The Role of Orthobiologics for Articular Cartilage Damage in the Knee

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WHAT THE HELL

YOU SAY?
Orthobiologics

- Bone Marrow Aspirate Concentrate
- Adipose-derived Mesenchymal Stem Cells
- Amniotic Fluid/membrane
- Platelet-Rich Plasma
- Umbilical Cord Tissue/Scaffold
- Micronized Allogeneic Cartilage
Chondral defect
Demonstration...
Bone Marrow Derived Stem Cells
Bone Marrow Derived Stem Cells

- Mesodermal Origin
- Precursors to Bone, Cartilage, fat, tendon, ligament
Mesenchymal Stem Cells

- Secrete Bioactive Molecules
  - Growth Factors
  - Cytokines
  - Chemokines
- In the Literature: Promising results with MSCs for Chondral defects and OA (AJSM, JBJS, Arthroscopy, etc)
Intra-articular Mesenchymal Stem Cell Therapy for the Human Joint

A Systematic Review

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Background: Stem cell therapy is emerging as a potential treatment of osteoarthritis (OA) and chondral defects. There is a great deal of heterogeneity in the literature. The indications for stem cell use, the ideal tissue source, and outcome measures for stem cell–based treatments have yet to be determined.

Purpose: To provide clinicians with a comprehensive overview of the entire body of the current human literature on safety and efficacy of intra-articular mesenchymal stem cell (MSC) therapy in all joints.

Methods: To provide a comprehensive overview of the current literature, all clinical studies investigating the safety and efficacy of intra-articular MSC therapy were included. PubMed, MEDLINE, and Cochrane Library databases were searched. All human clinical trials involving the use of MSCs for the treatment of OA and CD in all joints were identified. A total of 3867 publications were screened.

Results: Twenty-eight studies met the criteria to be included in this review. Fourteen studies treating osteoarthritis and treating focal chondral defects were included. MSCs originating from bone marrow (13), adipose tissue (12), synovial peripheral blood (2) were administered to 584 distinct individuals. MSCs were administered into the knee (523 knees), hip (5), and other joints (61). The mean follow-up time was 24.4 months after MSC therapy. All studies reported improvement in at least 1 clinical outcome measure, and no study reported major adverse events attributable to MSC therapy.

Discussion: The studies included in this review suggest that intra-articular MSC therapy is safe. While clinical and, in some cases, radiological improvements were reported for both OA and CD trials, the overall quality of the literature was poor, and a lack of reproducibility limits firm conclusions regarding the efficacy of these treatments.

Conclusion: This review provides strong evidence that autologous intra-articular MSC therapy is safe, with generally positive clinical outcomes.

Keywords: mesenchymal stem cell; MSC; intra-articular; stem cell; human

Adult cartilage is characterized by a limited intrinsic repair capacity after injury, owing to the sparse distribution of highly differentiated chondrocytes, the low supply of progenitor cells, and the lack of vascular supply. Osteoarthritis (OA) is a common disease affecting the joint cartilage and is characterized by progressive joint malalignment, pain, and disability. Osteoarthritis (OA) affects an estimated 20% of the world's population and is the most common joint disease in the United States.
19-y/o with medial chondromalacia s/p meniscectomy
Adipose-derived MSCs
## Adult Stem Sources

<table>
<thead>
<tr>
<th></th>
<th>Bone Marrow</th>
<th>Adipose Tissue</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Surgical Isolation Method</strong></td>
<td>Bone Marrow Aspiration</td>
<td>Lipoaspiration</td>
</tr>
<tr>
<td><strong>Method of Extraction</strong></td>
<td>Centrifugation</td>
<td>Centrifugation</td>
</tr>
<tr>
<td><strong>Enzymatic Digestion</strong></td>
<td>No</td>
<td>Depends</td>
</tr>
<tr>
<td><strong>Stem Cell Yield per Gram</strong></td>
<td>100 – 1,000 cells</td>
<td>~5,000 cells</td>
</tr>
<tr>
<td><strong>Abundance in Nucleated Cells</strong></td>
<td>0.001% - 0.01%</td>
<td>~2%</td>
</tr>
</tbody>
</table>

Bone Marrow Derived or Lipoaspirate Derived Adult Stem Cells are Mesenchymal Stem Cells (MSCs).
Amniotic Fluid/Membrane
Amniotic fluid cells produce high quantities of growth factor.

They have tremendous capacity for the production of growth and anti-inflammatory factors, and can differentiate into multiple tissues including bone, cartilage, and muscle among other cell types.
Growth factors found in Amniotic Fluid Concentrate

HGF: Hepatocyte Growth Factor → Myogenesis, Wound Healing, Organ Regeneration
EGF: Epidermal Growth Factor → Cell Growth, Proliferation, Differentiation
TNF-α: Tumor Necrosis Factor-Alpha → Apoptosis, Angiogenesis
GRO-α: Chemokine → Angiogenesis, Wound Healing
MCP-1: Monocyte Chemoattractant Protein-1 → Immune Modulation
TIMP (1,2,3,4): Tissue Inhibitor of Metalloproteinases (2,3,4) → Growth Promotion
IGF-1: Insulin-Like Growth Factor-1
IGF-2: Insulin-Like Growth Factor-2
IL1-RA: IL1-Receptor Antagonist → Anti-inflammatory
TGF-α: Transforming Growth Factor-Alpha
TGF-β1: Transforming Growth Factor-Beta 1
TGF-β2: Transforming Growth Factor-Beta 2
IL6: Interleukin 6 → Immune modulation
56-y/o with pain s/p PMM

