et al. claim that 75% of athletes who did not respond to non-operative treatments for tendinopathy for at least 4 months in their study were able to return to their pre-symptom sporting level after PRP treatment (909). This comes in contrast with operative measures, which only allow 50-70% of treated athletes to return to their pre-symptom sporting levels (Charousset et al. 909). These are almost indisputable numbers, and team physicians have

a responsibility to inform players about PRP therapy because they have a right to know what their options are.

In “Performance Outcomes after Repair of Complete Achilles Tendon Ruptures in National Basketball Association Players,” Nirav Amin et al. found that after Achilles tendon ruptures, not only did the NBA players have reduced performance in those activities that heavily relied on the tendon such as rebounds, steals and blocks, but they also had reduced abilities in every other component of the game (1869). Amin et al. also found that 36% of NFL players who had surgically repaired Achilles tendons and 39% of NBA players (sample size: 18) were unable to return to the game (1868). Parekh et al. in “Epidemiology and Outcomes of Achilles Tendon Ruptures in the National Football League” similarly found that 36% of NFL players who experienced Achilles tendon ruptures did not return to the game and add that those who did had over a 50% reduction in performance (285). If PRP is concurrently administered with surgical repair of the tendon, and this practice is standardized, then these numbers would be much lower. This assertion is corroborated in the study conducted by Sánchez et al., who reported that “two of the amateur athletes in the [PRP] group (soccer and basketball) attained preinjury level by 6 months, whereas the 2 remaining athletes (soccer) retired from competitive sports for reasons other than the injury; however, their level of activity was high” (249).

Surgical repair of Achilles tendon ruptures is usually the best option for professional athletes so that they can return to the game, but it is not enough for athletes to stay in the game. Andia, Sanchez, and Maffulli report that PRP treatment is also not without risk, as it can decrease the cross-sectional area of a tendon after 18 months (1419).

Dr. Nasim Baboldashti, a researcher at the University of Oxford, and his colleagues suggest that PRP can also be used as a “protective strategy at the time of local drug injection to prevent some of the adverse side effects of steroids and fluoroquinolones” (1931). Steroids have been shown not to be effective treatments against tendinopathy like in the experiment conducted by Baum et al., but PRP looks hopeful as a preventative treatment for tendon rupture when administering fluoroquinolones.

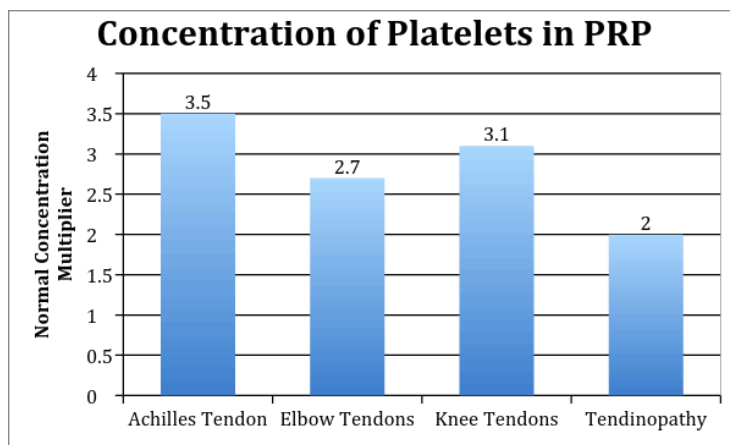


Figure 3. Chart detailing suggested concentrations of platelets in PRP formulations for recommended protocol in different tendons/tendinopathy.

dendon rupture (used concurrently with surgery; Figure 3).

