Injectable Orthobiologics: Hope or Hype?
Disclosures

- **Harvest**
  - International Consulting Agreement and Educational Speaker
- **Sonosite**
  - Educational Speaker
- **Tenex**
  - Minor investor
Outline

- Pathophysiology of Platelet Rich Plasma (PRP)
- PRP for Tendons
- PRP for Joints
- Beyond PRP
  - MSC
  - ADSC
  - IRAP
  - PDSC
Proliferative therapy (prolotherapy) was first described in the 1930’s.

Theory is that irritant solutions (classically dextrose) along with needling of soft tissues stimulates an inflammatory reaction which initiates a healing cascade for injured soft tissues.

This technique has been employed for chronic enthesopathies and ligamentous injuries/laxity.
Evolution of Orthobiologic Injections

- **1930’s**
  Kellgren, Gedney, Schultz described their use of injecting shoulders, necks, knees, backs, and TMJ with sclerosing solutions.


- **1941**
  Shuman described the use of prolotherapy for shoulder dislocations.

  Shuman D. Luxation recurring in the shoulder. *Osteopath Prof* 1941;8:11-14.

- **1954**
  Shuman published his results showing improvement in 75% of the cases he treated (SI, knee, back, shoulder)

Evolution of Orthobiologic Injections

• 1961
  George Hackett published a monograph based on his prolotherapy experience.
  1,857 LBP patients showed a cure rate of 82% followed from 2-12 years
  No Complications

And more...

1974 – Hemwall
- 2,007 patients treated with prolotherapy
- 6,000 treatments performed on 1,871 patients
- 75.5% (1,399) patients “cured”
- 24.3% (413) reported improvement
- 0.2% (25) reported no improvement

1980’s – Ongley
Treated LBP with manipulation + prolo or manipulation + saline
87% of the prolotherapy group improved.
39% of the saline group improved.

Evolution of Injectable Platelet Rich Plasma (PRP) in MSK medicine

- First use of PRP in US was in 1987 following open heart surgery
- Periodontal and wound healing were early successful clinical applications of PRP
- Bench work research demonstrated proliferation of GF’s with supraphysiologic amount of platelets
- Equine experiences with tendon regeneration with PRP
140 pts evaluated for lateral epicondylosis
- 20 had refractory pain an avg. of 15 months later
15 in treatment group, 5 in bupivacaine control group

Intervention
- Injection w/ autologous PRP once into common extensor tendon followed by gradual increase in rehab program through 4 weeks after which full activity allowed

Outcome
- A 46%, 60% and 81% improvement in VAS pain scores at 1, 2 and 6 months respectively in tx group
- 3/5 in bupivacaine group withdrew/ sought other tx
- At final F/U (12-38 months) 93% pain free (<10/100 VAS)
- No complications, no one got worse
What is PRP?

- Increase in platelets from baseline
  - Liquid
  - Injectable
Why Platelets?

1. Tissue injury
2. Coagulation Cascade
3. Inflammation: macrophage, neutrophil (natural debridement)
4. Fibroblast Activity
5. Tissue granulation, Collagen synthesis
6. Wound Contraction
7. Epithelization
8. Remodelling
9. Healed tissue
Why do we need to concentrate platelets?

- Studies have shown accelerated wound healing requires at least 4x-5x platelet concentration.
- An exponential increase in cell proliferation occurs as platelet concentration increases from 2.5x to 5x-10x baseline levels.
- Much lower volume needed to get high levels of platelets to area.
Why are we still debating if it works?

- **Need to define what is in the PRP**
  - Platelet concentration
  - Leukocyte count/presence
  - RBC +/- RBC -
- **Need to define the procedure**
  - US guidance
  - Needle tenotomy
    - How many times?
  - Activators used
- **Rehabilitation methods**
  - Need to be studied/validated
  - Immobilization
  - Timing of eccentrics
- **May need to separate out different body parts**
  - Do elbow tendons behave differently than Achilles tendon?
PRP for Tendinopathy
The continuum of tendinopathy.
PRP for Achilles Tendinosis

de Vos et al, JAMA, January 13, 2010
- **First** double blind, placebo controlled, RCT on PRP
- 54 randomized patients age 18 to 70 with chronic (at least 2 mo) achilles tendon pain 2 to 7 cm above calcaneus
- Either 6cc PRP or Saline was injected with US guidance into achilles tendon
- Rehab for both groups involved rest and then after 2 weeks, started on 12 week daily (180 repetitions) eccentric exercise program
- No sports for at least 4 weeks and then only if pain <= 3/10
- f/u questionnaire at weeks 6,12,24
PRP for Achilles Tendinosis

