Platelet-rich plasma (PRP) is a substance that has been gaining importance in treating orthopaedic and sports injuries. PRP, in addition to having high concentrations of platelets, has been found to have high concentrations of growth factors. Both of these promote tissue healing and regeneration. As a result, PRP has found applications in musculoskeletal injuries, most commonly in chronic tendinopathy and muscle injury. This article reviews the biological properties, physiological actions, methods of preparation, and uses of PRP.

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and platelets. There are many chemical substances that also promote healing.

In all tissues there are different phases of healing. It takes place in three overlapping phases. The first phase, inflammation, occurs by initial haemostasis followed by neutrophil and platelet activation. Macrophages then enter and remove debris. Mesenchymal stem cells, fibroblasts and endothelial cells then enter into the region and proliferate. In the second phase, the proliferative phase, the mesenchymal stem cells differentiate into their specific cell lines such as osteoblasts, chondrocytes or fibroblasts. This is dependent on a number of chemical and mechanical triggers. In the final phase, remodelling, the collagen that has been laid down becomes more structured and organised. This stage may take months to years (2,3).

In musculoskeletal tissue, healing takes place by slightly different processes depending on the tissue that is damaged. Healed musculoskeletal tissues never achieve the histological or mechanical features of the original (Box 1).

PATHOPHYSIOLOGY OF MUSCULOSKELETAL TISSUE HEALING

1. Tendon

Within the tendon, acute healing often goes on to become chronic and can be separated into a paratenon-related process and an intratendinous process. Paratenonitis is the inflammation of the tendon sheath often seen in cylindrical tendons. Depending on the type of tendon, there may be associated synovitis and fluid. Tendinosis or tendinopathy is an intratendinous process. In tendinosis, histologically, there is a loss of the normal ordered architecture of the tenocytes. The fibres are widely spaced with mucoid substance interspersed within it. The tenocytes are abnormal in shape and show features of fibrocartilaginous metaplasia. The pathophysiology of this process is poorly understood but is thought to be due to collagen fibre disruption causing direct damage to tenocytes or trigger mechanoreceptors. Pain and tenderness caused by this may be due to local noxious substance formation or proliferation of pain nerve fibres (2). These findings suggest that although steroids and NSAIDs may help in reducing the nociceptive stimuli and activity of pain fibres, they do not aid in healing (Box 2).

2. Muscle

Healing of muscle injury is unlike tendinosis. It is caused by recurrent acute muscle injuries. Unlike some native tissues such as skin that regenerate completely, in muscle, the area of injury gets replaced with fibrous scar tissue. This is mechanically and functionally inferior to the surrounding muscle and is therefore prone to re-injury (4). The commonest muscles injured tend to be the thigh muscles such as the rectus femoris or the hamstrings (4,5) (Box 3).

3. Ligaments

Ligamentous healing has a similar healing process to other tissues although the proliferative phase and remodelling phase differ slightly. During the proliferative phase fibroblasts that have migrated into the healing ligament start initially laying down a matrix of proteoglycan and collagen, especially type III. In the next 6 weeks, increasingly organised collagen is laid down, especially type I and V. During the remodelling phase there is ongoing organisation and matrix maturation. The histological findings though show that there is a change in the collagen make-up that is different from the uninjured ligament. For example, in the medial collateral ligament (MCL), the standard ligament studied, the ratio of type V to type I collagen is altered from its native state. This invariably causes changes in the mechanical nature of the ligament. Clinically, each ligament has a different healing potential, rate of healing and a different end-composition. Finally, despite the same apparent injury, ligaments heal differently in different people (6) (Box 4).

BOX 1: KEY POINTS OF GENERAL TISSUE HEALING

- Healing of musculoskeletal tissues consists of 3 overlapping phases: inflammatory, proliferative and remodelling phases, which differ only slightly depending on the type of tissue damaged
- Healed musculoskeletal tissues never achieve the histological and mechanical properties of the native tissues.

