INTRODUCTION
Platelet-rich plasma is an autologous blood product that is being increasingly used to treat musculoskeletal conditions. Platelets have a rich store of factors and cytokines within their alpha granules and dense granules, which makes platelet-rich plasma an appealing therapeutic alternative. Some of the important factors found within the alpha granules of the platelet include platelet-derived growth factor (PDGF), transforming growth factor-beta (TGF-β), insulin-like growth factor-1 (IGF-1), vascular endothelial growth factor (VEGF), and epidermal growth factor (EGF), among others. We previously revealed that Synovium-derived mesenchymal stem cells (SDSCs) had a great proliferation potential and multi-lineage differentiation potential in vitro [1]. Furthermore, because SDSCs can be obtained from patients by minimally invasive techniques, they may serve as a source for cartilage regeneration. In an attempt to produce a high-quality cartilage tissue, this study introduced a PRP gel encapsulated with SDSCs and determined that this gel has the potential to regenerate and repair articular cartilage defects.

MATERIAL AND METHODS
Isolation of SDSCs and Tracking dye labeling: Synovium obtained from knee joint of New Zealand white rabbits was minced in PBS solution and digested in 0.02% collagenase overnight at 37°C. Cells were plated and maintained in DMEM containing 10% FBS. The cells of the second passages were harvested for seeding in the PRP gel.

Preparation of PRP: PRP was prepared using two centrifugation techniques according to a modified as previously reported [2]. Surgical procedure and PRP treatment: After general anesthesia, osteochondral defect was made in the trochlear groove (0: 4 mm, depth: 2.5 mm) of the knee joint of rabbit with a custom-made surgical tool. Rabbit knee joints were divided into three groups: (1) control group, (2) PRP group (150 μL/defect) and (3) PRP/SDSC group. The PRP and thrombin solution were injected using a dual syringe. Animals were sacrificed at 4, 12 and 24 weeks after injection.

Macroscopic examination: Macroscopic evaluation of cartilage repair was assessed using the International Cartilage Repair Society (ICRS) evaluation score [26]. Histology & IHC: The dissected samples were embedded in paraffin blocks, and subjected to H-E and safranin-O/Fast Green staining. IHC was accomplished using ABC methods. Primary antibodies were used mouse monoclonal anti-type I and II collagen antibody. Histomorphologic findings: Each slice was evaluated by three different authors and scored according to a modified version of the grading system developed by O'Driscoll et al. GAG and DNA amount assay: The synthesized GAG was determined by binding to DMB dye and total amount of GAG was normalized to the amount of DNA by indole assay. Statistical analysis: All experiments were performed in triplicate and the results were analyzed using paired t-test. P values less than 0.05 were considered significant.

RESULTS
Macroscopic findings. The representative gross appearance of the repair tissue at 4, 12 and 24 weeks after surgery is shown in Figure 1. At 4 weeks after the surgery, the regenerated areas of the control groups were red or brown. In the PRP and the PRP/SDSC groups, the surface of the repaired tissues was filled with smooth, white tissue which was recognized the margins of the intact cartilage. At 12 and 24 weeks after the surgery, the defect of the control groups was only partially covered with red or purple tissue which was not cartilage-like in appearances and had irregular surfaces. In addition, there was a depression in the center of the repair surface. In the PRP and the PRP/SDSCs groups, macroscopic appearance of the repaired tissue became opaque and completely integrated with the surrounding normal cartilage. It was difficult to discern the boundary between the host and the repaired tissue. In some cases of the PRP groups, the central area of the defects was slightly depressed. But, the defect of the PRP/SDSCs groups was filled with the repaired tissue with a hyaline cartilage-remodeling tissue.

Histological findings
At 4 weeks: The control groups were characterized by fibrous tissue and blod clot remaining from the perforated marrow vasculature. In the PRP and PRP/SDSCs groups, the defects were almost filled with fibrous reparative tissue. At 12 weeks: None of the control group healed completely with hyaline-like cartilage. In the PRP group, the surface of the regenerated tissue was irregular and varied from fibrous to fibrocartilagenous. In the PRP/SDSCs groups, the transplanted area was repaired by cartilage-like tissue, but some fissures were present at the border between regenerative tissue and adjacent host original cartilage. At 24 weeks: In the control groups, the repaired tissue was obviously thicker than the surrounding cartilage and had incomplete integration with surrounding cartilage. In the PRP group, the cartilage-like tissue developed in the defect area, but the junction between repaired tissue and subchondral bone was irregular. In the Gel/SDSCs groups, The regenerated area was obviously repaired by cartilage-like tissue with thickness equivalent to that of the surrounding original cartilage, and no fissures were present at the border between regenerative tissue and adjacent host original cartilage. The subchondral bone and tidemark were well remodeled (Fig 2). GAG synthesis from defect embedded in PRP/SDSCs group was shown to have increased in 24 weeks.

DISCUSSION
In this study, we demonstrated that the transplantation of PRP gel containing SDSCs led to the repair of an osteochondral defect in rabbits. Therefore, these results suggest this PRP gel encapsulated with SDSCs can be a good therapeutic candidate for the restoration of damaged or diseased articular cartilage. Additionally, this cell-PRP combination product has the potential to be delivered using an arthroscope, thereby conferring advantages for clinical application of damaged or diseased articular cartilage.

REFERENCES
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2) Ishida K et al: Tissue Eng., 2007

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