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# Growth factor delivery methods in the management of sports injuries: the state of play

L Creaney,<sup>1</sup> B Hamilton<sup>2</sup>

## FDA CLEARED INDICATIONS FOR USE

The Double Syringe (ACP) System is used to facilitate the safe and rapid preparation of autologous platelet-rich-plasma (PRP) from a small sample of blood at the patient's point of care. The PRP can be mixed with autograft and allograft bone prior to application to an orthopedic surgical site as deemed necessary by the clinical use requirements.

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This article describes indications for use that are not cleared by the FDA.

## ABSTRACT

In recent years there have been rapid developments in the use of growth factors for accelerated healing of injury. Growth factors have been used in maxillo-facial and plastic surgery with success and the technology is now being developed for orthopaedics and sports medicine applications. Growth factors mediate the biological processes necessary for repair of soft tissues such as muscle, tendon and ligament following acute traumatic or overuse injury, and animal studies have demonstrated clear benefits in terms of accelerated healing. There are various ways of delivering higher doses of growth factors to injured tissue, but each has in common a reliance on release of growth factors from blood platelets. Platelets contain growth factors in their  $\alpha$ -granules (insulin-like growth factor-1, basic fibroblast growth factor, platelet-derived growth factor, epidermal growth factor, vascular endothelial growth factor, transforming growth factor- $\beta_1$ ) and these are released upon injection at the site of an injury. Three commonly utilised techniques are known as platelet-rich plasma, autologous blood injections and autologous conditioned serum. Each of these techniques has been studied clinically in humans to a very limited degree so far, but results are promising in terms of earlier return to play following muscle and particularly tendon injury. The use of growth factors in sports medicine is restricted under the terms of the World Anti-Doping Agency (WADA) anti-doping code, particularly because of concerns regarding the insulin-like growth factor-1 content of such preparations, and the potential for abuse as performance-enhancing agents. The basic science and clinical trials related to the technology are reviewed, and the use of such agents in relation to the WADA code is discussed.

Both tendinopathy and muscle strain injury are common sporting injuries with limited treatment options available. Until recently, rest, physical therapy and non-steroidal anti-inflammatory medications (NSAIDs) have been the mainstay of therapy.<sup>1</sup> However these techniques<sup>2,3</sup> may offer little beyond the body's own healing processes, and in fact it is possible that NSAIDs may impair the healing process.<sup>4</sup> The use of novel treatment techniques that utilise the body's own growth factors promises to provide a further therapeutic option to improve the quality and speed of recovery from injury.

It has long been recognised that growth factors are critical in wound healing,<sup>5</sup> but controlled delivery of growth factors has been a limitation to their clinical application. In recent years, a number of methods have been developed allowing the utilisation of the body's own growth factors, including systems that concentrate platelets,

commonly referred to as "platelet-rich plasma" (PRP) preparations. The technique of deriving PRP was developed in the mid 1990s for the discipline of maxillofacial surgery. Marx utilised the body's own blood to concentrate growth factors for use in dental and craniofacial surgery.<sup>6</sup> Its appeal has spread to plastic<sup>7,8</sup> and orthopaedic<sup>9</sup> applications and it is now commonly used in North America,<sup>7</sup> Spain<sup>10</sup> and Germany.<sup>9</sup> Its clinical utility has only recently been recognised in the UK.<sup>11</sup>

For those physicians working with elite athletes and potentially having the most to gain from their use, the challenge of utilising growth factor technology is both exciting and challenging in a sporting world where any mention of the words growth factor use is considered cheating.

This review assesses the current and potential use for growth factors and platelet preparations in sports medicine and considers its position relative to the World Anti-Doping Agency (WADA) code.

## BASIC BIOLOGY OF GROWTH FACTORS

Although this review primarily focuses on the therapeutic use of PRP, a brief overview of the subject of growth factors is required to place the technology in context.