BOX 2: KEY POINTS OF TENDON HEALING

- Tendon healing is poor due to its relative avascularity
- Tendon overuse results in a specific histological change known as tendinosis
- Pain in tendinosis is due to noxious stimuli and ingrowth of pain receptor nerve fibres, but not due to inflammation.

BOX 3: KEY POINT OF MUSCLE HEALING

- Healing of muscle is associated with a functionally and mechanically weaker muscle unit.

BOX 4: KEY POINTS OF LIGAMENT HEALING

- Ligament healing produces a mechanically and histologically different ligament
- Each ligament has a different healing potential, rate of healing and different collagen and proteoglycan composition
- Despite the same apparent injury, the same ligament will heal differently in different individuals.

EVIDENCE INFORMED PRACTICE
process of activation occurs by three known methods. Slow activation occurs when platelets come in contact with collagen or rough surfaces. Activation also occurs when platelets come in contact with calcium ions or thrombin. These secreted proteins not only recruit macrophages into the site of healing but also promote fibroblast and vascular ingrowth. Some of these secreted proteins released from platelets are absent in chronic inflammation (Box 5).

Another lesser known organelle in platelets is the dense granule. The dense granules contain a number of substances including adenosine, a nucleoside that plays a role in energy transfer and histamine, a local vasodilator and recruiter of immune mediators (8).

**PHYSIOLOGICAL ACTIONS OF PRP**

The idea that high concentrations of these growth factors could be prepared from human blood brought about the production of PRP. PRP contains a number of bioactive molecules that facilitate various components of healing and have higher concentrations than native blood. Typically, PRP contains 1 million platelets or more per microlitre, therefore having a significantly higher concentration of growth factors than normal blood (8). There are a number of commercially available methods that produce PRP.

PRP therefore has a number of beneficial properties. The most obvious action of PRP is the release of growth factors that restart healing in the injured tissues (11).

Once injected into tendons, it has been found to enhance gene expression of collagen. It has also been found to have mitogenic properties in tenocytes and stimulates the secretion of angiogenic growth factors such as VEGF and PDGF. This is helpful in neovascularisation, collagen deposition and organisation (12,13). It also has positive effects on chondrocytes and bone healing, although human studies have yet to confirm this (9).

Sanchez et al. have shown that PRP helps form the fibrin matrix, a necessary step in reconstituting the structural integrity of the tissue. It has been shown to bind not only platelets but also other cells such as fibroblasts and smooth muscle cells (14).

The ability of PRP to reduce local pain is a recently recognised property. The mechanism of this action is as yet unexplained (15).

Finally, an important effect of PRP has been its antibacterial and fungicidal properties. In vitro studies have found it has significant growth inhibitory actions on Staphylococcus aureus and Escherichia coli (16,17).

**METHODS OF PREPARATION**

PRP requires centrifuging of blood.
Although it is possible to use a standard laboratory centrifuge, the process entails multiple transfers of the plasma and two spins. Consequently sterility becomes an issue (3). There are a number of commercially available devices that produce PRP. They use different methods of preparation of PRP and also produce different concentrations of cells (3). These include systems such as the gPS system (Biomet, Warsaw, Ind), SmartPReP (Harvest Technologies Corp., Norwell, Massachusetts, USA) and Magellan (Medtronic, Minneapolis, Minnesota, USA). Table 3 shows some of the different products and methods of preparation used.

No consensus has been reached regarding standardisation of preparation or optimum concentrations. Different methods produce between 2- and 8-fold increases in platelet concentrations compared to native blood. Clinical benefits are more predictable between 4 and 5 times the normal blood concentration (12) although there is debate regarding this figure with some authors suggesting that a single spin may be sufficient for clinical purposes (18,19). There are also some reports saying that concentrations beyond 8-fold may be, paradoxically, detrimental to healing (3). Finally, as different growth factors favour different tissues, there is increasing interest in the quantities of growth factors that may promote different types of tissue repair (11,20).

USES OF PRP

PRP has found many uses in the practical management of sports injuries. These include tendon disease, ligamentous injuries, muscle injuries and more recently in chondral injuries.