- de Vos et al, JAMA, January 13, 2010
  - **RESULTS**
    - After 24 weeks, no statistical difference between the 2 groups
    - Both groups improved > 20 pts on VISA-A scores (0-100)
    - No adverse events in either group (ruptures/ infections)
  - **Discussion**
    - Eccentrics done on both groups (not done b4 PRP)
      - High success rate in treating tendinosis
    - Eccentrics done early (started at 2 wks)
    - Both groups improved
    - Only 1 treatment done
PRP vs Steroid Injection for Lateral Epicondylitis

- Peerbooms et al, AJSM, February 2010
- Double blind RCT with 1 year follow up of 100 pts
- No Ultrasound guidance was used
- Success defined as >25% reduction in VAS or DASH score

**RESULTS**

- At 1 yr, 49% of CSI group and 73% in PRP group were successful (p<.001)

<table>
<thead>
<tr>
<th>Time (wks)</th>
<th>CS Average ± SD</th>
<th>PRP Average ± SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>VAS</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>65.8 ± 13.8</td>
<td>70.1 ± 15.1</td>
</tr>
<tr>
<td>4</td>
<td>44.2 ± 26.4</td>
<td>55.4 ± 24.2</td>
</tr>
<tr>
<td>8</td>
<td>42.9 ± 29.2</td>
<td>46.9 ± 24.9</td>
</tr>
<tr>
<td>12</td>
<td>44.2 ± 27.1</td>
<td>38.7 ± 27.2</td>
</tr>
<tr>
<td>26</td>
<td>56.6 ± 23.2</td>
<td>32.6 ± 31.5</td>
</tr>
<tr>
<td>52</td>
<td>50.1 ± 28.1</td>
<td>25.3 ± 31.2</td>
</tr>
</tbody>
</table>
PRP vs Steroid Injection for Chronic Lateral Epicondylitis

- Gosens, Peerbooms et al; *AJSM*, March, 2011
- 2 yr f/u of data showed sustained benefit
**DESIGN**
- Randomized, Double-Blind, Placebo-Controlled Trial

**METHODS**
- N = 60
- PRP vs Saline vs glucocorticoid (+ Lidocaine)
- Primary end point - change in pain using Patient-Rated Tennis Elbow Evaluation (PRTEE) at 3 months
- Secondary Outcomes - were ultrasonographic changes in tendon thickness and color Doppler activity

**OUTCOMES**

1. **PRTEE Pain score**
   - Reduction at 3 months in all 3 groups with no statistically significant difference between the groups;
   - At 1 month glucocorticoid reduced pain more effectively than did both saline and PRP

2. **PRTEE Disability score**
   - At 1 month glucocorticoid was superior to saline and PRP
   - At 3 months no difference

3. **US eval (tendon thickness and color doppler)**
   - Glucocorticoid was more effective than PRP and saline in reducing color Doppler activity and tendon thickness

4. **Pain associated with INJXN**
   - PRP = 9 > Saline = 7 > CSI = 6
Neither injection of PRP nor glucocorticoid was superior to saline with regard to pain reduction in LE at 3 months.
Comparison of studies

Results of PRP can not be adequately measured with only 3 months follow-up

PRP vs CSI VAS Scores

Peerbooms, AJSM, 2010 & Gosen AJSM, 2011
Is it the Needle?

Platelet-Rich Plasma Significantly Improves Clinical Outcomes in Patients With Chronic Tennis Elbow

A Double-Blind, Prospective, Multicenter, Controlled Trial of 230 Patients

Allan K. Mishra,† MD, Nebojsa V. Skrepnik,‡ MD, PhD, Scott G. Edwards,§ MD, Grant L. Jones,‖ MD, Steven Sampson,¶ DO, Doug A. Vermillion,§ MD, Matthew L. Ramsey,‖ MD, David C. Karli,‡‡ MD, MBA, and Arthur C. Rettig,‡‡ MD

Investigation performed at Department of Orthopaedic Surgery, Menlo Medical Clinic, Stanford University Medical Center, Menlo Park, California

