P.R.P. Therapies

Self Stimulated Serum – S3
Dr. D. SISTER

From Wikipedia, the free encyclopedia:

Blood is a specialized bodily fluid that delivers necessary substances to the body's cells
– such as nutrients and oxygen
– and transports waste products away from those same cells.

- In vertebrates, it is composed of blood cells suspended in a liquid called **blood plasma**.
- The blood cells present in blood are mainly red blood cells (also called RBCs or erythrocytes) and white blood cells, including leukocytes and platelets.

- **Plasma**, which comprises 55% of blood fluid, is mostly water (90% by volume), and contains:
  - dissolved proteins,
  - glucose,
  - mineral ions,
  - hormones,
  - carbon dioxide (plasma being the main medium for excretory product transportation),
  - platelets and blood cells themselves.

Platelets are small (2 micrometer) but powerful.

- Consist of :
  - Pseudopodial extensions
  - Invaginations
  - 3 types internal vesicles (contain inactive growth factor)
    - Lysosomal vesicle
    - Dense vesicle
    - Alpha vesicle

- They contain a variety of Growth Factors :These are small molecules also known as cytokines that interact with the local cells and even send signals that initiate a variety of important events such as cell division and migration.

- ***Alpha vesicle*** store and releases the **Growth Factors,** these GF promote healing and regeneration (tissue healing and bio-cellular regeneration). Thus, the GF are relevant to wound-healing.
  - PDGFaa, PDGFbb, PDGFab (Platelet Derived Growth Factor)
  - TGFbeta1, TGFbeta2 (Transforming Growth Factor Beta)
  - VEGF (Vascular Endothelial Growth Factor)
  - EGF (Epidermal Growth Factor)
  - IGF (Insulin Growth Factor)

- They are proteins and need to be biologically active to work.
- The alpha-vesicles also contain cell-adhesion molecules and involved with vitronectin, fibronectin, fibrin.

Characteristics of Growth Factors

- **PDGF: (Platelet Derived Growth Factors)**
  - Chemo attractive to Mesenchymal Stem Cells and endothelial cells.
  - Differentiation for fibroblasts and osteoblasts.
  - Up regulate effects of other growth factors on cells such as macrophages.
  - Mitogenes of mesenchymal stem cells promote the synthesis of the extra cellular matrix

- **TGF: (Transforming Growth Factors alpha and beta)**
  - Promotes cell mitosis
  - Significantly increases type I Collagen production in tendon - Favours the **synthesis of collagen.**
  - Sheath fibroblast
  - Stimulation of DNA synthesis, proliferation of various types of cells.

- **VEGF: (Vascular Endothelial Growth Factor)**
  - Stimulates angiogenesis, chemo attractive for osteoblasts

- **EGF: (Epidermal Growth Factor)**
  - Important role in the regulation of cell growth, proliferation, and differentiation by binding to its receptor EGFR
  - Induce epithelial development and promote angiogenesis

  Stimulates proliferation and differentiation of **epidermis cells,** co-stimulating angionegesis.

- **IGF 1 and 2** (Insulin Like Growth Factors): Stimulates the proliferation and differentiation of osteoblats.

In addition the activated thrombocytes have onto their surface a multitude of signalisation molecules: CD9, CD-W17, CD41, CD42a-d, CD51, CD-W60, CD61, CD62P, CD63

What's PRP?

- **PRP** is autologous or derived from the same person, and is referred to as blood component therapy, rich in concentrated platelets (5 to 8 times)

- normal concentration of platelet: 200,000 per micro litre
  versus PRP: 1,000,000 per micro/L

PRP consist :
- Platelets: 94%
- Red Blood Cells: 5%
- White Blood Cells:1%

PRP is also a concentration of the 7 fundamental protein growth factors that are actively secreted by platelets to initiate all wound healing.

PRP includes 3 proteins in blood known to act as cell adhesion molecules: fibrin, fibronectin, vitronectin.

PLATELET-RICH PLASMA (PRP):
PATIENT'S OWN ENRICHED PLASMA

Science supports the use of **PRP** for the augmentation and regeneration of hard and soft tissues.

