SENESCENT SELLS EMERGE IN ARTICULAR CARTILAGE, SUBCHONDRAL BONE, SYNOVIUM, AND THE GREATER TUBEROSITY IN A RAT MODIFIED CUFF TEAR ARTHROPATHY MODEL

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INTRODUCTION: Osteoarthritis (OA) is a degenerative disorder involving not only the articular cartilage and subchondral bone but also joint capsules, ligaments, and tendons. Senescent cells contribute to the progression of OA through the secretion of senescence-associated secretory phenotype (SASP) factors. In knee OA animal models, senescence has been reported in chondrocytes and synovial fibroblasts[1)], and there are also reports of senescence in osteocytes, osteoblasts, and osteoclasts that may be associated with OA development[2)]. These findings suggest that cellular senescence is a common pathological mechanism in OA pathogenesis. Cuff tear arthropathy (CTA) is a shoulder secondary OA characterized by humeral head femoralization, a distinct morphological change not observed in other joint OA. Emerging evidence indicates that unloading induces the accumulation of senescent cells in cartilage, bone, ligaments, and tendons. Given that the shoulder is a predominantly non–weight-bearing joint, the pattern of senescent cell emergence in shoulder OA may differ from that observed in the knee. The aim of this study was to investigate the presence and distribution of senescent cells in the rat modified CTA (mCTA) model previously established by our group[3)].

METHODS: The animal experiment was approved by the Animal Experiment Ethics Committee of Kagoshima University (reference number: MD24036). Twelve 12-week-old male Sprague–Dawley rats were used. mCTA surgery was performed on the right shoulder involving transection of the supraspinatus (SSP) and infraspinatus tendons, the long head of the biceps tendon, and the superior half of the joint capsule, whereas sham surgery (deltoid split) was performed on the contralateral side. Euthanasia was performed at 4 weeks postoperatively, and glenohumeral joints were harvested. Immunohistochemistry (IHC) was performed to evaluate expression of $p16^{INK4a}$, a senescence marker, and the SASP-related cytokines TNF-α and IL-1β, in chondrocytes, osteocytes, synovium, and the SSP enthesis bone (greater tuberosity). Statistical analysis was performed using paired t-tests, with statistical significance defined as $P<0.05$ ($n=6$).

RESULTS SECTION: Similar to previously reported knee OA models, the humerus in the mCTA model also exhibited a marked increase in $p16^{INK4a}$-positive cells within the articular cartilage in the deep layer, synovial cells, and osteocytes of the subchondral bone (Fig. 1). In these regions, TNF-α and IL-1β were markedly elevated in the mCTA model, except for IL-1β in the subchondral bone (Fig. 2). As a finding specific to the mCTA model, $p16^{INK4a}$ expression at the SSP enthesis was markedly increased (Fig. 1), accompanied by a concomitant elevation of SASP factors (TNF-α and IL-1β) (Fig. 2).

DISCUSSION: Similar to knee OA models, the mCTA model exhibited an increase in senescent cells in the deep layers of articular cartilage, in osteocytes of the subchondral bone, and in synovial cells. These results suggest that joint loading patterns may not be strongly associated with the emergence of cellular senescence in OA. Alternatively, because rats are quadrupeds and the forelimbs bear some load, albeit less than the hindlimbs, it is possible that the mCTA model produced results similar to those observed in knee OA models. Conversely, a finding specific to the mCTA model was a marked increase in cellular senescence at the SSP enthesis. Notably, there is no evidence in ACL-transected knee OA models that senescent cells accumulate at the ACL footprint, suggesting that this phenomenon reflects changes in mechanical stress unique to the shoulder joint. Several reports have documented degenerative bone loss at the greater tuberosity following rotator cuff tears[4),5)], which may contribute to femoralization; the observation of senescence in SSP enthesis cells raises the possibility that such senescence contributes to this bone loss. Furthermore, we have previously reported decreased bone volume in the humeral head of CTA patients[6)]. If subchondral bone cellular senescence is a contributing factor, senolytic therapies could potentially serve as a novel treatment option, not only preventing OA progression in CTA but also mitigating bone loss–related humeral head collapse and femoralization.

SIGNIFICANCE/CLINICAL RELEVANCE: In this study, we demonstrated that senescent cells were markedly increased in the articular cartilage, subchondral bone, synovium, and the SSP enthesis in the mCTA model. Since these tissues are all responsible for CTA-related changes, if the accumulation of senescent cells represents one of the pathogenic mechanisms, senolytics may serve as a potential therapeutic agent for preventing CTA changes following rotator cuff tears or for treating established CTA.

REFERENCES:
1) Jeon OH, et al. *Nat. Med.,* 2017;23:775–781.
2) Li Y, et al. *Bone Res.*, 2025;13:18.
3) Ijuin T, et al. *Osteoarthr. Cartil. Open*, 2023;5:100389.
4) Jiang Y, et al., *Skeletal Radiol.*, 2002;31:522–528.
5) Waldorff EI, et al., *J. Shoulder Elbow Surg.*, 2011;20:904–908.
6) Tawaratsumida H., et al., *Osteoarthritis Cartilage*, 2025;33:101–115.

IMAGES AND TABLES:

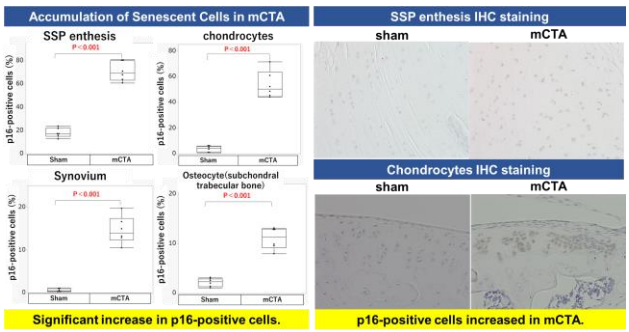

Fig.1

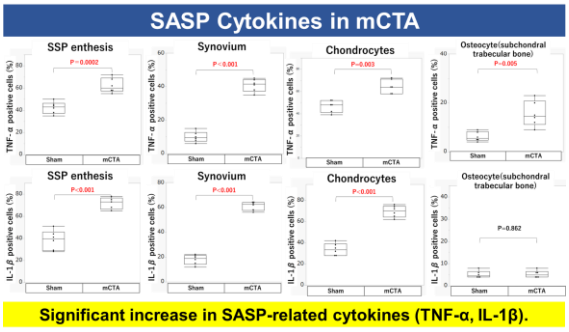

Fig.2