# Platelet-Rich Plasma (PRP) on Co-culture of Chondrocytes and Osteoblasts and Its Positive Role in Gap Repair Between the Bone and Articular Cartilage

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**INTRODUCTION:** The healing of the gap between subchondral bone and articular cartilage is crucial in the treatment of osteonecrosis of the femoral head (ONFH) patients. This is because continuous weight-bearing force hinders regeneration of the gap and leads to degeneration of articular cartilage, which eventually requires arthroplasty to reduce hip pain. In this study, we aimed to investigate the influence of platelet-rich plasma (PRP) on the co-culture of chondrocytes and osteoblasts as well as its potential positive effects on gap repair using a co-cultured scaffold, expanding on the previous research.

**METHODS:** Allogenic leukocyte-depleted PRP was obtained from blood supernatants after centrifugation at 150G for 10 minutes. PRP was then incubated with the PLGA mesh. Human chondrocyte cells harvested from ONFH patients were co-cultured with commercially available hFOB1.19 osteoblast cell line on the PLGA mesh. The experimental group utilized the PRP-treated PLGA mesh, while the control group used only the PLGA mesh. For proliferation analysis and phenotype expressions, we counted cell numbers and employed RT-PCR with specific primers representing chondrocytes and osteoblasts. Subsequently, gap-mimic constructs, composed of devitalized human bone discs, the PLGA mesh, and cartilage, were implanted into subcutaneous pouches of nude mice (Figure 1). The gap-mimic constructs consisted of two groups: PRP-free scaffold (n=8) and PRP-treated scaffold (n=8). After 4 and 8 weeks, the constructs were harvested for gross and microscopic analysis. The attachment between bone and cartilage in the harvested constructs was assessed by categorizing the entire construct length into 0-25%, 25-50%, 50-75%, and 75-100%. The presence of cellular invasion at the interface was also examined.

**RESULTS:** In the PRP-free group, the number of chondrocytes and osteoblasts approximately increased 3.7-fold (7.5x10^4 ± 1.4x10^4 cells) seven days post-seeding, whereas, in the PRP-treated group, the number of cells increased an approximately 4.5-fold (9.1x10^4 ± 0.6x10^4 cells). FE-SEM findings also highlighted a more dense and expansive coverage of the mesh surface in the PRP-treated group (Figure 2). Furthermore, the phenotypes of both chondrocytes and osteoblasts in each group were well preserved, as evidenced by the results of RT-PCR and fluorescent microscopic analysis. At the 4-week assessment, both groups had similar attachment levels (PRP-free: 2 cases with 0-25%, 1 case with 50-75%, and 1 case with 75-100% attachment; PRP-treated: 2 cases with 0-25%, and 2 cases with 75-100% attachment). Cellular invasion was not observed in any of the four cases in the PRP-free group, while in the PRP-treated group, it was observed in one out of the four cases. At the 8-week assessment, both groups showed comparable attachment (1 case with 25-50%, and 3 cases with 75-100% attachment in each group). However, cellular invasion was observed in 2 out of 4 cases in the PRP-free group, while it was present in all 4 cases in the PRP-treated group (Figure 3).

**DISCUSSION:** In the co-culture process of chondrocytes and osteoblasts, the use of PRP resulted in a higher cell count compared to when PRP was not used, while still maintaining the phenotypes of each cell. Moreover, PRP is presumed to play a positive role in inducing the healing of the gap between subchondral bone and articular cartilage.

**SIGNIFICANCE/CLINICAL RELEVANCE:** Cell transfer technique with the use of PRP can play a positive role in repair the gap between subchondral bone and articular cartilage in ONFH patients.

**IMAGES AND TABLES:**

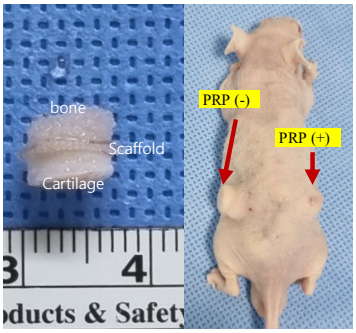


Fig 1. A gap-mimic construct (left), and mouse with implanted construct (right)

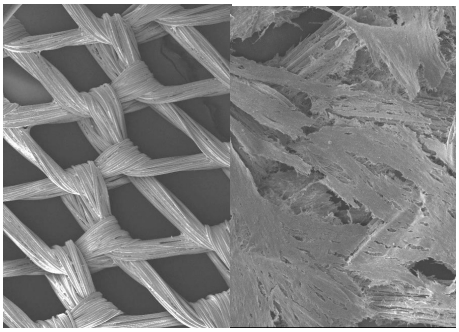

Fig 2. FE-SEM findings of PLGA mesh before (left) and 7 days after (right) the co-culture with PRP

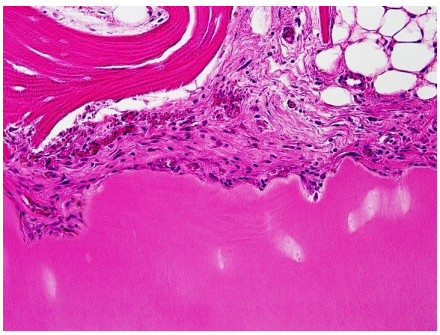

Fig 3. Interface with cellular invasion between bone and cartilage of the harvested construct