AJSM, 2013
Improvement in Pain Scores at 24 Weeks (p =0.027)

<table>
<thead>
<tr>
<th></th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>56.1%</td>
</tr>
<tr>
<td>PRP</td>
<td>71.5%</td>
</tr>
</tbody>
</table>
Patients Reporting Significant Elbow Tendernessness

- Control
  - Injection: 93%
  - 4 wks: 65%
  - 8 wks: 56%
  - 12 wks: 48%
  - 24 wks: 54%

- PRP
  - Injection: 93%
  - 4 wks: 52%
  - 8 wks: 43%
  - 12 wks: 37% *
  - 24 wks: 29% **

* p = 0.036
** p < 0.001
PRP vs PNT for RTC pathology

- Rha et al; *Clinical Rehabilitation*, 2012
- DBRCT comparing 2 PNT vs 2 PRP injections under US guidance for RTC tendinosis or small, partial tear
- Measured results using Shoulder Pain and Disability Index at baseline, 2wks after 1st injection, right before second injection, 2 wks after second injection, 3 months, 6 months
- Pts age 16-70 (avg 48 yrs)
- Greater than 6 months of pain (avg 36 months)
- Diagnosed by clinical exam plus MRI or diagnostic US
- Failed conventional treatments
  - Medications
  - Bracing
  - Stretching
  - Strengthening
  - CFM
  - Modalities
- PRP done under US guidance
Data of PRP procedures

- 325 patients with isolated tendinopathy sent questionnaire
  - Eliminated all non tendons and regional PRP treatments
- 180 responded (55%)
- All retrospective data analyzed with following questions:
  - Overall improvement
    - Not at all, slightly, moderately, mostly, completely
  - VAS score – pre and post
  - Overall satisfaction
  - Nirchl phase scale
<table>
<thead>
<tr>
<th>Tendon Type</th>
<th>Count</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lateral Epicondyle</td>
<td>30</td>
</tr>
<tr>
<td>Patella Tendon</td>
<td>27</td>
</tr>
<tr>
<td>Achilles</td>
<td>27</td>
</tr>
<tr>
<td>Rotator Cuff</td>
<td>21</td>
</tr>
<tr>
<td>Hamstring</td>
<td>17</td>
</tr>
<tr>
<td>Gluteus Medius</td>
<td>16</td>
</tr>
<tr>
<td>Medial Epicondyle</td>
<td>11</td>
</tr>
<tr>
<td>Plantar Fascia</td>
<td>9</td>
</tr>
<tr>
<td>13 other tendons</td>
<td>&lt;5 each</td>
</tr>
</tbody>
</table>
Overall Improvement

82% reported moderate to complete improvement – 50%- 100% relief of symptoms
70 % reported mostly to complete improvement -- 75-100% relief of symptoms
Pain Score pre and post PRP

➤ 74% Reduction in VAS
> 50% Improvement

Moderate improvement to complete resolution

PRP for Cartilage Disease/Osteoarthritis
Define the problem

- 18 million Americans are currently living with symptomatic knee OA.
  - 4.1 million Americans have difficulty with ambulation, therefore have failed conservative treatment, and are candidates for knee arthroplasty or high tibial osteotomy (HTO).
- 500,000 knee arthroplasties and HTOs are performed annually in the United States.
- Therefore, **3.6 million Americans** linger in a treatment gap, as they are unwilling or not a candidate to undergo arthroplasty or HTO, and remain there for an average of 20 years.

Osteoarthritis is characterized by an imbalance of anabolic and catabolic process in synovial joints, resulting in progressive cartilage damage.

PRP Properties:
- increase anabolic effect on chondrocytes
- decrease catabolic effect in the inflammatory environment
- Cartilage integrity maintained by a balance from cytokine-driven catabolic and anabolic processes

- Increases growth factors IGF, TGF

- IGF - precursor to GH

- IL-1
  - TNF-alpha
  - IL-1 Ra
  - Anticatabolic

- IL-4, 10, 13
  - PDGF, IGF, TGF, VEGF

- Increases growth factors IGF, TGF
  - Chondroprotectant
  - IGF-precursor to GH
Wang-Saegusa et al

- 312 patients with Knee OA (Outerbridge I-IV)
- >3 months symptoms
- 3 injections PRGF at 2 week intervals

Results:
- At 6 months patients reported significant improvement in pain, stiffness and function