Growth factors are a heterogeneous group of proteins (peptides) secreted by many different body tissues including connective tissue cells (eg, fibroblasts—fibroblast growth factor), haematopoietic stem cells (G-CSF—granulocyte colony-stimulating factor), white cells (interleukins, cytokines), platelets (PDGF—platelet-derived growth factor, vascular endothelial growth factor, transforming growth factor beta-1, epidermal growth factor and basic fibroblast growth factor) and solid organs such as the liver (IGF-1—insulin-like growth factor-1). They have a short biological half-life with quick systemic lavage leading to rapid disappearance of these substances from the circulation.<sup>12</sup> Subsequently, their effects are mostly confined to the site of delivery.<sup>13</sup>

Those growth factors found in platelets are stored within a cytoplasmic organelle called the  $\alpha$ -granule.<sup>14</sup> Platelets are the first cells to arrive at the site of an injury and, because of their capacity to release growth factors, they play a critical role in mediating healing of the injured tissue. This has led to the use of platelets as a delivery tool for growth factors.

## GROWTH FACTORS AND SOFT-TISSUE HEALING

Literature on the role of growth factors in tissue regeneration is abundant, and it is not the purpose of this review to examine the literature comprehensively with respect to animal studies or the use

of isolated growth factors, but rather to place the technology of PRP within context.

IGF-1 has been used in isolation by a number of investigators. Animal studies have suggested a role for IGF-1 in both the acceleration and enhancement of healing of tendon and muscle injuries.<sup>12 15</sup>

Menetrey *et al*<sup>12</sup> studied the use of fibroblast growth factor (FGF) in healing muscle alongside IGF-1, and demonstrated that it can also increase the number of regenerating myofibres and functional recovery compared to control, though to a lesser extent than IGF-1. Efthimiadou<sup>16</sup> found that basic FGF also increased angiogenesis in healing rat gastrocnemius muscle.

It is thought that PDGF may play a significant role in the early stages of healing where it may induce synthesis of other growth factors such as IGF-1.<sup>5</sup> Hildebrand *et al*<sup>17</sup> applied PDGF to ruptured rabbit medial collateral ligament. Biomechanical evaluation was performed at 6 weeks with improvement in the parameters of ultimate load to failure, energy absorbed to failure and ultimate elongation values of 1.6–2.4 that of controls. Further study in rat medial collateral ligament (MCL) demonstrated a 73% improvement in healed ligament strength against controls at only 12 days.<sup>18</sup>

## PLATELET-RICH PLASMA

Platelet-rich plasma is the therapeutic outcome of a technique involving the centrifugation of an autologous sample of human whole blood, which allows the extraction of that part of the plasma that contains a high concentration of platelets.

The methodology developed by Marx<sup>19</sup> requires a sample of blood to be obtained with the addition of an anticoagulant, such as anticoagulant citrate dextrose A, in order to prevent platelet activation before therapeutic use. The sample is spun twice, the first time to separate the red blood cells from the plasma and the second time to concentrate the platelets in the plasma. This second spin results in the formation of two layers within the plasma—a platelet-poor component (PPP) and a platelet-rich component (buffy layer) called platelet-rich plasma (PRP). The platelets are activated at the time of injection with the addition of calcium (Ca<sup>2+</sup>) and thrombin.

The resulting platelet-rich plasma has been found to contain up to 4–8 times the concentration of platelets found in whole blood<sup>6 7</sup> (table 1). The technology is still evolving and, consequently, the process of platelet concentration appears to result in a variable increase in  $\alpha$ -granule-derived growth factors.<sup>6 7</sup> (tables 1 & 2).

**Table 1** Absolute growth factor concentrations in PRP

PRP contents & normal values where known	Sanchez <sup>21</sup>	Eppley <sup>8</sup>	Anitua <sup>22</sup>	Marx <sup>19</sup>
Platelet count (150–400 × 10 <sup>9</sup> l <sup>-1</sup> )	634	1600	460	1086
$\alpha$ -Granule factors				
EGF (129) <sup>8</sup> (pg/ml)	481.5	470	442.5	–
VEGF (155) <sup>8</sup> (pg/ml)	383	955	297.5	–
TGF- $\beta_1$ (35) <sup>8</sup> (ng/ml)	74.99	120	37.83	170
PDGF (3.3) <sup>8</sup> (ng/ml)	35.62	17	13.33	133
bFGF	trace <sup>9</sup>	–	–	–
Plasmatic factors				
IGF-1 (ng/ml)	94.53	No $\uparrow$	115.71	No $\uparrow$
HGF (pg/ml)	593.87	–	435	–