### TABLE 1: GROWTH FACTORS RELEASED BY ALPHA-GRANULES AND THEIR ROLE

<table>
<thead>
<tr>
<th>GROWTH FACTOR</th>
<th>ROLES</th>
</tr>
</thead>
</table>
| Epidermal growth factor (EGF)          | ■ Attracts fibroblasts  
■ Causes cellular proliferation and differentiation |
| Insulin-like growth factor-1 (IGF-1)   | ■ Attracts fibroblasts  
■ Stimulates protein synthesis  
■ Enhances bone formation |
| Platelet-derived epidermal growth      | ■ Causes stimulation of epidermal regeneration factor (PDGF)  
■ Stimulates proliferation of keratinocytes and dermal fibroblasts |
| Platelet-derived growth factor (PDGF)  | ■ Stimulates angiogenesis and macrophage activation  
■ Causes proliferation, chemotaxis and collagen synthesis by fibroblasts  
■ Bone cell proliferation |
| Platelet factor 4 (PF4)                | ■ Attracts first wave of neutrophils  
■ Attracts fibroblasts |
| Transforming growth factor-β (TGF-β)   | ■ Causes proliferation of fibroblasts  
■ Stimulates type-1 collagen and fibronectin synthesis |
| Vascular endothelial growth factor (VEGF)| ■ Stimulates vascular endothelial cells to cause vascularisation of new tissue. |

Adapted from Engerbretsen et al. (9)

### TABLE 2: GROWTH FACTOR EXPRESSION IN MUSCULOSKELETAL TISSUES

<table>
<thead>
<tr>
<th>Factor</th>
<th>Muscle</th>
<th>Cartilage</th>
<th>Tendon/ligament</th>
<th>Bone</th>
</tr>
</thead>
<tbody>
<tr>
<td>GH</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>IGF-1</td>
<td>++</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>MGF</td>
<td>+++</td>
<td>?</td>
<td>+</td>
<td>?</td>
</tr>
<tr>
<td>B-FGF</td>
<td>+</td>
<td>+</td>
<td>±</td>
<td>+</td>
</tr>
<tr>
<td>PDGF</td>
<td>–</td>
<td>–</td>
<td>±</td>
<td>–</td>
</tr>
<tr>
<td>VEGF</td>
<td>+</td>
<td>–</td>
<td>±</td>
<td>–</td>
</tr>
<tr>
<td>TGF-β</td>
<td>±</td>
<td>+</td>
<td>±</td>
<td>+</td>
</tr>
<tr>
<td>BMP</td>
<td>+</td>
<td>+</td>
<td>–</td>
<td>–</td>
</tr>
</tbody>
</table>

**Abbreviations:** B-FGF, basic fibroblast growth factor; BMP, bone morphogenic protein; GH, growth hormone; IGF-1, insulin-like growth factor-1; MGF, mechano growth factor; PDGF, platelet-derived growth factor; TGF-β, transforming growth factor-β; VEGF, vascular endothelial growth factor.

Collagen deposition and remodelling of proteins, tenocyte differentiation, in angiogenesis, genetic expression, only recruiting cells, but also helping showing that PRP is helpful in not there has been mounting evidence and animal basic-sciences studies, tendinosis. From a number of in vitro shows PRP therapies help in healing of There is a large body of evidence that tendon disease but there are ongoing efforts towards this (9). In our practice, we have found good results in lateral epicondylitis and rotator cuff tendinopathy. In Achilles tendinopathy, I find better results using PRP with high volume paratenon stripping. My firm belief is that PRP injections should be combined with other treatments and should be tailored to the morphology of the tendon.

Muscle
PRP in the treatment of muscle injuries has been studied in the laboratory in animal studies. These include studies in rat models have been found to have good efficacy (33). A number of clinical studies, albeit having a low level of evidence, have pointed towards its efficacy in acute muscle injuries (8). So far, only one true level I study is currently in the literature (34).

The International Olympic Committee consensus paper on PRP published in 2010 mentions its use in acute muscle injury. It recognises the theoretical ability of PRP to help in muscle healing, but comments on a lack of scientific evidence in the world literature (9).

