Effectiveness of a Biomaterial Adhesive in Integrating a Hydrogel with Surrounding Tissue in Rabbit Cartilage Defects

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Introduction

Hydrogels are a potential scaffold for cell-based cartilage repair [1]. One approach to hydrogel therapy is use of in situ photopolymerization of an injected non-viscous gel in order to permit near-perfect matching of the scaffold construct to the defect geometry. An important consideration in the development of protocols based on this is the stability of the hydrogel within the defect. Arthroscopy is limited by its invasive nature and by the fact that it provides surface information only. In contrast, MRI may permit noninvasive longitudinal evaluation of implant integrity, including visualization of the interface region between implant and subchondral bone. Accordingly, we investigated the ability of MRI to evaluate this interface, using T_2 mapping, in surgically created chondral defects in the rabbit femoropatellar groove after a 4-week residence period in the living rabbit. We further investigated chondroitin sulfate-methacrylate-aldehyde (CS-MA-ald) as a bonding material. Reaction occurs between the aldehyde groups of CS-MA-ald and the amino groups of bone and cartilage present in the defect walls, while the methacrylate groups in the adhesive and the hydrogel covalently crosslink to each other during the photogelation process.

Materials and Methods

Animal model: Defects were created in 4 skeletally mature New Zealand white rabbits (3-6 months). Two 4-mm full-thickness chondral defects, depth ca. 0.2mm, were created within the left and right femorapatellar grooves, with care taken not to damage underlying bone. After extraction of the joints and filling of the defects as described below, samples were maintained in protease inhibitor (PI) solution at 4°C.

Hydrogel: The hydrogel used to fill the defects consisted of 10% PEODA (Polyethylene glycol diacrylate) + 2.5 mg/ml hyaluronic acid (HA) + 0.05% of the photoinitiator Igracure D-2959 {2-hydroxy-1-[4-(hydroxyethoxy) phenyl]-2-methyl-1-propanone (Ciba-Geigy, Tarrytown, NJ).

Control group (PEODA-only): In control defects, the liquid state PEODA solution was poured into the cartilage defects and subjected to UV-irradiation (365nm; 8mW/cm^2) for 5 min to induce gelation.

Experimental group (PEODA-CS-MA-ald): In bonding experiments, the surfaces of the cartilage defect were exposed to 25% CS-MA-ald for four 4 minutes to permit bonding. After washing, the defect was filled with PEODA solution and subjected to UV-irradiation as above.

MRI protocol: All MRI experiments were performed on a Bruker DMX imaging spectrometer (Bruker Medizintechnik, Ettlingen, Germany) coupled to a vertical-bore magnet operating at 9.4 Tesla. 8 defects filled with PEODA only and 8 defects filled with PEODA-CS-MA-ald were imaged at 4°C using a spin-echo sequence with a minimum TE = 14 ms and TR = 2000 ms. Geometrical parameters were: slice thickness = 200 µm, FOV = 4 mm x 4 mm, and matrix size = 256 x 256. T_2 maps were constructed by a 3-parameter fit to 50 echoes using Bruker ParaVision software (v.2.1). T_2 was mapped as a function of position perpendicular to the surface of the implant.

Results and Discussion

Figs 1A and 1B show images from the first echo for the two groups. The hydrogel cannot be visualized in the ex-vivo rabbit defects after 4 weeks of implantation. However a thin layer of bright contrast was observed in the PEODA-CS-MA-ald (Fig. 1B) group. Normalized T_2 characteristics across the interface between cartilage and implant are shown in Figs. 2A and 2B. The average normalized T_2 value in this region for cartilage was 404 ± 124 ms. The average normalized T_2 for the PEODA-CS-MA-ald group was 474 ± 238 ms, while for the PEODA-only group it was 571 ± 112 ms, consistent with more mature cartilage as was also seen in histologic sections from these samples. This is also consistent with more effective integration in the PEODA-CS-MA-ald group but owing to scatter in the data this was found to be not significant (p-value =0.3).. In all cases, the slope of normalized T_2 with respect to distance (Fig 2B) showed 2 minima and 1 maximum in a position which corresponds to the transition between cartilage and gel. This indicates that MRI detects the presence of two interfaces in both the PEODA-only and PEODA-CS-MA-ald groups, consistent with the initiation of cartilage repair at the interfaces between hydrogel and cartilage and bone and cartilage during the 4-week period of implant residence within the living rabbit, like due to the provision of cells with chondrogenic potential from the blood and marrow [2]. In conclusion, our results indicate that use of CS-MA-ald as a bonding agent in hydrogel filling of defects permits improved integration with surrounding tissue and augmented early cartilage repair.