bFGF, basic fibroblast growth factor; EGF, epidermal growth factor; HGF, hepatocyte growth factor; IGF-1, insulin-like growth factor-1; No  $\uparrow$ , no increase; PDGF, platelet-derived growth factor; PRP, platelet-rich plasma; TGF- $\beta_1$ , transforming growth factor beta-1; VEGF, vascular endothelial growth factor.

While the techniques for the isolation and differentiation of growth factors from PRP continue to develop, it is recognised that PRP contains the following growth factors.<sup>20</sup>

Platelet  $\alpha$ -granule-derived:

- ▶ platelet-derived growth factor (PDGF)
- ▶ vascular endothelial growth factor (VEGF)
- ▶ transforming growth factor beta-1 (TGF- $\beta_1$ )
- ▶ epidermal growth factor (EGF)
- ▶ basic fibroblast growth factor (bFGF)
- ▶ insulin-like growth factor-1 (IGF-1).

Plasma derived:

- ▶ hepatocyte growth factor (HGF)
- ▶ insulin-like growth factor-1 (IGF-1).

Excluding HGF and IGF-1, the above factors are locally acting paracrine factors with few or unknown systemic actions.<sup>20</sup> By contrast, IGF-1 has several sources. It is released systemically from the liver, under the control of pituitary growth hormone and from skeletal muscle following exercise,<sup>23 24</sup> as well as being present in and secreted from platelet  $\alpha$ -granules at sites of tissue injury.<sup>20</sup> While PRP does contain some IGF-1, the proportions derived from plasma versus platelets remains unclear. It is likely that platelets may release trivial amounts of IGF-1,<sup>8</sup> however, most of the IGF-1 present in PRP will be derived from the original plasma.<sup>6 8 9 22 25</sup>

PRP extract is injected directly into the damaged tissue, the aim being to enhance the wound healing through delivery of growth factors and theoretical optimisation of the healing environment.<sup>10</sup> Because it is an autologous sample the risk of allergy or the introduction of exogenous infection is considered negligible.<sup>6 13</sup> Once delivered the platelets begin active secretion of growth factors within 10 min, and more than 95% of the presynthesised growth factors are released within an hour.<sup>19</sup> Platelets are viable for 7 days and will continue to release growth factors into the tissue during this time.<sup>19</sup> While Marx<sup>19</sup> has suggested that a PRP preparation must contain at least 4–5 times the concentration of platelets compared to plasma in order to be effective, clinical efficacy of PRP has been demonstrated by other groups<sup>9 21</sup> with less concentrated preparations.

A theoretical advantage of PRP over the use of purified individual growth factors is that PRP contains several different growth factors, present in physiological proportions. Consequently a natural balance of proliferative and inhibitory effect would be expected, rather than the potentially unbalanced effects that may ensue when using purified isolated growth factors. Since the injected platelets are viable for a further 7 days in the tissue and continue to release growth factors for this period, more than one injection is unnecessary.<sup>19</sup>

## CLINICAL USE AND EFFICACY OF PRP

Within the field of musculoskeletal medicine, there is only limited clinical research on growth factor delivery methods.<sup>7 12 26–28</sup> Those limited clinical trials of tendon injury where these methods have been utilised tend to lack robustness, and have yet to be reproduced (table 3). Lateral and medial epicondylitis, patella tendinopathy and Achilles tendinopathy have all been investigated to varying extents in animals and humans<sup>7 21 26–31</sup> Studies of PRP technology in muscle injury are minimal.<sup>32</sup>

## Animal studies

Animal studies are numerous, though extrapolation of data to humans is of questionable validity. An injection of platelet concentrate into surgically injured Achilles tendons in rats led to a 30% increase in tensile strength at 1 week in a single study.<sup>33</sup>