Other tissues
PRP has been put forward as a novel method in the treatment of cartilaginous disorders of the knee and hip. There is some laboratory evidence to show that PRP has a beneficial effect in the growth and differentiation of mesenchymal stem cells into chondrocytes (35). There is literature that suggests PRP may help restore the levels of hyaluronic acid within the knee and may be beneficial in osteoarthritis (14). Filardo and Kon et al used PRP injections in patients with chondromalacia. The study showed improvement in pain and quality of life.

### Table 3: A Comparison of the Devices Producing PRP

<table>
<thead>
<tr>
<th>Device technology</th>
<th>Example device</th>
<th>What the device produces</th>
<th>Fold increase of platelets per ml</th>
<th>Platelet yield (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Centrifugation (standard)</td>
<td>AutoloGel System (Cytomedix)</td>
<td>PRP</td>
<td>1–2</td>
<td>78</td>
</tr>
<tr>
<td>Cell-saver based</td>
<td>Electa</td>
<td>PRP</td>
<td>4–6</td>
<td>75</td>
</tr>
<tr>
<td>Computer-aided</td>
<td>Magellan (Artericyte Medical Systems)</td>
<td>PRP</td>
<td>51</td>
<td>76</td>
</tr>
<tr>
<td>Direct siphoning</td>
<td>GenesisCS</td>
<td>PRP</td>
<td>6</td>
<td>68</td>
</tr>
<tr>
<td>Direct aspiration</td>
<td>Secquire</td>
<td>PRP</td>
<td>16</td>
<td>31</td>
</tr>
<tr>
<td>Floating buoy</td>
<td>Biomet GPS</td>
<td>PCP</td>
<td>3.2</td>
<td>70</td>
</tr>
<tr>
<td>Platelet separation</td>
<td>Vivostat PRF</td>
<td>PRF</td>
<td>6</td>
<td>65</td>
</tr>
<tr>
<td>Platelet filtration</td>
<td>Caption</td>
<td>PC</td>
<td>4.3</td>
<td>–</td>
</tr>
</tbody>
</table>

PRP: platelet-rich plasma; PCP: platelet concentrated plasma; PRF: platelet-rich fibrin; PC: platelet concentrate.

### Tendon disease
There is a large body of evidence that shows PRP therapies help in healing of tendinosis. From a number of in vitro and animal basic-sciences studies, there has been mounting evidence showing that PRP is helpful in not only recruiting cells, but also helping in angiogenesis, genetic expression of proteins, tenocyte differentiation, collagen deposition and remodelling (12,13,21–24).

This has spurred a number of groups to look at the effects of PRP injections in tendinopathy and measure the outcomes. The largest studies have focused on lateral epicondylitis. A Level 1 study performed by a Dutch group, Gosens and Peerboms et al, showed PRP has a positive effect at two years compared to steroids. In their study using 100 patients, the group given PRP was found to have improved VAS (visual analogue scale) and DASH (disabilities of the arm, shoulder and hand) scores by greater than 25% compared to the group given corticosteroids (25). Thanasas’ group used a randomised controlled study to look at the effect of PRP versus autologous blood in lateral epicondylitis. Using similar outcome measures to the Dutch group, they found that the PRP produced better overall improvement in pain and function compared to the autologous blood group (26).

The results in Achilles tendinopathy have been less convincing. Although case reports and low level studies have suggested its efficacy, a Level I study by de Vos in 2010 using a double-blinded placebo-controlled randomised method with a cohort of 54 patients did not show much improvement in pain and function outcomes using the standardised VISA-A (Victorian Institute of Sport Assessment-Achilles) scores (27). We suspect this was due to the nature of the Achilles tendon compared to a flat tendon such as the common extensor origin. I believe that the paratenon in the Achilles tendon plays a significant role in pain and function disability here. In fact, histology of the tendinopathic Achilles tendon shows neovascularisation of the tendon with the sensory fibres running along with the blood vessels (28,29). Therefore, treatment of Achilles tendinopathy needs to be targeted towards both the tendon and paratenon.