PRP is not only effective because of the molecules it contains but also because of those it does not. Charoussat et al. assert that in many studies such as McCarrel and Fortier’s, it was “demonstrated that leukocyte concentration is positively correlated with catabolic gene

Recommendations on the Use of PRP

There are approximately $1.4 - 4.0 \times 10^{11}$ platelets/liter of blood, and each platelet has a lifespan of ~10 days (Woodell-May et al. 749). According to the review conducted by Baksh et al., formulations of PRP were dependent on the individual and varied from 2 – 7 times the normal platelet concentration. In this study, these concentrations and their effectiveness were taken into account and matched with types of tendinopathy/tendon

expression in tendons and ligaments, which suggested that delivery of concentrated leukocytes to a site of injury might not provide a favorable environment for tissue healing” (909). Introducing leukocytes to the tendon during its healing process is not favorable, as the leukocytes are attacking the tendon tissues in the absence of pathogens and causing sterile inflammation. Creating an environment with minimal leukocytes may promote better and faster healing, as it prevents the onset of sterile inflammation. However, it may put the patient at risk for infection if they have an already weakened immune system. For this reason, few white blood cells should be included in the formulation. Fontenot et al. agree with this assertion to an extent, claiming that white blood cells aid in tissue repair by warding off microbes and maintaining an immune system presence in the injured area (1270).

Commercial preparation of PRP is very efficient and the best option in the long-term. Although the upfront cost is significantly greater than other methods, the efficiency, reduction in chance for human error, and simple processing of whole blood make it suitable for a clinical setting. Charousset et al. used the Arthrex ACP system to isolate platelet-rich plasma from whole blood. The ACP contained more than two times the density of platelets than whole blood (907). The preparation process is simple according to Charousset et al. who drew 15 mL of venous blood “into the Arthrex Double Syringe System (Arthrex Inc.) and then centrifuged at 1700 rpm for 5 minutes. A final volume of 6 mL of pure PRP was obtained” (907).

The most effective concentration of platelets in the PRP in relation to whole blood is debated and varies from 2-10 times. However, as Fontenot et al. point out, “some studies suggest that high platelet numbers are no better than moderate numbers or may actually have detrimental effects” (1267). It is better to be on the safe side and inject a lower concentration of platelets and then administer more injections than to do the opposite. After reviewing the literature and comparing different tendon densities, tensile strengths, and size, a guide to concentration levels was developed (Figure 3).

PRP can be harmful or useless without the correct formulation, so balancing how much PRP injected is of paramount importance. The current protocol devised by Dr. Peter Wehling, according to the Chicago Tribune, is to extract PRP serum from ~60mL of blood to and inject it into the patient’s tendons six times over the treatment course. This treatment can be used as a guide but needs to be tailored according to tendon density and size to be most effective. A blanket treatment may even do more harm than good if too much tissue is regenerated according to the mechanism described by Wang and Avila in “Platelet Rich Plasma: Myth or Reality?” Andia, Sanchez, and Maffulli suggest that injecting patients weekly with PRP over 2-3 weeks using ultrasonography as a guidance tool is an effective way to administer the treatment (1420). Charousset et al. agree with this procedure protocol, adding that a 16-MHz probe gave a clear image of the internals and that a 22-gauge needle was used to inject the PRP “within and around the hypoechoic area” (907). Ultrasound imaging is optional but is recommended as it allows the physician to accurately guide the injection. This allows for a more effective treatment and fewer injections over the course of the treatment. Charousset et al. add “75% [of the treated patients] were able to return to their presymptom sporting level ... after a mean period of 3 months, and this sporting level was maintained until the 2-year follow-up” (909). The 3-month MRI in 57% of patients confirmed a complete repair of the tendon. This conservative procedure is an alternative treatment to surgery, which has allowed only 50% to 70% of the treated patients (either arthroscopic or open surgery) to return to a presymptom sporting level” (Charousset et al. 909).

Even though the Arthrex Inc. preparation system mentioned previously yields high-grade PRP, the delivery method needs to be refined. PRP needs to be injected in and around the tendon uniformly, presenting a challenge as one cannot just move up the tendon, injecting PRP at

regular intervals on the skin. A possible solution is using the same technique used for lubricating joints with Orthovisc (high molecular weight hyaluronan) fluid—moving the needle while it is inside the patient. This may cause the hypodermic needle to stretch the entrance hole, but this is better than creating multiple injection sites.

