Biopsies and Imaging at ACL Reconstruction

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Blunt impact often leaves a visible bone bruise. What about the overlying cartilage injury??
T1p Cartilage-Specific Imaging at 4.7T

trochlea sulcus

PG 18.7μg/μl PG 15.7μg/μl
Tissue Defined Zones
PG = 18.7μg/μl

- Superficial: < 0.01% Total PG
- Transitional: 25% Total PG 4.675 μg/μl
- Radial: 67% Total PG 12.5 μg/μl
- Deep: 7.9% Total PG 1.48 μg/μl

<table>
<thead>
<tr>
<th>T1rho Relaxation Times for Each Zone</th>
</tr>
</thead>
<tbody>
<tr>
<td>PG: 18.7 15.7 31.9 21.3 28.1 μg/μl</td>
</tr>
</tbody>
</table>

Mean T1rho Relaxation Times for Each Zone

<table>
<thead>
<tr>
<th>T1rho Relaxation Times Over All Specimens</th>
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</thead>
<tbody>
<tr>
<td>ROI</td>
</tr>
<tr>
<td>------</td>
</tr>
<tr>
<td>Superficial</td>
</tr>
<tr>
<td>Transitional</td>
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<tr>
<td>Radial</td>
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<tr>
<td>Deep</td>
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<tr>
<td>In Vitro Full-Thickness</td>
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<tr>
<td>In Vivo Full-Thickness</td>
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</tbody>
</table>

Normal Lateral Femoral Condyle Articular Cartilage
T1p Relaxation Time and PG

Sera Collected from 3 Patient Cohorts within 30 Days of Injury

1. Tibial plafond fractures (IAF) 20-75 years old
2. Normal – no joint injury 21-70 years old
3. Anterior cruciate ligament tears (ACL) 23-40 years old

<table>
<thead>
<tr>
<th>Marker</th>
<th>Units</th>
<th>IAF Mean</th>
<th>IAF Std.Dev</th>
<th>IAF n</th>
<th>Normal Mean</th>
<th>Normal Std.Dev</th>
<th>Normal n</th>
<th>ACL Mean</th>
<th>ACL Std.Dev</th>
<th>ACL n</th>
</tr>
</thead>
<tbody>
<tr>
<td>C2C</td>
<td>ng/mg</td>
<td>1795</td>
<td>900</td>
<td>18</td>
<td>0.072</td>
<td>0.031</td>
<td>10</td>
<td>705</td>
<td>99</td>
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<tr>
<td>BAP</td>
<td>µg/mg</td>
<td>0.065</td>
<td>0.034</td>
<td>19</td>
<td>12.4</td>
<td>4.4</td>
<td>28</td>
<td>0.095</td>
<td>0.021</td>
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<tr>
<td>Trap</td>
<td>mU/mg</td>
<td>17.0</td>
<td>6.9</td>
<td>18</td>
<td>4.7</td>
<td>2.2</td>
<td>27</td>
<td>28.4</td>
<td>8.5</td>
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</tr>
<tr>
<td>3B3</td>
<td>ng/mg</td>
<td>3.9</td>
<td>4.4</td>
<td>19</td>
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</tbody>
</table>
Serum Biomarkers

1. *Acute* indicators of bone and cartilage turnover in patients with:
   - **Intra-articular fractures**
     - matrix turnover from fracture healing
   - **Anterior cruciate ligament damage**
     - turnover from cartilage or ligament repair?

2. CS3B3 - an OA marker in these patients?

Collagen Degradation Marker: C2C

AKA “Col2- 3/4Clong”, C2C is a marker for collagen degradation by collagenases.
Bone Synthesis Marker: BAP

Bone alkaline phosphatase is an enzyme involved in matrix mineralization by osteoblasts (bone turnover).

Bone Degradation Marker: TRAP5b

Tartrate-resistant alkaline phosphatase is a bone mineral-degrading enzyme synthesized by osteoclasts (bone turnover).
The 3B3\(^{-}\) chondroitin sulfate epitope is made in immature & osteoarthritic cartilage, possibly synthesized by injury-responsive chondrogenic progenitor cells\(^1\).\(^2\)


The 3B3: GAG ratio was substantially higher in CPC cultures at 24 hours and 72 hours. The 3B3(-) epitope and total glycosaminoglycans (GAG) were measured in medium samples from primary cultures of chondrogenic progenitor cells (CPC), and medium from primary cultures of normal chondrocytes (NC). The columns are averages of two different experiments.
Conclusions at 2-4 weeks post injury

• T1p relaxation times were elevated from normal.
• Serum biomarkers TRAP and C2C were significantly higher in IAF patients, consistent with bone and cartilage turnover expected during fracture healing.
• CS3B3 was significantly higher in ACL patients, consistent with early osteoarthritic changes? Or activation of chondrogenic progenitor cells

OAI / OARSI OA Biomarkers Workshop 2012
Conclusions

- We propose that swelling and soft-tissue fluid infusion associated with the inflammatory response to injury were responsible for some difference in preoperative imaging markers of cartilage composition.
Bone sialoprotein is normally trapped within bone, but is released by osteoclast activity (bone degradation).

Conclusions at 2-4 weeks post injury

- T1ρ relaxation times were elevated from normal.
- There were significant differences in serum biomarkers in blunt trauma injured ACL and IAF patients.
  - TRAP and C2C were significantly higher in IAF patients, a finding consistent with bone and cartilage turnover expected during fracture healing.
  - CS3B3 was significantly higher in ACL patients, consistent with early osteoarthritic changes.
- The relationships of these imaging and biomarker data are inconclusive, but continued complementary sample collection and recommendations of this workshop should enhance that search.