**Table 2** Biological roles and relative increase (multiplication factor) from baseline of growth factors derived from platelet-rich plasma (PRP) and autologous conditioned serum (ACS) technique compared with concentrations in whole blood (ABI)

	Source	Role	PRP	ACS
Platelets	Blood	Initial control of haemorrhage, release GF at injury site	3–8 <sup>6–8</sup>	N/A (serum contains no platelets)
PDGF	Platelets	Stimulates cell replication, angiogenesis, mitogen for fibroblasts <sup>14</sup>	5–29 <sup>9 20</sup>	No difference <sup>26 29</sup>
VEGF	Platelets	Angiogenesis <sup>34</sup>	6–52.7 <sup>8 9</sup>	No data
TGF- $\beta_1$	Platelets	Key regulator in balance between fibrosis and myocyte regeneration <sup>4 35</sup>	3.5–27 <sup>8 9 20</sup>	1.3 <sup>26 29</sup>
FGF	Platelets	Simulates proliferation of myoblasts, angiogenesis <sup>1 8 12</sup>	"detected" <sup>19</sup>	7.5 <sup>26 29</sup>
EGF	Platelets	Proliferation of mesenchymal & epithelial cells, potentiation of other GFs <sup>36</sup>	3 <sup>8</sup>	No data
HGF	Plasma	Angiogenesis, mitogen for endothelial cells, <sup>21</sup> anti-fibrotic <sup>10</sup>	No increase from baseline <sup>22</sup>	1.3 <sup>26</sup>
IGF-1	Plasma/liver	Stimulates myoblasts & fibroblasts, mediator in growth & repair of skeletal muscle <sup>35 37</sup>	No increase from baseline <sup>8 9 22 25</sup>	No difference <sup>26</sup>

EGF, epidermal growth factor; GF, growth factor; HGF, hepatocyte growth factor; IGF-1, insulin-like growth factor-1; PDGF, platelet-derived growth factor; PRP, platelet-rich plasma; TGF- $\beta_1$ , transforming growth factor beta-1; VEGF, vascular endothelial growth factor.

Carda *et al*<sup>32</sup> looked at surgically induced muscle injury in sheep, and demonstrated accelerated healing in PRP-treated animals. Lefaucheur *et al*<sup>35</sup> examined mouse muscle injury with antibodies to neutralise bFGF, IGF-1 and TGF- $\beta_1$ . They found that there was attenuation of the healing response, demonstrating that removal of these factors leads to poorer healing.

### Human studies

Sanchez *et al*<sup>38</sup> applied PRP to the ruptured Achilles tendons of a professional basketball player and a professional footballer in

conjunction with operative repair, and reported a return to full match fitness in 14 weeks. A further report by this same group<sup>21</sup> described a case series of six athletes undergoing open suture repair following complete Achilles tendon rupture, and compared the outcome with a comparable group who received the same operation with the addition of an injection of PRP to the wounded ends when sutured together. Their results showed that PRP-treated patients recovered their range of motion (ROM) sooner, had no wound complications and took less time to run and resume training. The cross-sectional area of the

**Table 3** Summary of studies using growth factor application methods in animal and human trials of tendon, ligament and muscle healing

