DPP4 is a specific surface marker for senescent cells in osteoarthritis cartilage.

Sun Young Wang1, Ha Ru Yang2, Hee Jung Park2, You Jung Kim2, Ji Yoon Kim1, Gun Hee Cho1
Byung Sun Choi1,2, Hyun Cheol Bae2 and Hyuk-Soo Han1,2

1 Department of Orthopedic Surgery, Seoul National University, College of Medicine, Seoul, South Korea
2 Department of Orthopedic Surgery, Seoul National University Hospital, Seoul, South Korea
*wangsy@snu.ac.kr

INTRODUCTION: Senescent cells, state of cell proliferation arrest, accumulate in more vertebrate tissues as they age and are found in elderly diseases. Increase in senescent cell that occurs with aging appears to play a major role in driving life-limiting age-related diseases. These senescent cells are also a major factor in OA, senescent chondrocytes were found in cartilage tissue isolated from patients undergoing joint replacement surgery. It has been confirmed that the removal of senescent chondrocytes suppresses development of OA and promotes regeneration by creating an appropriate environment. To develop or screen effective senolytics for the selective elimination of senescent chondrocytes, it is necessary to understand senescent cell-specific signaling mechanisms by separating them. In this study, we aimed to identify a new senescence marker that enables cell separation in a living state through FACS analysis.

METHODS: Senescent chondrocyte markers were screened using flow cytometry and transcriptomic analysis using National Center for Biotechnology Information Gene Expression Omnibus. Senescence phenotypes, such as senescence-associated-galactosidase (SA-β-gal), proliferation, senescence gene expression, OA phenotypes, such as anabolic and catabolic gene expression, hypertrophic marker expression and histological analysis were evaluated in DPP-4+ chondrocytes.

RESULTS: 35 candidate genes for surface factors selectively expressed on OA chondrocytes are selected by cell surface marker screening by flow cytometry and transcriptomic analysis. Among the 35 genes, CD28 and DPP4 were significantly upregulated in OA patient. Through analysis of surface markers for non-OA, OA and oxidative stress-induced senescent chondrocytes DPP4 was selected as a final senescent chondrocyte marker (Fig1.). Immunostaining revealed expression of p16INK4a and DPP4 increases in correlation with severity of lesions found in human OA cartilage. FACS analysis showed that DPP4+ chondrocytes were increased in human OA cartilage (Fig2A-B). FACS-mediated sorting of DPP4+ chondrocytes show increased mRNA expression of p16 and p21 compared with DPP4- chondrocytes from OA cartilage and decreased proliferation rate (Fig2C-E). Chondrocytes expressing DPP4 exhibited decreased COL2A1 expression and increased hypertrophic marker MMP13 expression (Fig2F). And these results confirmed by decreased safranin-O staining of DPP4+ chondrocytes pellets. These data indicated that DPP4+ senescent chondrocytes may play a role in mediating OA development and progression. But the exact mechanisms by which DPP-4 expression leads to chondrocyte senescence or DPP4 upregulation and its downstream pathways are currently unknown, further study is required.

DISCUSSION: Our finding shows DPP4 is a potential cell surface marker of senescent chondrocytes in OA development and progression. But the exact mechanisms by which DPP-4 expression leads to chondrocyte senescence or DPP4 upregulation and its downstream pathways are currently unknown, further study is required.

SIGNIFICANCE/CLINICAL RELEVANCE: We suggest DPP4 as senescent chondrocyte markers of OA chondrocytes. And we also propose the therapeutic strategy for OA treatment by selectively removing DPP4+ chondrocytes.