(Archives Orthopaedic Trauma Surgery 2010)
Spakova et al, AJPMR, April 2012
- 120 pts received either 3 PRP or 3 HA injections
  - PRP group with 4.5x baseline count

![Graph showing NRS Scores and WOMAC Score changes over time for PRP and HA groups.](image)
RCT comparing PRP vs HA

55 patients in each group
- Received weekly IA injections x3
  - PRP was leukocyte rich
- Evaluated up to 12 months post injection

Overall, clinically significant improvement
- But not statistically significant

HOWEVER......
Platelet-rich plasma vs hyaluronic acid to treat knee degenerative pathology: study design and preliminary results of a randomized controlled trial.
Comparison Between Hyaluronic Acid and Platelet-Rich Plasma, Intra-articular Infiltration in the Treatment of Gonarthrosis

Fabio Cerza,† MD, Stefano Carni,‡ MD, Alessandro Carcangi,⁎ MD, Iginio Di Vavo,⁎ MD, Valerio Schiavilla,⁎ MD, Andrea Pecora,⁎ MD, Giuseppe De Biasi,‖ and Michele Ciuffreda‖

Investigation performed at the Paolo Colombo Hospital of Velletri, Rome, Italy

AJSM, 2013

TABLE 1
Demographic Information

<table>
<thead>
<tr>
<th></th>
<th>ACP Group</th>
<th>HA Group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Average age (SD), y</td>
<td>66.5 (11.3)</td>
<td>66.2 (10.6)</td>
</tr>
<tr>
<td>Male patients, No. (%)</td>
<td>25 (42)</td>
<td>28 (47)</td>
</tr>
<tr>
<td>Female patients, No. (%)</td>
<td>35 (58)</td>
<td>32 (53)</td>
</tr>
<tr>
<td>Left knee, No.</td>
<td>17</td>
<td>12</td>
</tr>
<tr>
<td>Right knee, No.</td>
<td>43</td>
<td>48</td>
</tr>
<tr>
<td>Gonarthrosis, No. (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Grade I</td>
<td>21 (35)</td>
<td>25 (42)</td>
</tr>
<tr>
<td>Grade II</td>
<td>24 (40)</td>
<td>22 (37)</td>
</tr>
<tr>
<td>Grade III</td>
<td>15 (25)</td>
<td>13 (21)</td>
</tr>
</tbody>
</table>

⁎ACP, autologous conditioned plasma; HA, hyaluronic acid; SD, standard deviation.

Figure 2. Mean Western Ontario and McMaster (WOMAC) scores for the autologous conditioned plasma (ACP) group in relation to degree of gonarthrosis.

Figure 3. Mean Western Ontario and McMaster (WOMAC) scores for the hyaluronic acid (HA) group in relation to degree of gonarthrosis.
Treatment With Platelet-Rich Plasma Is More Effective Than Placebo for Knee Osteoarthritis: A Prospective, Double-Blind, Randomized Trial
Sandeep Patel, Mandeep S. Dhillon, Sameer Aggarwal, Neelam Marwaha and Ashish Jain
DOI: 10.1177/0363546512471299

Enrollment
Assessed for eligibility (n = 150 patients)
Excluded (n = 72 patients):
- Not meeting inclusion criteria (n = 54)
- Declined to participate (n = 18)
- Other reasons (n = 0)
Randomized (n = 78 patients)

GROUP A
Allocated to intervention (n = 27 patients; 54 knees):
- Received allocated intervention (n = 27 patients; 54 knees)
- Did not receive allocated intervention (n = 0)

1 injection
- Lost to follow-up (n = 1 patient; 2 knees): underwent TKR elsewhere.
- Discontinued intervention (n = 0)
- Analyzed (n = 26 patients; 52 knees)
- Excluded from analysis (n = 0)

GROUP B
Allocated to intervention (n = 26 patients; 50 knees):
- Received allocated intervention (n = 25 patients; 50 knees)
- Did not receive allocated intervention (n = 0)

2 injections
- Lost to follow-up (n = 0)
- Discontinued intervention (n = 0)
- Analyzed (n = 25 patients; 50 knees)
- Excluded from analysis (n = 0)

GROUP C
Allocated to intervention (n = 26 patients; 52 knees):
- Received allocated intervention (n = 23 patients; 46 knees)
- Did not receive allocated intervention (n = 3; 6 knees) [later refused for treatment when reported for injection]

