HYALURONIC ACID-BASED POLYMERS IN THE TREATMENT OF OSTEOCHONDRAL DEFECTS

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Introduction: Articular cartilage in adults has a limited ability for self repair. A number of methods have been devised to augment its natural healing response. These methods generally lead to filling of the defect with fibrous tissue or, in the best case, fibrocartilage, which may provide a temporary articulating surface, but lacks the mechanical characteristics of the articular cartilage and often fails with time. Transplantation of osteochondral grafts and periosteum also fails to produce a long lasting, satisfactory integration and regeneration. Recently, Tissue Engineering techniques have been used with natural and synthetic materials, alone or seeded with chondrocytes or marrow-derived mesenchymal progenitor cells. Some of these materials, alone or in combination with cells, allow the regeneration of hyaline cartilage[1], but this response is often accompanied by variable amounts of fibrocartilage. We have used two hyaluronic acid (HA)-based polymers to facilitate the natural healing response of osteochondral defects. These HA polymers mimic an embryonic-like milieu[2] in which, driven by local factors, endogenous mesenchymal progenitor cells from the marrow space can differentiate into the appropriate phenotypes to regenerate the injured tissues.

Methods: Following an IACUC-approved protocol, 39 four month-old rabbits received bilateral, 3 mm diameter x 3 mm deep osteochondral defects on the weight-bearing surface of the medial femoral condyle of the knee joint. Twenty-four defects were left untreated, 27 were treated with ACP™ sponge and 27 were treated with HYAFF®11 sponge. Both of these HA polymers were generously provided by Fidia Advanced Biopolymers srl (Abano Terme, Italy). Rabbits were sacrificed at 4 and 12 weeks after surgery. The condyles were fixed in formalin, decalcified, embedded in paraffin, cut and stained with Toluidine blue for histologic evaluation. All 12-week defects were scored with a modification of O'Driscoll's twenty-four point scale[3] described in Table I. Scores were compared with a Mann-Whitney rank sum test.

• Percentage hyaline articular cartilage	up to 8
• Surface regularity	up to 3
• Degenerative changes	up to 3
• Structural integrity	up to 2
• Thickness	up to 2
• Integration	up to 4
• Bone filling	up to 2
• Tidemark	up to 1
• Degenerative changes in adjacent cartilage	up to 3
• TOTAL	up to 29

Table I. Histologic scoring system used to grade the 12-week specimens

Results: The repair tissue of the untreated empty defects was similar at both time points in that most were filled with bony tissue up to or beyond the level of the tidemark, while the non-calcified top layer varied from undifferentiated fibrous to fibrocartilaginous tissue to tissue with more hyaline-like characteristics. No empty defect repaired completely with hyaline-like cartilage. Most empty defects presented varying amounts of the three tissue types with the 12-week specimens presenting a much thiner non-calcified top layer. Both ACP™ and HYAFF®11 sponges stayed in the defects, as revealed by the presence of sponge material in pilot 1-week sacrifices. Four weeks after surgery, the defects treated with ACP™ sponge exhibited a rapid endochondral bony fill to the level of the tidemark, except for the central area of the defect where some hypertrophic cartilage was observed. The top layer was composed of hyaline-like cartilage which showed impressive integration with the adjacent cartilage (Figue 1A). The 12-week specimens exhibited bone up to or beyond the tidemark level, and the non-calcified layer was mainly hyaline-like cartilage which was half as thick as the normal cartilage (Figure 1B).

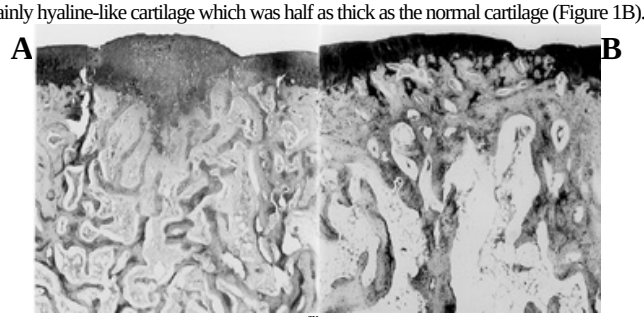

Figure 1. Histologic appearance of an ACP™ sponge-treated defect 4 (A) and 12 (B) weeks after surgery. Toluidine Blue staining.