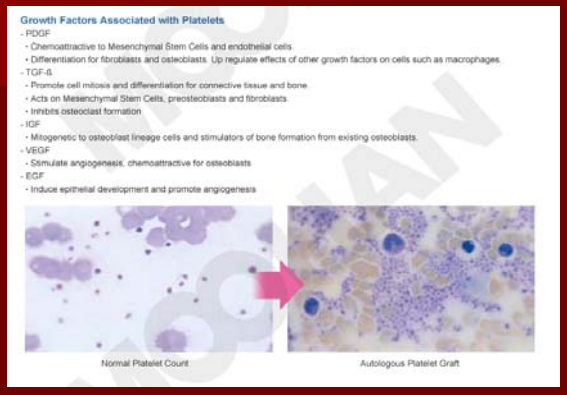
Today, Platelet Rich Plasma can easily be separated from blood, through a process of centrifugation.

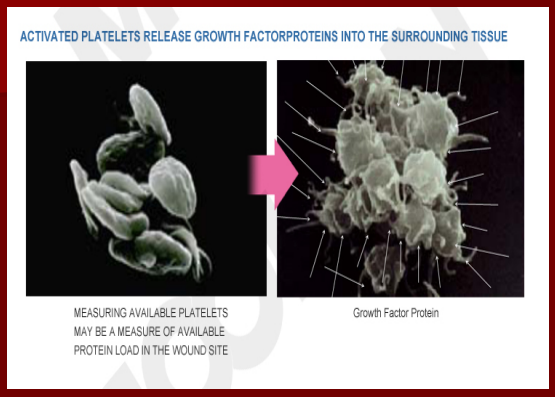
Using the enriched plasma permits the body to heal faster and more efficiently. Because the patient's own plasma (autologous) is used, there is no danger of disease transmission, anaphylaxis or neoplasia.

In general medicine, platelet transfusion is common practise in treating blood platelet disorders, including very low counts associated with bleeding.

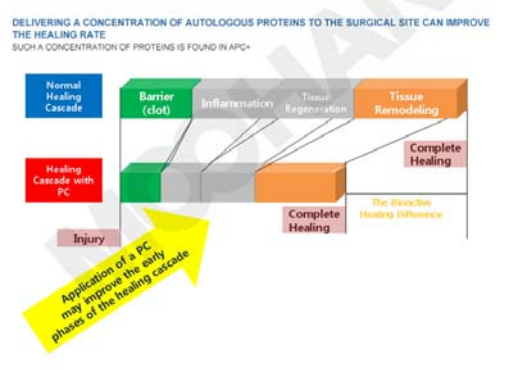
Biological actions:

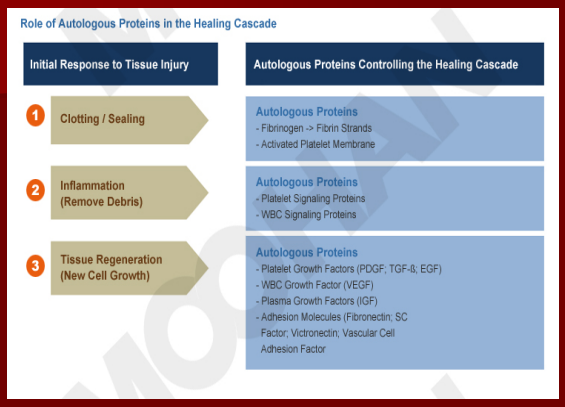
- stimulate mesenchymal stem cells and resident adult cells to replicate,
- osteoblast replication,
- endothelial cell-replication,
- fibroblast and osteoblast-replication to produce collagen,
- enhance bone-regeneration,
- stimulate matrix-formation, scaffold for tissue regeneration
- stimulation of pericytes, (As a relatively undifferentiated cell, it serves to support these vessels, but it can differentiate into a fibroblast, smooth muscle cell, or macrophage as well if required. epidermal-regeneration and re-surfacing)
- Heamostatic sealant
- Stem cell binding
- Growth factor concentration





PRP does not contain stem cells of any relevance, but may be directed at resident stem cells that are up-regulated after injection to replicate together with other mesenchymal cells.





SCIENCE BEHIND PLATELET-RICH-PLASMA (PRP) FOR AESTHETIC PURPOSES

Classic wound healing and regeneration pass through the phases of:
- haemostasis,
- inflammation,
- tissue regeneration
- and tissue remodelling (maturation).

Recent anecdotal reports indicate that it is possible to move the curve to the left by intra dermal and hypodermal injections (for facial aesthetic purposes), containing activated autologous platelets that release growth factors.

