THE SYNOVIUM HAS IMPORTANT EFFECTS TO REPAIR THE CARTILAGE DEFECTS

- The role of the synovium in cartilage defects using the synovium of GFP rats-

+ Hiroshima*University, Japan.
ayato-m@d6.dion.ne.jp

Introduction
It has been described in vitro that there are multipotential mesenchymal stem cells having chondrogenic potential in the synovium. Moreover, it was also observed that the transitional zone between the articular cartilage and the synovial membrane invaded the cartilage defects in an osteoarthritic knee or a rheumatic knee. However, the effects of the synovium on the cartilage defects and its derivation are still not clear. We examined the derivation of the tissue at the cartilage defects and the role of the synovium in cartilage repair using the synovium of Green Fluorescent Protein transgenic rats (GFP rats) and Sprague-Dawley (SD) rats.

Material and Method
Twenty-four male SD rats aged 8 weeks were used in study 1, and Twenty-four male SD rats aged 8 weeks and twelve male GFP rats aged 10 weeks were used in study 2. The protocol of this experiment was approved by the Ethical Committee of Hiroshima university.

Study 1: The knees of SD rats, 8 weeks of age, were used in this study. Intraperitoneal anesthesia was administered and the knee joint was opened using a paramedian approach. In experimental group 1, a 5 × 3 × 0.8 mm full-thickness cartilage defect in the transitional zone between the articular cartilage and the synovial membrane at the medial condyle of the femur was induced. In control group 1, in the contralateral knee, a full-thickness cartilage defect was induced in the same manner as in the experimental group 1, and a section of 5 × 5 mm was resected from the synovium of the transitional zone between the articular cartilage and the synovial membrane extending up to the cartilage defect. The rats were allowed to move freely in the cage after the surgery. They were sacrificed after 2, 4, 6, and 8 weeks, and the bilateral femoral condyles were harvested for macroscopic, histological, and immunohistochemical evaluation. Histological findings were performed by staining with hematoxylin and eosin as well as toluidine blue, safranin-O, and immunohistochemical staining for type II collagen. The histological evaluation using the scale of Pineda were performed, and the total mean scores for both groups were analyzed statistically.

Study 2: The knees of SD rats and GFP rats, 8 weeks of age, were used. After anesthesia, the knee was opened using a paramedian approach. In experimental group 2, a 5 × 3 × 0.8 mm full-thickness cartilage defect in the transitional zone between the articular cartilage and the synovial membrane at the medial condyle of the femur was induced. A section of 5 × 5 mm was resected from the synovium of the transitional zone extending up to the cartilage defect. The synovium of the GFP rat harvested from the same lesion as the medial condyle of femur was transplanted into the synovium defect of SD rat. In control group 2, a full-thickness cartilage defect was induced and the synovium of SD rats was resected; however, the synovium of GFP rats was not transplanted into the contralateral knee. The rats were sacrificed after 2, 4, 6, and 8 weeks, and were evaluated using the same manner as study 1. Furthermore, the expression of aggrecan and type II collagen in the cells located in the cartilage defect was assessed using reverse transcriptase-polymerase chain reactions (RT-PCR).

Results
1. Macroscopic and histological findings
Study 1: After 2 and 4 weeks, there was no macroscopic and histological difference in the degeneration of the cartilage between experimental group 1 and control group 1. After 6 and 8 weeks, the macroscopic degeneration of the cartilage was not progressive in experimental group 1 compared to that in control group 1. Histologically, the synovium-like tissue that invaded the cartilage defect was observed, and the cartilage-like tissue was found in almost specimens of the experimental group 1.

Study 2: After 2 and 4 weeks, there was no macroscopic and histological difference in the degeneration of the cartilage between experimental group 2 and control group 2, and GFP positive cells were observed, but histological difference in the defect was not obvious between two groups. After 6 and 8 weeks, GFP positive cells were observed in the invading tissue in experimental group 2; however, they were not observed in control group 2. The cartilage-like tissue was found in almost specimens of the experimental group 2 after 8 weeks.

2. Pineda’s histological grading score
Study 1: After 2 and 4 weeks, there was no significant difference between two groups. After 6 and 8 weeks, the score for experimental group 1 was significantly better than that for control group 1 (P<0.05).

Study 2: After 2, 4, and 6 weeks, there was no significant difference between two groups. After 8 weeks, the score for the experimental group 2 was significantly better than that for control group 2 (P<0.05).

3. Immunohistochemical evaluation
After 6 and 8 weeks, immunohistochemical staining for type II collagen was positive in specimens with a cartilage-like tissue in study 1 and study 2; however, it was not positive in control groups.

4. The expression of aggrecan and type II collagen using RT-PCR.
In experimental group 2, RT-PCR showed mRNA expression for aggrecan and type II collagen in the cells located in the cartilage defect, after 6 and 8 weeks no expression was showed in control group 2.

Discussion
Our results showed that the synovial cells invaded the cartilage defect. The synovial cells were derived from the transitional zone between the articular cartilage and the synovial membrane. From these results we could concluded that the synovial cells might have important effects to repair the cartilage defect.