- Lost to follow-up (n = 0)
- Discontinued intervention (n = 0)
- Analyzed (n = 23 patients; 46 knees)
- Excluded from analysis (n = 0)
Treatment With Platelet-Rich Plasma Is More Effective Than Placebo for Knee Osteoarthritis: A Prospective, Double-Blind, Randomized Trial
Clinical and MRI Outcomes After Platelet-Rich Plasma Treatment for Knee Osteoarthritis

Brian Halpern, MD, Salma Chaudhury, MD, PhD, MRC, Scott A. Rodeo, MD, Catherine Hayter, MD, Eric Bogner, MD, Hollis G. Potter, MD, and Joseph Nguyen, MPH

- 17 patients/ 18 knees
  - Kellgren 0-II, with knee pain
- Prospective - 1 PRP Injection
  - Cascade system used (low WBC, lower plt)
- MRI data collected pre/ 1 yr post procedure on 15 patients
- Functional outcomes assessed at 1 wk, 1,3,6 and 12 months
Clinical and MRI Outcomes After Platelet-Rich Plasma Treatment for Knee Osteoarthritis

- **Results**
  - 56% reduction of VAS at 6 mo, 59% at 1 year
  - 24% improvement in function at 1 year
  - 24% improvement in ADL’s at 1 year
  - WOMAC pain scores reduced by 41% at 6 mo and 56% at 1 year
  - Overall 73-83% of patients with repeat MRI had no worsening of MRI changes in lat, med, PF comp.
    - Studies suggest 4-6% volume loss per year of cartilage thickness
Talus OLT
PRP vs HA

- Mei-Dan et al, AJSM, Jan 2012
- 32 patients, randomized to HA vs PRP
  - 3 injections weekly
  - PRP 2-3 x platelet concentrate

### Outcome Measures: Time by Group Interaction

<table>
<thead>
<tr>
<th>Outcome Measure</th>
<th>Week</th>
<th>Group 1: HA</th>
<th>Group 2: PRP</th>
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</thead>
<tbody>
<tr>
<td>AHFS&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0</td>
<td>66.4 (15)</td>
<td>68.0 (14)</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>78.2 (14)</td>
<td>84.3 (7)</td>
</tr>
<tr>
<td></td>
<td>12</td>
<td>81.2 (14)</td>
<td>89.7 (7)</td>
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<tr>
<td></td>
<td>28</td>
<td>78.3 (14)</td>
<td>92.5 (8)</td>
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<tr>
<td>VAS pain</td>
<td>0</td>
<td>5.6 (1.7)</td>
<td>4.1 (2.1)</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>3.7 (1.5)</td>
<td>1.6 (1.5)</td>
</tr>
<tr>
<td></td>
<td>12</td>
<td>3.0 (2.1)</td>
<td>0.9 (1.0)</td>
</tr>
<tr>
<td></td>
<td>28</td>
<td>3.1 (2.1)</td>
<td>0.9 (1.4)</td>
</tr>
<tr>
<td>VAS stiffness&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0</td>
<td>5.1 (2.8)</td>
<td>5.0 (2.3)</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>3.2 (2.0)</td>
<td>2.5 (1.7)</td>
</tr>
<tr>
<td></td>
<td>12</td>
<td>3.0 (2.4)</td>
<td>1.4 (1.8)</td>
</tr>
<tr>
<td></td>
<td>28</td>
<td>2.9 (2.3)</td>
<td>0.8 (1.2)</td>
</tr>
<tr>
<td>VAS function&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0</td>
<td>5.8 (1.9)</td>
<td>4.7 (2.1)</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>4.0 (2.0)</td>
<td>2.2 (1.4)</td>
</tr>
<tr>
<td></td>
<td>12</td>
<td>3.5 (2.5)</td>
<td>1.1 (1.1)</td>
</tr>
<tr>
<td></td>
<td>28</td>
<td>3.5 (2.6)</td>
<td>0.8 (1.2)</td>
</tr>
<tr>
<td>Subjective global function&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0</td>
<td>56 (18)</td>
<td>58 (22)</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>67 (18)</td>
<td>79 (14)</td>
</tr>
<tr>
<td></td>
<td>12</td>
<td>71 (21)</td>
<td>90 (9)</td>
</tr>
<tr>
<td></td>
<td>28</td>
<td>73 (20)</td>
<td>91 (10)</td>
</tr>
</tbody>
</table>
Sanchez et al