Technique	Species	Tissue type	Study details	Results	Type
ABI	Animal	Tendon Ligament		No harmful effects	Crossover
	Human	Tendon	Edwards 2003 <sup>28</sup> —lateral epicondylitis	79% patients complete pain relief	Cohort
		Ligament	Connell 2006 <sup>27</sup> —medial epicondylitis Connell 2006 <sup>27</sup> —lateral epicondylitis	No pain at 6 months	
ACS	Animal	Muscle	Wright-Carpenter 2004 <sup>26</sup> —mice gastrocnemius	Increased satellite cells & myofibres	Controlled trial
	Human	Muscle	Wright-Carpenter 2004 <sup>29</sup> —human skeletal muscle	Improved recovery 22.3 vs. 16.6 days	Controlled trial
PRP	Animal	Tendon Ligament	Aspenberg 2004 <sup>33</sup> —rat Achilles tendon rupture	30% improved strength at 1 week	Cohort
		Muscle	Carda 2005 <sup>32</sup> —skeletal muscle tears	Improved healing at 6 days	Cohort
	Human	Tendon	Mishra 2006 <sup>7</sup> —elbow tendinopathy	60% ↓ VAS at 8/52 vs. 16% ↓ control	Controlled trial
		Ligament	Sanchez 2005 <sup>38</sup> —Achilles tendon rupture	Full recovery 14 vs. 21 weeks	Case report
			Sanchez 2007 <sup>21</sup> —Achilles tendon rupture		Non-randomised trial
Muscle	Sanchez 2005 <sup>39</sup>	Full recovery in half the time of controls	Case series		
Suramin	Animal	Muscle	Chan 2005 <sup>31</sup> —mice gastrocnemius	↓ Scar tissue ↑ Tetanic strength	Controlled trial
Relaxin	Animal	Muscle	Negishi 2006 <sup>40</sup> —mice skeletal muscle	↓ Fibrosis ↑ Myofibre regeneration	Crossover

ABI, autologous blood injections; ACS, autologous conditioned serum; PRP, platelet-rich plasma.

treated tendons increased to a lesser degree compared to non-treated tendons.<sup>21</sup>

Mishra *et al* investigated the use of PRP in elbow epicondylar pain in a partially randomised trial of a series of 15 patients with chronic lesions (mean 15 months) who had failed conservative therapy and a control group of 5 patients. At 4 weeks post-injection, PRP-treated patients reported a mean of 46% improvement in visual analogue pain scores compared with 17% in controls. At 8 weeks, they reported a 60% improvement in pain scores compared with a 16% reduction in the controls, at which point three out of five controls had dropped out of the study to seek alternative treatment. At 6 months pain scores were reduced by 81% in the treatment group. At final follow-up (2 years) PRP-treated patients reported a 93% improvement in pain, and 94% returned to sport and work.

### Human studies on muscle injury

Sanchez *et al* also published a case study of 20 professional athletes with small muscle tears who were injected with PRP under ultrasound scan (USS) guidance, and found that athletes reported functional recovery in half the time expected.<sup>38</sup> Significantly, this study did not appear to result in any excessive fibrosis, which may have been suspected from the involvement of TGF- $\beta_1$ .

### AUTOLOGOUS BLOOD INJECTION

Autologous blood injection (ABI) refers to the re-injection, at an injury site, of a few millilitres of blood taken from the patient. The injection will contain some platelets capable of releasing growth factors, but at much lower levels than seen with the PRP technique. In a study using rabbits, normal patella tendons injected with ABI were found to have normal histology and a 15% increase in tensile strength compared with controls at 12 weeks.<sup>30</sup> The purpose of this study was to demonstrate the safety of ABI.

In humans, the most extensively investigated pathologies are medial and lateral epicondylitis. Connell *et al*<sup>27 41</sup> used ultrasound-guided ABI in two series of 20 and 35 patients with medial and lateral epicondylitis. They reported reduction in pain scores of 60% at 2 months and 100% at 6 months. This same group also recently evaluated the effect of ABI plus physiotherapy in patella tendinosis in a series of 47 knees. They found that 44 patients returned to sport at a mean of 14.8 months.<sup>42</sup>

### AUTOLOGOUS CONDITIONED SERUM

Autologous conditioned serum (ACS) involves incubating the blood with glass beads and spinning the blood down to extract the serum containing the released growth factors. This method produces a lower yield of growth factors than PRP<sup>26 29 43</sup> since the method was originally described<sup>45</sup> for the production of inflammatory cytokines (IL-4) rather than growth factors. This technique has been investigated in muscle strain injury and has been shown to be effective.<sup>26 29</sup> Evidence from studies in rats has suggested the process of muscle healing is often incomplete due to the formation of scar tissue (fibrosis) at sites where muscle satellite cells need to rebuild injured sarcomeres.<sup>26 29</sup> Two studies, one in rats and the other in humans, looked at the effects on histological and clinical outcomes of injecting ACS into injured muscle<sup>26 29</sup> (see table 3). In ACS-treated rat muscle injury, there was an 84% increase in satellite cell activation, a 27% increase in regeneration of myofibres and increased angiogenesis versus controls. In ACS-treated human muscle strain injury, there was complete versus partial regression of

subjectively assessed MRI findings in ACS-treated patients and return to sport at a mean of 16.6 days versus 22.3 days in controls.