The topical efficacy, and level - evidence, has been demonstrated in the treatment of diabetic-foot ulceration. Numerous indications for PRP are described in cranio-facial surgery: bone grafting, rhytidectomy, face-lift, fat-grafting and transfer.

From a cell biology perspective, this innovative approach of administering a combination of target specific growth factors, intra and hypo dermally, addresses such issues as epithelial growth, regeneration of the ECM, epithelization and angiogenesis.

The regenerative model as mentioned before is summarised as follows:

- Formation of 3D fibrin mesh and platelet cohesion (via calcium and thrombin)
- Platelet and leukocyte release of growth factors: (PDGF, TGBF, IGF, EGF, FN, VEGF, Osteopontin).
- Local stem cell proliferation and Local tissue stem cell differentiation (and therefore enhanced tissue regeneration and tissue remodelling)
- From a PRP cell biology perspective:
  - Enhanced proliferation of fibroblasts, stem cells and myo-fibroblast
  - Enhanced restoration of the extra cellular matrix(ECM)
- Enhanced proliferation and differentiation of keratinocytes that are essential for a smooth complexion.
- Enhanced release of collagen from activated fibroblasts, thus restoring the ECM and enhancing thickening of the dermis.

REJUVENATION means to "make someone or something looks younger or lively" (Oxford Paperback Dictionary and Thesaurus 2001).

Dorland's Illustrated Medical Dictionary provides an excellent description of rejuvenation: (to become young): a renewal of youth or of strength and vigour".

So really it does not matter what cosmetic treatment you use. As long as the patient looks younger, lively and shows signs of renewal of youth, strength of vigour, then you have achieved your goal in rejuvenation.

CHALLENGES FACED BY ALL MEDISPA'S, CLINICS, AESTHETIC PRACTITIONERS WHEN REJUVENATING THE FACE, NECK, AND HANDS BY NON-INVASIVE OR MINIMALLY INVASIVE TECHNOLOGY WITH BIOLOGICALS

- Rejuvenation is a gradual process and takes months.
- Formation of new ECM, collagen and fibroblast takes months and is a very gradual process. So, keep notes to gauge process.
- Most often results of treatment are highly variable and inconsistent (includes such treatments as RF, IPL and fractional photo light therapy).
- Post inflammatory hyperpigmentation (PIH) is a major problem and headache as well as hypopigmentation, for those engaged in the use of IPL and lasers. Many sessions of treatment zone needed and maintenance therapy is strongly advised to deal with the ravages of the ongoing aging process that accelerates between the ages of 50 and 90 years, and fuelled by alcohol and tobacco. One has to rely on creams in the interim to hide the formation of lines and wrinkles.

BIGGEST CHALLENGES AT THE MOMENT FOR PATIENT:

- Premature aging of the face due to genetic make up
- Solar aging
- Damaged face due to alcohol and tobacco abuse
- Atrophy of hard and soft tissue of the face
- Loss of 3D facial structure and contours
- Sagging deep lines, due to loss of anti-gravity support of the overlying skin
- Blotches (lentigens), pigmentation, telangiectasis and melasma
- Sagging redundant skin
- Sagging neck
- Mid face ptosis

What are your primary priorities?

- Do you want to treat the pigmentation, crowfeet wrinkles or vertical/horizontal crepe lines affecting the face, chin and décolleté?
- The dyschromia may well respond initially, but melasma will return.
- At best, whatever machine is used (fractional laser IPL, or LED), a 68.5% improvement in wrinkles will be detectible at 12 weeks.
- Mid-face ptosis is different to treat in persons older than 60-years. Skin tightening is not always easy to achieve, and it does not occur after only one treatment.
- Loss of structural (collagen) integrity will test the best of therapists, and often one has to resort to filler.

BRIEF OVERVIEW OF AUTOLOGOUS PLATELET-RICH PLASMA AND ACR

- Platelet-rich derived growth factors play an important role in wound healing and the regeneration process.
- Platelet rich plasma (**PRP**) is abbreviated as **A-PRP** or **c-PRP**.
- Preparation in the side-room is possible as a high concentrate from plasma-rich plasma **(c-PRP**) for clinical application
- Concentrated PRP (**c-PRP**) can be generated from **PRP**
- Platelet concentration in **c-PRP** is higher than the patients plasma
- TGF-beta 1 can be checked in the **c-PRP**
- Dermal rejuvenation is possible via autologous cellular regeneration (**ACR)**. The process is referred to as **ACR-PRP** rejuvenation.