PRP alone cannot miraculously cure tendinopathy and fix tendon rupture, but instead, it has to be combined with a variety of other treatments to be most effective. One of these is physical therapy (PT). Stanish, Rubinovich, and Curwin suggest that the PT program consists of stretching, progressive loading, and increase in the speed of exercise (66). The stretching helps with stratifying the tendon and pain from joint movement. The load on the tendon should be increased weekly and lessened if the patient experiences pain. Speeding up the contractions allows the tendon to take on more force according to Stanish, Rubinovich, and Curwin (66). The exercise program should last for at least 4 weeks but 6 weeks is recommended to ensure complete healing and to minimize the risk of rerupture. Charousset et al. suggest that running should be taken up after 6 weeks and sporting after 8 weeks (908).

Many studies lay claim to the ineffectiveness and potential harmful effects of PRP, yet they fail to provide concrete data to corroborate these assertions. Studies have also erred on the side of caution, arguing that more research is needed before PRP can be administered to patients, as it may present extremely negative side effects in the long term. Although these claims might be well warranted under normal circumstances in testing of synthetic drugs, PRP is derived from the patients' bodies and unused portions of it is metabolized soon after it is administered. For this reason, any side effects will manifest in the short term, and no significant ones have been documented. Of course, administering too much of any substance that interferes with cell signaling can be harmful in large quantities, which is why in this study, the formulation standard lies more on the conservative side. Athletes should not be over-vigilant when it comes to making the decision of whether to try PRP or not. If alternate treatments continue to fail, they should consider PRP as a serious option, as it has displayed extremely positive results in the past. In the case that the treatment is successful, athletes will not only enjoy the benefit of a fast and more complete recovery from tendon rupture and tendinopathy but will also be providing more data on which scientists can rely and perfect PRP technology for widespread use.

PRP is a promising treatment for alleviating tendinopathy and preventing tendon re-rupture postoperatively. Although the treatment has been developed extensively in the past few years, a standard for its administration has not been set. Gathering recommendations from previous studies and taking into account the different tendons of the body, a comprehensive protocol was developed in this study (Table 1). Figure 3 and Table 1 should serve as a guide for administering PRP to athletes after tendinopathy or rupture in the tendons outlined. Ultimately, however, the provider must make the final decision on the formulation, the injection volume, and the exercise program until there is more research on the administration of PRP.

Table 1. Procedure for Administering Platelet-Rich Plasma	
1	Establish a baseline platelet count
2	Draw 15 mL of whole blood
3	Centrifuge at speed recommended by the Arthrex ACP Double Syringe System for 5 minutes
4	Isolate platelets and check concentration, matching it to Figure 3; do not exceed an injection volume of 2.5 mL
5	Inject PRP in and around the tendinous lesion (optional: use ultrasonography to guide the injection)
6	Repeat procedure once a week after the first injection for 2 more weeks, for a total of 3 injections
7	Enroll patient in a therapy program focused on stretching, light extensions of the tendon, and mild exercise; increase difficulty on a weekly basis