- 45 pts with moderately severe hip OA
- US Guided Hip Injection weekly x 3 wks

Results
- 57.5% had clinically relevant reduction in pain
- 40% were excellent responders (pain reduction by 6-7 wks)
  - Which was sustained at 6 months

*Rheumatology, November, 2011*
Conclusion of PRP and OA

- PRP looks promising as a tool for the management of knee OA
  - When compared to Hyaluronic acid, PRP seems to have improved outcomes.
  - Conflicting results based on degree of OA
- Pain and function is improved clinically best at 6 months for those treated with PRP but also at 1 yr
  - Might serve as preventative for further damage
  - Unlikely to “regrow cartilage” in the knee
  - PRP likely mechanism is an increase anabolic effect on chondrocytes and a decrease catabolic effect in the inflammatory environment
    - May prevent Apoptosis of chondrocytes.
Beyond PRP

BMC
ADSC
PDSC
IRAP
Beyond PRP

- PRP is likely just the tip of the iceberg in regenerative medicine
- Stem cells are an emerging form of regenerative medicine
  - Currently... FDA will allow removal of stem cells and re-injection the same day with MINIMAL manipulation of the cells
- Some suggest better results can be obtained with culturing and expanding these cell lines.
  - Some concern for tumors (not likely)
  - Some concern for diminished efficacy with storing cells before re-injection
Beyond PRP

**Mesenchymal Stem Cells**
- These are the adult stem cells that help rebuild bone, muscle, tendon, cartilage
- They act as the repairman in the body
  - Growth factor’s are the site managers
- 2 most common location for MSC’s are
  - Bone Marrow
  - Fat
How MSCs Work

It is not clear how MSCs work however they are able to go to sites of injury to help with tissue restoration

- MSCs secrete an array of cytokines and chemokines involved with immunomodulation, anti-inflammation, antiapoptosis, proangiogenesis, proliferation, and chemoattraction

- MSCs orchestrate the process of differentiation with differentiated and undifferentiated cells

Caplan AI, Dennis JE. Mesenchymal stem cells as trophic mediators. Journal of cellular biochemistry. 2006 Aug 1;98(5):1076-84.
Two Different Types of Bone Marrow Stem Cell Processes

Bone Marrow Nucleated Cell Isolation
The stem cell fraction of bone marrow is isolated via a centrifuge and re-injected the same day.

The stem cell fraction of bone marrow is isolated via a centrifuge and re-injected the same day.

Bone Marrow Mesenchymal Stem Cell Culture
The stem cells themselves are isolated and cultured to greater numbers over a few weeks. This produces a “pure” population of stem cells which is different than the mix of cells produced by same day procedures.

Bone Marrow Mesenchymal Stem Cell Culture
The stem cells themselves are isolated and cultured to greater numbers over a few weeks. This produces a “pure” population of stem cells which is different than the mix of cells produced by same day procedures.

Three Different Types of Fat Stem Cell Processes

Same Day

Simple Adipose Graft
The fat is separated from the oil and liquid and the fat is injected (however the stem cells are still trapped in the fat and are not concentrated).

Stromal Vascular Fraction (SVF)
The fat is separated and then chemically digested to release the stem cell fraction, which is then concentrated.

Advanced

Adipose Mesenchymal Stem Cell Culture
The stem cells are isolated and cultured to greater numbers over a few weeks. This produces a “pure” population of stem cells which is different than the mix of cells produced by same day procedures.

Non-adherent Cells
MSCs adhere to the plastic

Fat
Liquid

Stem Cells

Non-adherent Cells
MSCs adhere to the plastic

Fat
Liquid

Stem Cells

Non-adherent Cells
MSCs adhere to the plastic

Fat
Liquid

Stem Cells

Non-adherent Cells
MSCs adhere to the plastic

Simple Adipose Graft
The fat is separated from the oil and liquid and the fat is injected (however the stem cells are still trapped in the fat and are not concentrated).