WHAT AUTOLOGOUS "PRP" (AUTOLOGOUS PLATELET RICH PLASMA) OFFERS REGARDING FACIAL REJUVENATION.

Quantifiable improvement of skin complexion with visible changes noticeable in 3-4 weeks-especially forehead, cheeks, neck and back of hands.
PRP is not a volumetric filler, but biological cell therapy with patient's own cells and enriched plasma.
Facial soft tissue augmentation without synthetic filler or animal products: Augments the dermis and epidermis by enhancing the growth of keratinocytes, fibroblasts and deposition of collagen.
This improves skin tone, texture and colour.
No need for costly lasers, IPL, or RF
One-off treatment, negligible down time, can ameliorate mild to moderate skin changes but needs redoing 6-12 monthly because of the ongoing chronological aging process and senescence of fibroblasts that eventually stall forming collagen and elastin due to age-exhaustion.

- Our results show that the most responsive skin areas following **PRP-mesotherapy** are the forehead and malar areas, with improvement of skin texture and tone.
- All persons injected, by the "**PRP-MESOTHERAPY"**, respond subjectively with positive improvement of skin complexion. The effects of PRP wear off at the 6 month mark. PRP does not improve pigmentation but acne scarring is responsive.
- Best PRP response is in persons of either sex, with modest wrinkling and Glogau scores, in the age group 40-60 years.
- Post-PRP outcomes are enhanced by application of vitamin-A anti wrinkle creams or LED phototherapy.
- Advanced objective digital skin analysis post-PRP mesotherapy in our laboratory, shows quantitative improvement of wrinkle micro relief, skin biometrics, and epidermal morphology. This includes improvement of peak-trough amplitude and anisotropy/topography. Botulinum Toxin administered into the glabellar region can improve the results.

- **CONTRA INDICATIONS TO THE PRP.**
- Facial cancer, past and present. This includes SCC, BCC and melanoma
- Systemic cancer, chemotherapy, steroid therapy
- Dermatological diseases affecting the face (ie porphyria)
- Blood disorders and platelet abnormalities
- Anticoagulation therapy
- Certain herbal products
- Aspirin or anti-platelet agents
- Platelet dysfunction syndrome, critical thrombocytopenia, hypofibrinogenaemia, haemodynamic instability, sepsis, acute and chronic infections, chronic liver disease, aspirin and possibly vitamin E consumption
- It is safe treatment because autologous plasma is used. Expect mild redness, swelling, bruising and minor discomfort.

FACIAL REJUVENATION WITH AUTOLOGOUS PLATELET-RICH PLASMA: PREFERRED METHOD OF A-PRP OR C-PRP DELIVERY TO SKIN DERMIS AND HYPODERMIS.

- Ameliorating facial wrinkles with IPL, RF, LED and non-ablative fractional resurfacing requires many interval treatments is labour intensive and results are usually modest and variable.
- Non-invasive face rejuvenation devices are unable to affectively address, jowls, mid-face ptosis, sagging and creasing of the forehead and neck, such as a face lift.
- Autologous platelet-rich plasma (**PRP**) offers the patient a one-off biologic treatment that lasts for 6-8 months. Results from Japan, France, Korea, Thailand, Europe, England and South Africa indicate that the aesthetic use of PRP is safe and efficacious
- Enriched platelet-rich plasma is extracted from a centrifuged venous blood specimen, as a side-room procedure and then injected intra dermally/hypodermically to induce skin epidermal and dermal rejuvenation.

MODUS OPERANDI OF FACIAL REJUVENATION PROCESS BY BIOSTIMULATION AND PRP MESOTHERAPY.

- Doctor takes a sample of venous blood from patient's arm.
- Blood is placed in a special **Collection Tube** and the PRP is processed. PRP generation takes 20 minutes.
- Cover face with Emla® cream, or equivalent, a good 30-45 minutes before injection and the face must feel numb. Local anaesthesia may still be needed.
- Multiple small jab injections are made close together into the skin releasing the PRP in order to kick-start rejuvenation by GF stimulation that is derived from the platelets in the PRP. The neck and dorsums of the hands can also be done at one session.
- Recovery is fast and expect results in 3-4 weeks. Some persons show more swelling of the skin under the injections than other patients. It is impossible to identify who will show excessive swelling.