Works Cited

- Abrams, C. S., David C. Dugdale, III, and B. J. Bain. "Platelet Count: MedlinePlus Medical Encyclopedia." *MedlinePlus*. U.S. National Library of Medicine, 2 Feb. 2013. Web.
- Amin, Nirav H., Andrew B. Old, Loni P. Tabb, Rohit Garg, Nader Toossi, and Douglas L. Cerynik. "Performance Outcomes after Repair of Complete Achilles Tendon Ruptures in National Basketball Association Players." *The American Journal of Sports Medicine*. 41.8 (2013): 1864-1869. Web.
- Andia, Isabel, Mikel Sanchez, and Nicola Maffulli. "Tendon Healing and Platelet-Rich Plasma Therapies." *Expert Opinion on Biological Therapy*. 10.10 (2010): 1415-1426. Web.
- Baboldashti, Nasim, Raewyn Poulsen, Sarah Franklin, Mark Thompson, and Philippa Hulley. "Platelet-Rich Plasma Protects Tenocytes from Adverse Side Effects of Dexamethasone and Ciprofloxacin." *The American Journal of Sports Medicine*. 39.9 (2011): 1929-1935. Web.
- Baksh, Nikolas, et al. "Platelet-Rich Plasma in Tendon Models: A Systematic Review of Basic Science Literature." *Arthroscopy: The Journal of Arthroscopic & Related Surgery*. 29.3 (2013): 596-607. Web.
- Baum, H. S. Böttcher, R. Abel, H. J. Gerner, and H. G. Sonntag. "Tissue and Serum Concentrations of Levofloxacin in Orthopaedic Patients." *International Journal of Antimicrobial Agents*. 18 (2001): 335-400. Web.
- Charoussat, Christophe, Amine Zaoui, Laurence Bellaiche, and Benjamin Bouyer. "Are Multiple Platelet-Rich Plasma Injections Useful for Treatment of Chronic Patellar Tendinopathy in Athletes?" *American Journal of Sports Medicine*. 42.4 (2010): 906-911. Web.
- Fontenot, Robin L., et al. "Simple Tube Centrifugation for Processing Platelet-Rich Plasma in the Horse." *The Canadian Veterinary Journal* 53.12 (2012): 1266-1272. Print.
- Fox, Alice J. S., et al. "Fluoroquinolones Impair Tendon Healing in a Rat Rotator Cuff Repair Model: A Preliminary Study." *The American Journal of Sports Medicine*. 42.12 (2014): 2851-2859. Web.
- Golliver, Ben. "Lakers' Kobe Bryant Discusses Knee Treatment in Germany, Achilles Progress." *SI.com*. Sports Illustrated, 9 Oct. 2013. Web.
- "Graphic: Unique Knee Surgery." Sports Column. Chicago Tribune, 22 Aug. 2012. Web.
- Joseph, Michael F., et al. "Achilles Tendon Biomechanics in Response to Acute Intense Exercise." *Journal of Strength and Conditioning Research*. 28.5 (2014): 1181-1186. Web.
- Parekh, Selene G., Walter H. Wray III, Olubusola Brimmo, Brian J. Sennett, and Keith L. Wapner. "Epidemiology and Outcomes of Achilles Tendon Ruptures in the National Football League." *Clinical Research*. 2.6 (2009): 283-286. Web.

- Rekhter, Mark D. "Collagen Synthesis in Atherosclerosis: Too Much and Not Enough." *European Society of Cardiology*. 41.2 (1998): 376-384. Web.
- Rosengarten, Samuel D, et al. "Australian Football Players' Achilles Tendons Respond to Game Loads within 2 Days: An Ultrasound Tissue Characterisation (UTC) Study." *British Journal of Sports Medicine*. 49.3 (2014): 183-187. Web.
- Rutland, Marsha et al. "EVIDENCE-SUPPORTED REHABILITATION OF PATELLAR TENDINOPATHY." *North American Journal of Sports Physical Therapy: NAJSPT* 5.3 (2010): 166–178. Print.
- Sánchez, Mikel, Eduardo Anitua, Juan Azofra, Isabel Andia, Sabino Padilla, and Iñigo Mujika. "Comparison of Surgically Repaired Achilles Tendon Tears Using Platelet-Rich Fibrin Matrices." *American Journal of Sports Medicine*. 35.2 (2007): 245-251. Web.
- Simonin, Marie-Agnes, Pascale Gegout-Pottie, Alain Minn, Pierre Gillet, Patrick Netter, and Bernard Terlain. "Pefloxacin-Induced Achilles Tendon Toxicity in Rodents: Biochemical Changes in Proteoglycan Synthesis and Oxidative Damage to Collagen." *Antimicrobial Agents and Chemotherapy*. 44.4 (2014): 867-872. Web.
- Stanish, William D., R. Mitchell Rubinovich, and Sandra Curwin. "Eccentric Exercise in Chronic Tendonitis." *Clinical Orthopaedics and Related Research*. 65.8 (1986): 65-68. Web.
- Wang, Hom-Lay, and Gustavo Avila. "Platelet Rich Plasma: Myth or Reality?" *European Journal of Dentistry* 1.4 (2007): 192–194. Print.
- Woodell-May, Jennifer, Dayna N. Ridderman, Matthew J. Swift, and Joel Higgins. "Producing Accurate Platelet Counts for Platelet Rich Plasma: Validation of a Hematology Analyzer and Preparation Techniques for Counting." *Journal of Craniofacial Surgery*. 16.5 (2005): 749-756. Web.
- "2013 Prohibited List." *World Anti-Doping Agency*. Anti-Doping Administration & Management System, 1 Dec. 2014. Web.