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Paper #21

# A Bioactive Scaffold Enhances Articular Cartilage Regeneration after Microfracture in a Rabbit Model

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**Summary**:
In a rabbit model of trochlear cartilage defects, treatment with a 3D-printed aggrecan-functionalized scaffold resulted in greater cartilage thickness, more chondrocytes, and enhanced glycosaminoglycan content compared to microfracture only.

**Abstract**:

## Introduction

The treatment of articular cartilage defects remains challenging due to the limited endogenous regeneration ability of cartilage and poor integration with implantations. Microfracture is a common treatment for full thickness cartilage lesions, but often leads to fibrous cartilage formation with decreased mechanical properties. We developed a 3D printed bioactive scaffold that can retain the released bone marrow cellular components after microfracture. This biological function was achieved by incorporating aggrecan, a major proteoglycan component present in native cartilage, to the scaffold. Our hypothesis was that the addition of aggrecan to a scaffold would improve the quality and thickness of the cartilage restoration after microfracture.

## Methods

14 New Zealand rabbits (female, 7-9 lbs) underwent a unilateral surgery to create a full thickness chondral defect in the central trochlea. The animals were then placed into four groups: 1) No treatment (n=3); 2) Microfracture only (n=3); 3) Microfracture with scaffold + fibrin glue (n=4); and 4) Microfracture with aggrecan functionalized scaffold + fibrin glue (n=4). All scaffolds were made of poly(L-lactide-co-e-caprolactone) and were 3D printed using an extrusion-based printer. Half of the scaffolds were surface functionalized with aggrecan. After surgery, the animals were observed for locomotion evaluation throughout the study using a modified Basso, Beattie and Bresnahan (BBB) scale. At 9 weeks postop, the animals were euthanized and the hind-limbs were harvested and analyzed with optical coherence tomography (OCT) and histology. Every limb underwent OCT scanning to determine the thickness of the regenerative trochlear cartilage. Histological analysis was then performed with both H&E and Alcain Blue staining. Groups were compared using the ANOVA statistical test.

## Results

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SECTION: All animals thrived postoperatively and there were no perioperative complications. Locomotion Evaluation: All experimental groups demonstrated a trend of increasing scores over time. There was no improvement in the defect control group. OCT Scanning: The OCT results revealed a more homogenous thickness distribution in the aggrecan-scaffold group compared to other experimental groups. The aggrecan-scaffold group had significantly greater average regenerated cartilage thickness (264 um) than the no treatment group (47 um), microfracture only group (139 um), and the non-aggrecan scaffold group (155 um) ($p<0.05$). Histologic Analysis: On H&E staining, there were ~3 times more chondrocytes in the regenerated layer present in the aggrecan scaffold group compared to the microfracture only group. The Alcian Blue staining further confirmed this therapeutic effect of the aggrecan-functionalized scaffold. The glycosaminoglycan content was greatly enhanced (440% increase) in the aggrecan scaffold group compared to microfracture only group.

## Discussion

In our rabbit model of trochlear cartilage defects, we have demonstrated improved cartilage regeneration when treated with a microfracture and a 3D printed scaffold. The use of an aggrecan-functionalized scaffold resulted in thicker and higher quality cartilage compared to use of a non-functionalized scaffold. The further development of this functionalized scaffold has great potential to improve the results of current cartilage restoration techniques.