## POTENTIAL RISKS OF USING GROWTH FACTORS IN SPORTS MEDICINE

There is the potential for both local and systemic adverse effects when using growth factor delivery methods.

### Potential local complications

A potential local complication of growth factor administration is induction of excessive fibrosis in the healing tissue. Muscle healing takes place in the following four overlapping stages<sup>31 35 44</sup>:

- ▶ degeneration<sup>35</sup>
- ▶ inflammation (first few days)<sup>35</sup>
- ▶ regeneration (beginning day 5, peaking at day 14)<sup>35</sup>
- ▶ fibrosis (beginning in second week), which may become an overly aggressive healing response in extensively injured muscles.<sup>31 44 45</sup>

Fibrosis is problematic in muscle healing since complete muscle regeneration cannot occur in the presence of fibrosis.<sup>44 45</sup> A key regulator of this process is TGF- $\beta_1$ , which appears to regulate the balance between regeneration and fibrosis.<sup>31</sup> It is possible that use of multiple growth factors in muscle injury may result in increased fibrosis and impair long-term outcomes.

Until recently, the use of NSAIDs has been promoted in muscle injury. However, Wei-Shen<sup>4 44</sup> has demonstrated that NSAIDs may impair muscle healing and promote fibrosis by increasing expression of TGF- $\beta_1$  and reducing expression of prostaglandin E<sub>2</sub>. Prostaglandin E<sub>2</sub> plays a role in the proliferation and differentiation of muscle satellite cells. Thus, NSAIDs may impair muscle healing by delaying muscle regeneration and increasing scar tissue formation.

In order to address this problem, a number of substances have been tested including Decorin, Relaxin,<sup>40</sup> matrix metalloproteinases<sup>46</sup> and Suramin<sup>31</sup> in the hope that they may provide therapeutic options to limit fibrosis. Suramin, a polysulphonated naphthylurea, is an antiparasitic and antitumor drug that acts as an inhibitor of TGF- $\beta_1$  by competitively binding to the growth factor receptor. It has been shown to significantly reduce fibrotic tissue and increase the number of regenerating myofibres in mice when injected on day 14 post injury.<sup>31</sup> Furthermore, there was increased fast-twitch and tetanic strength compared to controls. Relaxin, an ovary-derived hormone structurally related to IGF,<sup>40</sup> plays a role in pregnancy, softening the symphysis pubis and cervix in preparation for labour. It also has effects on collagen production and degradation.<sup>40</sup> Relaxin has also been used to prevent muscle fibrosis after injury with promising results.<sup>40</sup>

### Potential systemic complications and effects

#### Infection

Since PRP is an autologous preparation the risk of introducing foreign material is effectively eliminated, although the entire procedure must be carried out in sterile conditions. The use of autologous blood products in this manner reduces the risk of transmissible infection and allergic reaction. Earlier techniques relied upon the use of topical bovine thrombin, containing contaminants such as bovine factor Va as a platelet activation source.<sup>19</sup> This resulted in antibodies to factors V and VI, with potentially life-threatening coagulopathies resulting,<sup>13</sup> and as a

consequence is no longer utilised in commercially available techniques in the UK.

