Propagation of Autologous Chondrocytes Using A Thermosensitive Polymer Gel Culture

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Management of Full Thickness Articular Cartilage Defects

- Drilling
- Microfracture
- Abrasion Arthroplasty for pluripotential stem cell stimulation
- Reattachment
- Allograft transplantation
- Mosaicplasty

Friedman et al. Clinical Orthopaedics. 1984
Minas et al. Instructional Course Lectures. 1999
High Failure Rates

• Fibrous Tissue at drilling and abrasion sites
• Failure in fixation / reattachment
• Immunogenicity
• Large cartilage defects at donor sites
Previous results have driven research towards autologous cartilage regeneration and transplantation.
Swedish Series of Autologous Chondrocyte Culture and Reimplantation

- 100 patients
- 2-10 year follow up
- Improvement in
  - 92% isolated femoral condyle defects
  - 74% femoral condyle & ACL Reconstruction
  - 84% OCD lesions
  - 69% patellar defects
  - 58% trochlea defects
  - 75% patients with multifocal defects

Peterson, AAOS 1998
Swedish series continued

- Histological Evaluation Via Biopsy
  - 25 biopsies (14 pts)
    - 13 Hyaline-like
    - 6 Combination hyaline/fibrous
    - 6 Fibrous
  - Strong correlation between hyaline-like cartilage and good clinical outcome
  - Strong correlation between fibrous tissue and poor outcome
Matrix or scaffold used to culture the cells may be very important in maintaining the original chondrocyte phenotype.
Thermosensitive Polymer

- Copolymer of poly(N-isopropylacrylamide) and acrylic acid (PNiPAAm-co-Aac)
- Lower Critical Solution Temperature (LCST) of 37 C
  - gels at 37 C or higher
  - liquid under 37 C
- Provides a Three Dimensional Matrix for Chondrocyte Culture
Hypothesis: Chondrocytes can

- Duplicate in the copolymer gel
- Regain their chondrocyte phenotype
- Be easily recovered from the gel by simply lowering the temperature below 37°C
Chondrocytes Harvested From Adult Rabbit Scapula

• Cultured in monolayer until confluent (approx. 2 wks)
• Cells seeded into copolymer gel and cultured for 3 wks
• Cells seeded into two control groups
  – monolayer gel
  – agarose gel
Monolayer Culture:
Fibroblast like
Agarose Culture: Good Phenotype but Poor Recovery from Media

Safranin O
Thermosensitive Polymer: Round Chondrocytes

H & E
Thermosensitive Polymer

Positive ALP
Second Study

- Harvested scapular chondrocytes from two NZW rabbits
- Cultured in monolayer until confluent
- Chondrocytes seeded into thermosensitive polymer and cultured for 3 wks
- Injection of polymer/chondrocyte scaffold into subcutaneous tissue on dorsum of original NZW rabbits
- After 4 wks, the rabbits were sacrificed and tissue examined
Gross Appearance:
Gross Appearance:

New Cartilage !?
Inflammatory Reactions?
Histology 10x

Safranin O & Hematoxylin
Histology 10x

Safranin O & Hematoxylin
Histology 10x

Safranin O & Hematoxylin
Histology 40x

Safranin O & Hematoxylin
Histology 40x

Safranin O & Hematoxylin
Histology 40x

Safranin O & Hematoxylin
Conclusion

- Current methods of autologous chondrocyte implantation (Carticel) show promising results
- High cost, technically difficult procedure
- Still has 8-42% failure rate depending on site of lesion
- Improved methods of autologous chondrocyte propagation are being investigated
Conclusion

• Autologous Chondrocytes can propagate in the Thermosensitive Polymer Culture in vitro
• Three Dimensional Matrix for culture may be superior to current monolayer culture techniques for maintenance of the original chondrocyte phenotype
• Initial in vivo studies yielded predominance of inflammatory reaction
• A 2nd generation thermosensitive polymer which is more biodegradable is being investigated currently
• More studies in vivo will need to follow
Thank you!