Stromal Vascular Fraction (SVF)
The fat is separated and then chemically digested to release the stem cell fraction, which is then concentrated.
Bone Marrow Aspirate Concentrate
BMAC
• Anesthetize periostium of pelvis with lidocaine/marcaine
• Can use US or fluoro guidance
• Tap trochar into bone marrow with hammer

• Withdraw BMAC using 60 cc syringe.
• Slowly aspirate and withdraw BMAC to maximize stem cell concentration
MSC’s

Human Mesenchymal Stem Cells in Bone Marrow

MSC / Marrow Cells

0.000001

0.0000008

0.000006

0.000004

0.000002

0.00000002

0.00000001

Age

Newborn
Teen
35 yrs
50 yrs
80 yrs

1/10,000

1/100,000

1/250,000

1/400,000

1/2,000,000
Adipose Derived Stem Cells
50 year old active male with 1.5 year history of R shoulder pain
Told by ortho. he has full thickness RTC and needs surgery
Fat vs BMAC

2000 ml of fat
50 million nucleated cells/100 ml of fat

Stem cells per cc Tissue

Knee OA treated successfully with MSCs using platelet lysate technique
- There was a decrease in the VAS by 95%
- Increase in ROM by 5 degrees
- Increase in cartilage by 19% as measured by MRI at 3 months post procedure.

Case Reports in Humans

Pre-injection cartilage

6 months Post-injection cartilage

meniscus

Roi-1

Roi-2

Roi-1

Roi-2
Human Studies

- Prospective study of 6 females who underwent injection of MSCs without addition of growth factors
  - 1 year follow up decrease in mean pain on VAS, joint function and walking distance compared to baseline
    - No adverse events reported
  - 3 out of 6 showed improvement on MRI in cartilage thickness, extension of repair tissue over subchondral bone, and decrease subcondral edema

Injectable BMC in Knee OA

Patient Demographics

<table>
<thead>
<tr>
<th></th>
<th>Regenexx-SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>N</td>
<td>184 patients</td>
</tr>
<tr>
<td>Male</td>
<td>134 (73%)</td>
</tr>
<tr>
<td>Female</td>
<td>50 (27%)</td>
</tr>
<tr>
<td>Age</td>
<td>52 (19-83)</td>
</tr>
<tr>
<td>Height</td>
<td>69” (60-80)</td>
</tr>
<tr>
<td>Weight</td>
<td>174 (100-295) lbs.</td>
</tr>
<tr>
<td>BMI</td>
<td>25.5 (17.5-39.8)</td>
</tr>
</tbody>
</table>

For both graphs above: N=86 at 4-6 weeks (47% reporting), n=47 at 3 mo (29% reporting), 79 at 6 mo (55% reporting), n=48 at 12 mo (55% reporting), n=35 at 18 mo (58% reporting). For example, 55% reporting means that only about half of the patients treated answered our questionnaires.
## Injectable Stem Cells Vs. Knee or Hip Arthroplasty

<table>
<thead>
<tr>
<th></th>
<th>Stem Cells</th>
<th>Knee/Hip Arthroplasty</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pain associated with procedure</td>
<td>Minimal</td>
<td>Considerable</td>
</tr>
<tr>
<td>Post procedure recovery time</td>
<td>Minimal</td>
<td>Considerable</td>
</tr>
<tr>
<td>Joint Infections</td>
<td>0.0% (reports of 2 patients with infections at bone marrow draw sites)</td>
<td>0.72%</td>
</tr>
<tr>
<td>DVT/PE</td>
<td>0.0%</td>
<td>as high as 3.76%/1.19% (with anticoagulation)</td>
</tr>
<tr>
<td>Nerve injury</td>
<td>0.0%</td>
<td>Up to 0.03%</td>
</tr>
<tr>
<td>Limb length discrepancy</td>
<td>0.0%</td>
<td>Up to 62%</td>
</tr>
<tr>
<td>Cost of procedure</td>
<td>Up to $11,000</td>
<td>Up to $35,000/$44,816</td>
</tr>
<tr>
<td>Need for Revision</td>
<td>Unknown</td>
<td>5,082/7,852 in UK in 2010</td>
</tr>
</tbody>
</table>
THR vs BMAC

Mitchell, Sheinkop et al, awaiting publication

Mean Harris Hip Score

- THA Preop (101) 56
- THA Post (24) 94
- BMAC Preop (28) 68.75
- Post BMAC (18) 82.89

Harris Hip Pain Score

- THA
  - Pre op
  - Mod to marked: 77%
  - Post op
  - None: 80.0%
- BMAC
  - Pre op
  - Moderate: 93%
  - Post op
  - None: 25%
  - Slight: 50%
  - Moderate: 25%
TKR vs BMAC