### Carcinogenesis

Growth factors act on cell surface receptors, do not enter the cell and do not cause DNA mutation. There is no plausible mechanism by which growth factors result in neoplastic development, and there have been no reports of this in the literature.<sup>6, 19</sup>

### Effect on serum growth factor levels

Recent research by Banfi's group in Italy<sup>47</sup> looked at the potential systemic effects of locally administered PRP. They found that a locally administered injection of PRP (4 patella tendons, 1 elbow) led to a fall in the serum concentration of EGF. There was no statistically significant difference in the concentration of VEGF, measured at 30 min, 3 h and 24 h post injection; other growth factors were not measured. A limitation of this study unfortunately was its small size ( $n = 5$ ), but the implication is that locally administered PRP will impact on systemic levels of growth factors, but in a negative manner.

### THE USE OF GROWTH FACTOR METHODS AND CONFLICT WITH THE WADA CODE

In the UK, the use of autologous blood products containing growth factors entered the public arena when a premiership football club sports physician made enquires to the national anti-doping organisation (UK Sport) and WADA regarding the legality of their use in sport.<sup>48</sup> The question asked forced WADA to consider their position on both the use of autologous blood injections and any autologous product that contains growth factors.

The response from WADA was quite clear, that the use of either of these techniques is prohibited under the terms of the WADA Prohibited List (B Hamilton, personal communication). The use of ABI as described above was considered prohibited under section M1<sup>49</sup> while the use of any autologous product that contains growth factors was prohibited under section S2. This section specifically mentions growth hormone, IGF-1 and mechano growth factor (MGF).

### IGF-1 content of PRP: therapy versus doping

IGF-1 is a 7.5 kDa polypeptide that is structurally similar to insulin.<sup>50</sup> It induces proliferation, differentiation and hypertrophy of multiple cell lines, in particular skeletal muscle, and has an additional role of facilitating glucose entry into skeletal muscle cells.<sup>23, 50</sup>

IGF-1 is secreted as the result of a hypothalamic-pituitary-liver axis. The hypothalamus secretes growth hormone-releasing hormone (GHRH), which stimulates the pituitary to release growth hormone, which in turn stimulates the liver to release IGF-1.<sup>23</sup> Like most endocrine systems, the system is controlled by negative feedback, thus in normal individuals, exogenous administration of IGF-1 will lead to suppression of the axis. Whereas growth hormone secretion is pulsatile, with greatly varying levels in a 24-hour period, serum IGF-1 levels are relatively stable within a 24-hour period; hence, measurement of the serum IGF-1 level is now the favoured test for acromegaly or growth hormone deficiency.<sup>51</sup>

IGF-1 circulates in the serum 99% bound to the carrier protein insulin-like growth factor binding protein-3 (IGFBP-3). Only 1% of serum IGF-1 is "free" (fIGF-1), and it is the free portion that is believed to exert the biological effects upon binding to the IGF-1

receptor (IGF-1R).<sup>51</sup> IGF-1 has a serum half-life of 10 min ( $t_{1/2}$  10 min) when unbound to IGFBP-3,<sup>52</sup> and it is in this unbound form that IGF-1 is administered within PRP. In contrast, the IGF-1/IGFBP-3 complex has a much longer half-life of 16 h ( $t_{1/2}$  16 h).<sup>52</sup>

IGF-1 has at least three isoforms: IGF-1Ea, IGF-1Eb and IGF-1Ec. IGF-1Ea is the circulating form of IGF-1, which is released from the liver, whereas IGF-1Ec, also known as mechano growth factor (MGF), is the tissue isoform released from skeletal muscle cells and is believed to exert exclusively autocrine/paracrine actions.<sup>23</sup>

The different isoforms have slightly different biological actions. IGF-1Ea is known to stimulate terminal differentiation of muscle cells into myotubes and promote stem cell-mediated muscle regeneration, whereas MGF is damage sensitive, controls local tissue repair and is more potent than IGF-1Ea at causing hypertrophy.<sup>23</sup> MGF is rapidly degraded in the serum.<sup>23</sup>

These varying biological actions of IGF-1 isoforms are important since IGF-1 derived from PRP (IGF-1Ea), which is used for therapeutic purposes, may not have the same performance-enhancing effects as skeletal muscle-derived IGF-1Ec (MGF).