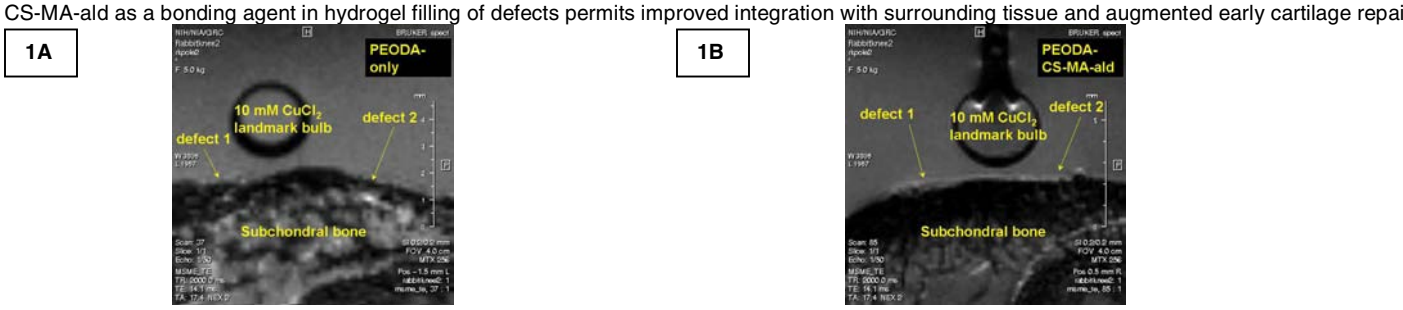

Figure 1: (A) MR Image of control group (without CS-MA-ald). (B) MR image of experimental group (with CS-MA-ald)

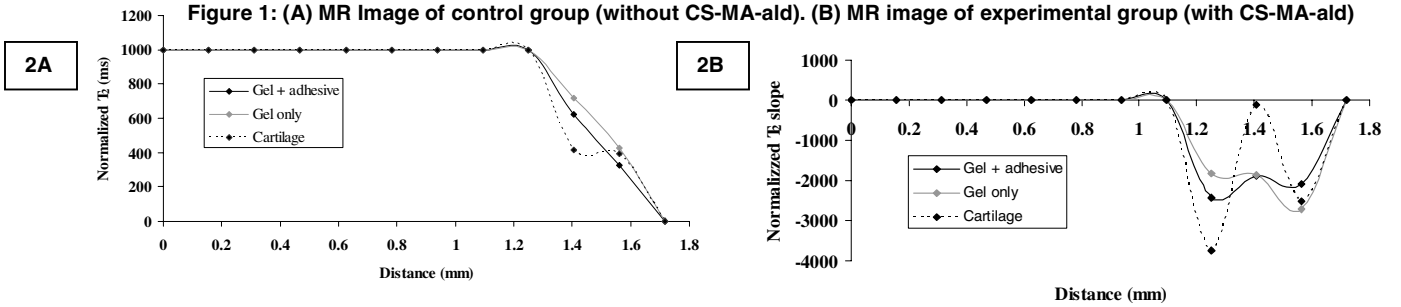

Figure 2: Normalized T_2 (Fig. 2A) and Normalized T_2 slope (Fig. 2B) distributions across the transition region in the 2 groups. (The transition region spans from a distance of 1.25 mm to 1.72 mm)

References

1] Wang, DA and Elisseeff, J.H, Encyclopedia of Biomaterials and Biomedical Engineering, New York; 2004. 1212-1225.

2] Breinan HA, Hsu HP and Spector M. Clin Orthop Relat Res. 2001. Oct;(391 Suppl):S219-30.