POTENTIAL SIDE-EFFECTS OF ACR FACIAL MESOTHERAPY AND REJUVENATION

- Minimal: Expect minimal swelling, Peri-orbital swelling (reversible in all cases), bruising and redness for 12-24 hours. Some clients experience headache.
- Modest: Some patients may feel faint. A bruise at the venapuncture site may be visible for 2-3 days.
- Severe: A low morbidity procedure. In rare cases, skin cellulitis may occur which can be treated with antibiotics and cold compresses. Occasionally hospitalization is needed in these cases. Secondary skin infection is a rare complication.
- Intra-vascular injection (thrombus)
- Nerve trauma (very rare)
- Secondary infection (very rare)
- Almost no down time. No need for hospitalization. Anaphylaxis does not occur

HOW TO ENHANCE THE REGEN PROCESS OF FACIAL REJUVENATION WITH PRP

- Application of PRP plus 4-6 sessions of LED in the doctors rooms
- Avoid further sun exposure and use sun screens
- Don't be drowned in heavy moisturizers. Aging is not a lack of oil.
- Consider conditioners containing glycolic acid
- cream or glycolic formulation will enhance the results even more.
- Very acceptable and promising results have been reported from England, France and Japan and the longest follow up is about 2 years at the time of posting.

PLATELET-RICH PLASMA (PRP) AND ACR CAN AMELIORATE FACIAL ACNE SCARRING: NEW ROLE IN BIOLOGICAL REJUVENATION

Platelet-rich plasma (PRP) has been utilized in aesthetic medicine (UK, Japan, and Asia, Europe) to rejuvenate and ameliorate the aging process and face. This refers to mesenchymal and epithelial rejuvenation by application of the persons own enriched autologous plasma. Treatment of acne scarring is, however, challenging for the aesthetic physician and dermatologist, especially in pigmented skins.

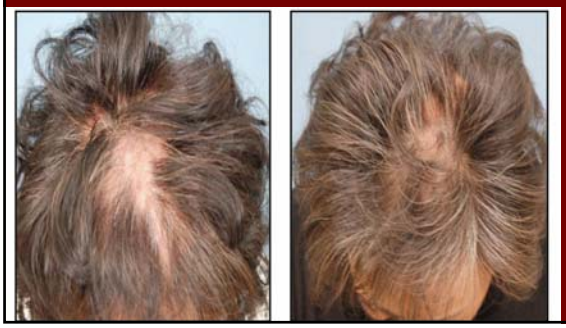
WRINKLES:

Aging of the human face is a given and something we have to live with. The process is gradual, starting at the age of 25 years and progresses rapidly after 60 years. Most people, of both genders, choose to age gracefully.

However, in the era of anti-aging medicine, the tendency is to turn back the clock and undergo some form of rejuvenation to reduce facial wrinkles, lines and blemishes. For decades, rejuvenation of the face by surgical face-lift has been popular.

Today, more non-invasive treatments are available to improve the skin complexion of the aging face. This is because people want to look better with as little as possible down time and morbidity. There are newer treatment options available to ameliorate aging aspects that affect the face, neck, décolleté and the back of the hands. For decades the backbone of facial rejuvenation has been massage, facials, and cosmeceuticals including moisturisers, sunscreens, micro-dermabrasion, serums and foundation cosmetics. Botulinum and fillers have also become popular. Currently, popular methods to rejuvenate, tighten and resurface the face include radiofrequency, non-ablative photo-thermolysis, IPL's and light-emitting diode (LED) phototherapy. Also popular is combination therapy consisting of IPL, laser and cosmeceuticals, including vitamin A derivatives.

whatever option is used, the aging process cannot be prevented and skin improvement by these treatments is gradual, subtle, modest and variable. Avoidance of excessive sunlight is important to prevent accelerated photo-aging, blotches, lentigenes and skin cancer. A new approach, appealing to clients seeking a more natural approach to facial rejuvenation, is rejuvenation with your own cells and more specifically, autologous platelet-rich plasma **(PRP)**. This is also referred to as autologous cellular regeneration (**ACR)** an aesthetic practise, is a safe and tolerable procedure.