Four weeks after surgery, the HYAFF®11 sponge-treated defects presented a rim of chondrogenic cells at the interface with the host tissue, while only empty sponge material was present in the center of the defect. The top layer was variable, with hyaline-like cartilage in some cases and fibrocartilage in others. Importantly, the defect tissue was always integrated with the adjacent cartilage (Figure 2A). At the 12-week time point, most of the defects exhibited bony fill with an area in the center which was either unfilled or filled by undifferentiated fibrous tissue. In some of the defects, a considerable amount of hypertrophic cartilage was still found in the defect. The non-mineralized surface layer was composed mainly of hyaline-like cartilage with, in some cases, cracks and fissures (Figure 2B).

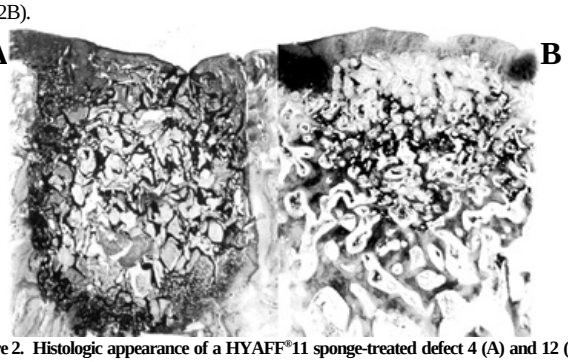

Figure 2. Histologic appearance of a HYAFF®11 sponge-treated defect 4 (A) and 12 (B) weeks after surgery. Toluidine Blue staining.

The histologic scores of the 12-week specimens revelaed differences between the groups (Table II). The ACP™ sponge-treated group presented higher scores than both the untreated ($p<0.002$) and HYAFF®11 sponge-treated ($p<0.05$) defects. The differences between the HYAFF®11 sponge-treated group and the empty (untreated) group were not significant ($p=0.29$). Power analysis suggests that a larger sample size is required to identify significant differences.

Group	Score	n
Empty	15.5 ± 3.47	10
ACP™	20.1 ± 3.00	15
HYAFF®11	17.3 ± 3.33	8

Table II. Histologic scores (mean ± std. dev.) of the 12-week specimens

Discussion: Although it is possible that different cell pools (bone marrow, synoviocytes, chondrocytes) contribute to the repair process of an osteochondral defect, the mesenchymal progenitor cells from the bone marrow are probably the main contributors to the natural repair process. Such natural reparative tissue in the untreated defects fails to withstand the function of normal articular cartilage and degenerates over time. The introduction of a biocompatible and biodegradable porous material into the defects with the appropriate chemical composition to allow differentiation of reparative cells must also provide the scaffolding for the reparative cells to regenerate and integrate. Hyaluronic acid-based polymers are excellent cell-delivery vehicles[4] and posses the unique biochemical composition to recreate an embryonic-like environment that is favorable for the regenerative process.

The materials studied in this series of experiments have different resorption characteristics that are important in the dynamics of the repair process. ACP™ sponge is resorbed quickly and only traces of the material can be found in some of the 4-week specimens; HYAFF®11 sponge is present in the defect for a longer period of time and was observed in all the 4-week specimens. The HYAFF®11 sponge was resorbed in most of the 12-week specimens. The presence of the HYAFF®11 sponge material over a longer period of time and the slow bony filling of the defect may be responsible for the lower scores relative to the ACP™ sponge-treated group where rapid resorption of the material results in subchondral bone formation by 4 weeks. This bony fill of the defect probably results in a more mechanically stable environment for the reparative process. Further work is required to fully assess the value of HYAFF®11 sponge and also the long-term outcome of osteochondral defects treated with these biopolymers.

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References:

[1] Wakitani, S *et al. J Bone Joint Surg*, 76: 579, 1984
[2] Toole, BP *J Internal Med*, 242: 35, 1977
[3] O'Driscoll, SW *et al. J Bone Joint Surg*, 68: 1017, 1986
[4] Solchaga, L *et al. J Orthop Res*, submitted, 1998

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