Other tendons that are showing good results with PRP include the rotator cuff and patellar tendon (30–32).

As such, there are only a few Level I studies in the use of PRP in tendon disease but there are ongoing efforts towards this (9). In our practice, we have found good results in lateral epicondylitis and rotator cuff tendinopathy. In Achilles tendinopathy, I find better results using PRP with high volume paratenon stripping. My firm

- **Figure 3:** Ultrasound guided PRP injection of the common extensor origin
scores in patients up to 12 months after the PRP injection. There was a steady decline and recurrence of symptoms at 24 months (36–38). The maximum efficacy was found in patients with Kellgren–Lawrence knee scores between 1 and 2 (36,39). The results of the study performed by Filardo et al. suggest it has better outcomes than viscosupplementation in the knee (37). Sanchez et al. have performed a similar study in 40 patients with established severe osteoarthritis of the hip. This study also shows encouraging results with improvement of pain and function up to 6 months after the PRP injection (40).

The treatment of ligament injuries with PRP has not been studied in much depth. The vast majority of studies both in animals and humans have looked at its efficacy in the healing of the anterior cruciate ligament graft. The studies have not only looked at the rate of ligamentisation of the ACL graft and its effect on prevention of tunnel widening (41,42). Only one study has looked at the effects of PRP on medial collateral ligament healing in rabbits (43). There are also case reports and retrospective studies showing good outcomes in plantar fasciitis (44).

METHOD OF INJECTION

The treatment regimes for PRP injections are still a matter of debate. Most studies suggest between 2–6 ml of PRP is to be injected (3,27,45) into tendons, although there is no consensus on the quantities given. Most studies have given a single injection for tendinopathy and 3 consecutive intra-articular injections spaced 1–2 weeks apart for cartilage abnormalities (27,32,36,40,46).

A large-bore drawing up needle (<22 gauge) is suggested to prevent activation of the PRP. Once drawn up, mixing with calcium ions or bovine thrombin can activate the PRP. Calcium chloride has been recommended as the ideal activating calcium agent, although it is known to be an irritant to skin and can cause sloughing (47). Calcium gluconate, though, is a better agent as it can safely be injected subcutaneously.

Once informed consent has been obtained, the site of the injection is prepared with antiseptic solution. Local anaesthetic is infiltrated locally into the skin. In my experience, good local anaesthesia is necessary for areas such as lateral epicondylitis or plantar fasciitis as most patients experience moderate pain while the procedure is performed. This has to be weighed against the inactivation of PRP by local anaesthetics including Lignocaine and Marcaine (48). For procedures such as these, we regularly perform nerve blocks in addition to local skin infiltration to reduce discomfort from the procedure. For patellar and Achilles injections, we have found good results with high volume stripping of the paratenon prior with a combination of 0.5% Marcaine and 0.9% saline or a 25% dextrose-saline solution mixed with Marcaine (49). This not only disrupts the vessels and nerve fibres entering into the tendon but also provides excellent anaesthesia of the region.

It is recommended that the PRP injection should be performed under imaging (9). Ultrasound is a quick and easy real time method of injection. As there is no ionising radiation involved, it is safe. It can also be draped for aseptic precautions. Ultrasound also provides good visualisation of the adjacent traversing structures such as blood vessels and nerves.

In tendinopathy, it has been suggested that the tendon should undergo some dry needling to allow better dispersal of the PRP (45). Once the PRP has been injected, the area is bandaged. It is important to protect the tendon or joint injected. The tendon is theoretically weaker after dry needling and during the tissue healing period. No standardised rehabilitation regimes are recommended in the literature, but a period of 2 to 3 weeks of protection followed by physiotherapy is sensible. Physiotherapy with eccentric exercise regimes for tendons is an important part of the rehabilitation, especially in the common extensor origin and Achilles tendon. This is essential to re-align the collagen fibres laid down during the proliferative phase for optimal remodelling of the tendon (50).

