• PGE2 signal through EP2 agonist promotes regeneration of injured articular cartilage

INTRODUCTION:
Chondrocytes in articular cartilage are terminally differentiated cells with minimum proliferating potential and low metabolic activity. These cells are fully responsible for the production of cartilage matrix consisting of collagens and proteoglycans, hence considerable damage to articular cartilage is unrepairable, initiating a sequence of catabolic events leading to a pathological condition known as osteoarthritis (OA). Meanwhile Prostaglandin E2 (PGE2) exhibits pleiotropic effects in various types of tissue through four types of receptors, EP1-4. Among them, EP2 was identified as the major PGE2 receptor expressed in articular cartilage. An EP2 agonist increased intracellular cAMP in articular chondrocytes, stimulating DNA synthesis in both monolayer and 3D cultures. The effect of the PGE2 signal through prostaglandin E receptor 2 (EP2) receptors on the repair of injured articular cartilage was investigated in vivo using rabbit knee joints and a selective agonist for EP2 in this study.

METHODS: The Animal Research Committee of Kyoto University, approved this investigation. Japanese white rabbits were at least 5 months old and had a body weight of 3 kg. Two types of cartilage defect were made according to depth. A chondral and a osteochondral defects were prepared on the rabbit femoral concave in both knee joints, and gelatin containing polyactic-co-glycolic acid (PLGA) microspheres conjugated with or without the EP2 agonist was placed nearby. (Fig.1)

Animals were sacrificed at 4 or 12 weeks post-operation, and regenerated cartilage tissues and subchondral structure remodeling were evaluated by histological scoring. The quality of regenerated tissues was also evaluated by the immunohistochemical staining of EP2, type II collagen, and proliferating cell nuclear antigen (PCNA). As an evaluation of side effects, the inflammatory reaction of the synovial membrane was analyzed based on histology and the mRNA expression of matrix metalloproteinase3 (MMP3), and interleukin-1 beta (IL-1 beta). Also, the activity of MMP5 and the amount of tumor necrosis factor-alpha (TNF-alpha) and C-reactive protein (CRP) in joint fluid were measured. in both models. All statistical analyses were performed using Statcel software. The results are shown as the mean ± SD. The Mann-Whitney U test was used differences in the scores between two groups. A p value <0.05 was considered significant.

RESULTS: The EP2 agonist enhanced the regeneration of the type II collagen-positive tissues containing EP2- and PCNA-positive chondrocytes (Fig.2), and the histological scale of the regenerated tissue and subchondral bone was better than that of on the control side, particularly at 12 weeks post-operation. No inflammatory reaction in the synovial membrane was observed, and no induction of pro-inflammatory cytokines was found in joint fluid.

DISCUSSION:
The ideal regeneration-promoting therapeutics will be small molecules which can be produced in a large amount, promote the regeneration of articular cartilage with a physiological structure, and have no adverse effects in other tissues either locally or systemically. In this study, selective stimulation of the PGE2 signal through EP2 receptors by a specific agonist promoted regeneration of cartilage tissues with a physiological osteochondral boundary, suggesting the potential usefulness of this small molecule for the treatment of injured articular cartilages. Further confirmation of the effect of EP2 agonists in combination with a more effective drug delivery system and experimental OS models in larger animals may lead to a new way to treat OA.

SIGNIFICANCE:
This study relieved that the clinical possibility and availability of EP2 signal for injured cartilage repair in articular cartilage. The EP2 agonist may be a potent therapeutic agent for the cartilage damage.