Serum IGF-1 levels vary greatly among individuals and are dependent on genetic influences and nutritional status; however, a typical value of 300 ng/ml (range 94–506 ng/ml) is seen in 17 to 20-year-old adults and 250 ng/ml (range 117–358 ng/ml) in 21 to 30-year-olds (C Camacho-Hubner and L Perry, personal communication). In order to achieve such physiological levels, children with Laron syndrome, a rare form of growth hormone resistance typified by very low levels of natural IGF-1, are given exogenous IGF-1 160 µg/day for many months.<sup>53</sup> Contrast this with a typical dose of a single locally administered PRP injection in the treatment of elbow extensor tendinopathy—3 mls of PRP containing ~100 ng/ml of IGF-1 (total dose 300 ng)<sup>7</sup> and there is a demonstrable  $5 \times 10^2$  fold difference in even a single dose.

It is also important to mention that exercise has some effect on circulating levels of IGF-1. Berg *et al*<sup>24</sup> studied changes in serum IGF-1 in relation to acute bouts of exercise. This group demonstrated a 27% increase in serum IGF-1 following 10 min of moderate exercise in healthy adults, corresponding to changes of 10–28 µg/l. This was likely to be IGF-1 released from skeletal muscle, therefore, it would be difficult to differentiate changes in serum IGF-1 as a result of exercise from changes caused by exogenous administration.

Thus, there appear to be several compelling reasons to support the belief that PRP is unlikely to be a potent ergogenic aid:

- ▶ The unbound IGF-1 has too short a half-life to be able to exert systemic effects (10 min versus 16 h).
- ▶ The isoform IGF-1Ea found in PRP is not the isoform principally responsible for skeletal muscle hypertrophy (IGF-1Ec/MGF).
- ▶ The doses of IGF-1 in PRP are subtherapeutic in terms of producing systemic anabolic actions by a factor of 500 (300 ng versus 160 µg).

A recent International Olympic Committee (IOC) Medical Commission Consensus Statement on the use of growth factor technologies in therapy appears to welcome further research in the field "to ensure these therapies are optimised" and "to ensure athlete/patient safety". We would welcome such assertions also, though the statement by the IOC somewhat contradicts the WADA Code, which prohibits all use of growth factor therapies in elite sport.<sup>54</sup>

### What is already known on this topic

- ▶ Growth factors mediate tissue repair following injury.
- ▶ Various techniques have been developed to deliver increased concentrations of growth factors to sites of injury, including autologous blood injections and PRP.
- ▶ Robust data of clinical efficacy are lacking.
- ▶ The use of growth factors is prohibited by the WADA code.

### What this study adds

- ▶ Growth factor technologies have the potential to accelerate healing in soft tissue injury.
- ▶ The debate of therapeutic use versus doping needs to be opened in relation to the WADA code.

Notwithstanding these concerns expressed by WADA, it is possible to apply to a WADA approved anti-doping organisation for a therapeutic use exemption (TUE) to utilise these techniques for specific clinical indications in elite athletes. Given the obvious difficulties associated with detection of these techniques and the bureaucratic delays the TUE process entails, it is unclear whether this approach has been widely utilised. Indeed, the only research known to have been conducted on professional athletes makes no mention of any anti-doping concerns.<sup>47</sup> The authors would strongly support the use of this approach in order for WADA to develop awareness of the current clinical utility of these techniques.

### CONCLUSION

Medical technology continues to advance at a furious pace. The use of growth factors promises to herald a new era of accelerated healing of injured tissues, and is already commonplace in many fields of medicine. The technology is still in its infancy with respect to soft tissue injuries, and the precise mechanisms of action and optimum therapeutics need to be developed. The use of PRP promises to become a powerful therapeutic modality for use in muscle, tendon and ligament injury in the future, but at present its use is considered a doping violation under the WADA code, so research and treatment is restricted to non-elite sportspersons. In the future an ironic dichotomy may exist whereby the general public will be able to benefit from state of the art medical technology utilising growth factors, but elite athletes will be excluded because of doping restrictions. WADA and the International Olympic Committee must work with scientists to allow athletes to benefit from the best medicine available in a both a safe and fair environment.

**Competing interests:** None.

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