Pain relief with paracetamol or acetylsalicylic acid is suggested during the first 6 weeks after the PRP injection. We provide patients with an information leaflet to prepare them for the procedure and have an in-house review at 6 weeks and 6 months to assess outcomes.

COMPLICATIONS AND CONTRAINDICATIONS

The only strict contraindication to the procedure is infection at the wound site. Others include platelet dysfunction and thrombocytopenia. Relative contraindications include use of local or systemic steroids.

Theoretically carcinogenesis is not possible with PRP. There is no evidence to support carcinogenesis in the literature to date.

The main complications in this therapy include infection although PRP has been found to prevent infection by some bacteria such as S. aureus and E. coli (16,17).

“PRP THERAPIES ARE EXTENSIVELY USED IN SPORTS AND MUSCULOSKELETAL INJURIES, MOST COMMONLY IN CHRONIC TENDINOPATHY AND MUSCLE INJURIES”
WADA CO CO AND USE OF PRP
The World Anti-Doping Agency (WADA) after much deliberation has removed PRP from its Prohibited Substances list in 2011 stating that PRP does not produce any systemic effects that may enhance the performance of athletes (51).

CONTROVERSIES WITH PRP STUDIES
As noted previously, much of the world literature on PRP is strong on the theoretical and laboratory evidence. Human studies on the effects of PRP in musculoskeletal injuries though are poor and consist of level 2 to 4 studies.

The most thoroughly investigated uses have been in tendinopathies, especially lateral epicondylitis and Achilles tendinopathy. The results, using level 1 study design, have been encouraging in lateral epicondylitis but equivocal in Achilles tendinopathy.

The quality of evidence peters out in other musculoskeletal injuries with most studies at best being level 2 or 3. There have been calls by the IOC and other stakeholders within the sports medicine community for better study design. The IOC consensus statement on PRP in sports, although generally encouraging, has highlighted the lack of conclusive evidence and criticised its use in sports medicine. This lack of level 1 studies has implications in not only standardising treatment regimes but also in designing rehabilitation programs and funding.

There has also been some interest amongst government health departments about its efficacy. A recent report by the National Institute for Health and Clinical Excellence suggested that PRP treatments should be given after clinical governance department at the local health trust was informed and that clear written information be given regarding the “uncertainty of the procedures efficacy” (52). The Ministry of health and Welfare in Korea have raised similar concerns (53).

Insurance companies and other private fund holders are also showing interest in the treatment. Although a difficult area to obtain information about, this procedure does appear to be funded by most private health insurers.

SUMMARY
PRP therapies are an exciting new development in the treatment of musculoskeletal injuries in sport. There are theoretical arguments and laboratory evidence to show that it can significantly improve healing in tissues. Clinically, this has lead to a boom in PRP treatments of various sports injuries with variable but generally good outcomes. Unfortunately, there is little level 1 evidence of its efficacy in the world literature. This has led the IOC; other sports medicine stakeholders and governments to call for better designed studies to look at its efficacy in various sports injuries. With the removal of PRP from the Prohibited Substances list by WADA in 2011, level 1 studies in elite athletes may become more common.

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KEY READING

REFERENCES
Due to the large number of references accompanying this article, for space reasons, we’ve had to restrict these to the online version. To view, please login to www.sportex.net or download the iPad/iPhone app which is free to subscribers.

THE AUTHOR
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If you have a current subscription, login at [www.sportex.net](http://www.sportex.net) to view this video or download the mobile apps which are free to subscribers with online access.

**ACHILLES TENDINOSIS**

(a) Ultrasound image showing the site of moderate Achilles tendinosis

(b) Video: Ultrasound doppler images of deep surface vascularity in Achilles tendinopathy

Research shows that the new vessels bring with them pain fibres that then contribute to the pain in Achilles tendinosis. The recent paper by de Vos (27) attempted to treat tendinosis with PRP injections. The present research into Achilles tendinosis suggests that pain is multifactorial. Disruption of the neovascularisation and paratenon adhesiolysis in addition to PRP in my practice has been of benefit.