- **AUTOLOGOUS PLATELET-RICH PLASMA (A-PRP) CAN AMELIORATE ACUTE SPORTS INJURIES SUCH AS HAMSTRING INJURIES, AND OTHER MUSCULO-TENDINOUS TEARS.**

- Autologous platelet-rich plasma (A-PRP) refers to clinical applicable cell therapy with the patient's own plasma enriched with platelets and release of growth factors such as PDGF, TGF, EGF AND VEGF once the PRP is activated with calcium chloride.
- Will injected **A-PRP** be able to stimulate skeletal muscle myoblasts/ satellite cell regeneration to facilitate the healing process? The answer is yes, because ex vivo tissue culture studies show that PRP is a potent stimulator for the proliferation of myoblasts and satellite cells that play key roles in the regeneration and healing of skeletal muscle. It has been suggested that **A-PRP** is more effective than steroid injection in inducing pain relief. The reason is that the **A-PRP** has anti-inflammatory effects as well as growth factor enhancement. Skeletal muscle myoblasts or satellite cells (progenitor cells) can be generated in monolayer culture when skeletal muscle is co-cultured ex vivo in **PRP-enriched media** using advanced tissue culture (TC) technologies.

- This is of considerable interest and relevant to sports science physicians and biokineticians that deal with muscle and tendon injuries that pose significant challenges in athletes (in terms of healing time and recurrence of the injured tendon/muscle).

- **PRP**, if correctly generated, can release TGF,PDGF, EGF and VEGF, which together with myriads of other factors, is important in the wound healing cascade Theoretically then, **PRP** injected carefully and under controlled conditions, taking into account all the relevant local anatomical structures, into injured muscles and musculo-tendinous junctions , can stimulate proliferation of satellite cells (myoblasts: progenitor cells that regenerate adult skeletal muscle) in situ at the injection point or point of care.
- This cell-based treatment option, based on platelet-derived growth factors, has been sporadically applied with success in conjunction with conventional strategies such as RICE, biokinetics and physiotherapy.

- **REFERENCES ON A-PRP:**

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- **Brown GL** et al. Enhancement of wound healing by topical treatment with epidermal growth factor. N Engl J Med 321: 76-79, 1989
- **Chajchir A** et al. Growth factors in Plastic Surgery. Aesth. Plast. Surg. 29: 295-299, 2005
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- **Sanchez M et al**. Comparison of surgically repaired Achilles tendon tears using platelet –rich fibrin matrices (Am J Sports Med 2007, 245-51)

- **Relevant scientific publications regarding PRP, tissue culture, translational research regarding musculo-tendinous injuries are listed below:**

- **Acta Orthop** 2006:Tendon repair can be stimulated by thrombin and platelets. Currently, in the RSA, a new device enables the rapid side-room preparation (REGEN-PRP-THT), with unsurpassed high platelet yield and growth factors. New reports indicate that autologous–PRP (A-PRP) may suppress harmful cytokine release, limit inflammation and thereby promote tissue, muscle and tendon regeneration.
- **Anitua** et al 2005, from Spain, reported that autologous PRP may be beneficial to the treatment of tendon injuries by inducing cell proliferation and promoting the synthesis of angiogenic factors during the healing process (J ORTOP RES 2005).
- **de Mos** et al 2008Human tenocytes in PRP driven tissue culture can stimulate cell proliferation and total collagen production
- **J Orth Res** 2005Autologous biological production of plasma rich in GF promote proliferation and induce VEGF and HGF production by human tendon cells in culture.
- **Kajikawa** et al 2008: In experimental models, PRP can enhance Type I and III collagen immuno reactivity in early phase tendon healing. This suggests that locally injected PRP may well enhance the initial tendon healing process by increased cellular proliferation and collagen production.

- **Murray** et al 2007Collagen-platelet rich plasma hydrogel enhances primary repair of porcine anterior cruciate ligament
- **Sariguney** et al 2008: Activated platelets release various GF, some of which are recognized to improve nerve regeneration (): see results of sciatic nerve re-myelinization in animal models.

- **Schnabel** et al 2007: PRP can enhance anabolic gene expression patterns in flexor digitorum superficialis tendons. Tendons cultured in 100% PRP show enhanced gene expression of matrix molecules such as COL 1A1, COL3A1, and Comp.
- **Virchenko** et al 2006Injection of PRP, can improve repair in various rat Achilles tendon transsection models.