Mitchell, Sheinkop et al, awaiting publication

Knee Society **Assessment Score**

- TKA
  - Pre op 48
  - Post op 80
- BMAC
  - Pre op 69.08
  - Post op 82.44

Knee Society **Function Score**

- TKA
  - Pre op 40
  - Post op 73
- BMAC
  - Pre op 71.15
  - Post op 90.31
## TKR vs BMAC

### Physician Global Assessment of the Knee

<table>
<thead>
<tr>
<th></th>
<th>Pre BMAC</th>
<th>Post BMAC</th>
</tr>
</thead>
<tbody>
<tr>
<td>None</td>
<td>0%</td>
<td>38%</td>
</tr>
<tr>
<td>Mild</td>
<td>35%</td>
<td>50%</td>
</tr>
<tr>
<td>Moderate</td>
<td>65%</td>
<td>12%</td>
</tr>
<tr>
<td>Severe</td>
<td>0%</td>
<td>0%</td>
</tr>
<tr>
<td>Extreme</td>
<td>0%</td>
<td>0%</td>
</tr>
</tbody>
</table>

- **None:** no pain, no symptoms and no limitation of function
- **Mild:** mild pain, mild symptoms and mild limitation of function
- **Moderate:** moderate pain, moderate symptoms and moderate limitation of function
- **Severe:** severe pain, severe symptoms and severe limitation of function
- **Extreme:** extreme pain, extreme symptoms and extreme limitation of function

*88%*
TJA vs BMAC Conclusions

- After TJA, Patients decrease athletic participation and intensity and also diminish types of activity
- After BMAC Patients restore and increase athletic participation and intensity after BMAC
- Knee patients overall, had better outcomes vs Hip patients, esp. compared to surgical treatment but both groups did well

Mitchell, Sheinkop et al, awaiting publication
Placenta derived MSC’s

- In last 2-3 years, placenta has been discovered as source of MSC
- Only starting to consider orthopedic use of this treatment
- Very high concentration of GF’s and some literature to support use
- Very Sparse orthopedics literature
IRAP

- **Orthokine/ Regenokine**
  - Available in Europe as there is storage and manipulation of cells so controversial in US
  - Growth factors all released as bolus due to preactivation of platelets
  - Targets IL’s, especially IL-1 RA
    - IL-1 is main cytokine associated with catabolism in knee joint and OA production
      - May be better for prevention/ progression of OA?
  - Most speculate that this is not a regenerative solution
PRP Procedure
14 patients with OA
3 PRP injections at 4 week intervals of 6cc injection under ultrasound guidance

Results:
- No significant difference in ultrasound cartilage thickness at 6 months
- 8 of 13 improved based on VAS & KOOS, 3 of 13 no change,
- 2 of 13 worse

(Journal of Physical Medicine and Rehab 2009)
60 Patients with knee OA
30 patients with PRGF and 30 patients with hyaluronic acid injections
3 weekly injections

Results:
At 5 weeks (based on WOMAC scale)
- 33.3% success PRGF
- 10% Hyaluronic Acid

(Sports Medicine 2009)
Kon et al

- 114 patients with degenerative articular lesions of the knee
- 3 PRP injections 5cc q 21 days

Results:
- Significant improvement all clinical scores at 12 months with max improvement at 6 months

Knee Surgery Sports Traumatology 2010
Kon et al

- 120 patients with III-IV changes knee OA
- 60 patients PRP
- 60 patients hyaluronic acid injection

Results:
- At 12 months significant diff with PRP having better and longer efficacy compared to hyaluronic acid in decreased pain and increased function.
A Randomized Clinical Trial Evaluating Plasma Rich in Growth Factors (PRGF-Endoret) Versus Hyaluronic Acid in the Short-Term Treatment of Symptomatic Knee Osteoarthritis

Mikel Sánchez, Ph.D., Nicolás Fiz, Ph.D., Juan Azofra, Ph.D., Jaime Usabiaga, Ph.D., Enmanuel Aduriz Recalde, Ph.D., Antonio Garcia Gutierrez, Ph.D., Javier Albillos, Ph.D., Ramón Gárate, Ph.D., Jose Javier Aguirre, Sabino Padilla, Ph.D., Gorka Orive, Ph.D., and Eduardo Anitua, M.D., D.D.S., Ph.D.