- Prior surgery by outside surgeon (PMM)…continued pain
- No relief with PT/cortisone/meds
PRP (Platelet-Rich Plasma)

- When soft tissue is injured, platelet cells are delivered to the site of injury
- These platelets contain growth factors that help to heal the tissue
- PRP therapy amplifies this response
- The platelet-derived growth factors:
  - Increase collagen production
  - Stimulate blood flow
  - Enhance Stem cell proliferation
Platelet-Rich Plasma

- Growth factors (Alpha granules storage units)
  - Transforming Growth Beta Factor
  - Vascular Endothelial Growth Factor
  - Platelet Derived Growth Factor
  - Epithelial Growth Factor
  - Fibroblast Growth Factor

- Stimulate Cell Replication, angiogenesis, epithelialization, granulation tissue formation, extracellular matrix formation and regulation of bone cell metabolism
PRP Extraction
Umbilical Cord Tissue/Stem Cells

- Stem cells are found in the Wharton’s Jelly (smooth muscle cells surrounding the cord vessels)
- PolyCyte: growth factors, HA, cytokines
- CoreCyte: same + 1,000,000 MSC/cc
- Hypoimmunogenic, homologous, younger/higher proliferation

**Predictive Technologies**
MEMORANDUM

Subject: Memo on File
From: Doug Schmid, PhD
Date: 01/15/2017
Re: Ohio State University Viability Study

3 samples of varying concentration were sent on dry ice, overnight via FedEx, to Ohio State University for viability testing. Using a dye similar to DAPI, cellular counts and viability were obtained on a Countess II cell counter. The data are as follows:

A. **Sample 1**: (our count was 2 million cells/mL)
   - 1.82 million total cell count per mL (average of 2 counts), 78% viability

B. **Sample 2**: (our count was 5 million cells/mL)
   - 4 million total cell count per mL (average of 2 counts), 73% viability

C. **Sample 3**: (our count was 500,000 cells/mL)
   - 555,000 total cell count per mL (average of 2 counts), 64% viability

In storing cells, cellular concentration is a key component of viability—if the cell density is too low (less than 1 million cells per mL) or too high (more than 10 million cells per mL), cellular viability upon thaw will be affected. These numbers can vary by cell type; some cell types can survive the freeze/thaw cycle better than others.

From these viability measurements (and others made in our lab), we have determined that the optimal freezing density for the CoreCyte™ product is 2 million cells/mL. This concentration consistently yields approximately 80% viability and at least 1 million cells/mL in the freshly thawed product. These numbers were again confirmed in-house.
### ASSAY RESULTS

#### Fresh Sample ID

Excluding RBC contamination, cellularity consisted of 9.7x10^5 viable cells per mL. Viability based on exclusion of DAPI equals 67.6%. Cellular phenotype consisted of:

- CD90 = 59.2%
- CD105 = 55.4%
- CD73 = 86.7%
- CD45 = 3.19%

Total Viable Mesenchymal Stem Cell Count: 1,200,000 per mL. (This number is an approximation based on the use of bead markers. There is to be expected normal cell loss based on the steps to process the sample before Flow Cytometry testing. The 1,200,000 per mL number is the cell count that remains after testing protocol preparations.)

#### Frozen Sample ID

Excluding RBC contamination, cellularity consisted of 7.8x10^5 viable cells per mL. Viability based on exclusion of DAPI equals 43.1%. Cellular phenotype consisted of:

- CD90 = 28.0%
- CD105 = 35.1%
- CD73 = 84.4%
- CD45 = 4.06%

Total Viable Mesenchymal Stem Cell Count: 800,000 per mL. (This number is an approximation based on the use of bead markers. There is to be expected normal cell loss based on the steps to process the sample before Flow Cytometry testing. The 800,000 per mL number is the cell count that remains after testing protocol preparations.)

James Marvin
Director, Flow Cytometry SRL
Micronized Allogeneic Cartilage- BioCartilage
BioCartilage

- Allograft cartilage with extracellular matrix (Type 2 Collagen, proteoglycans, growth factors)
- Serves as a scaffold over the Microfractured defect
- More closely resembles native hyaline cartilage than microfx alone (AJSM, 2016)
BioCartilage

Treatment of Medial Femoral Condyle Cartilage Defect

Pre-Operative

2 Years Post-Op
BioCartilage™
Treatment of Lateral Femoral Condyle Cartilage Defect

Pre-Operative – Sag PD

1 Month Post-Op – Sag PD
BioCartilage™
Treatment of Lateral Femoral Condyle Cartilage Defect

Pre-Operative – Sag T2 FS

1 Month Post-Op – Sag T2 FS
15yo with large cyst of MFC. Autologous bone graft topped with BioCartilage + ACP. Non-compliant patient, patient became stiff. 3mo post-op, manipulation performed.
Thank-you