ing insulin sensitivity. Recently, adiponectin has also been found to regulate immune responses and inflammation. Adiponectin is found in OA joints but its role in the pathogenesis of OA and in cartilage metabolism is not clear. In the present study, we investigated the relation of circulating adiponectin and biomarkers of cartilage degradation (COMP and MMP-3) in patients with OA, and the effects of adiponectin on human OA cartilage.

**Methods:** Blood samples were collected from 38 male OA patients (BMI 29.5±0.8 kg/m²) undergoing knee replacement surgery because of severe OA, and adiponectin, COMP and MMP-3 concentrations were measured by immunoassay. Cartilage samples collected from OA patients under total knee arthroplasty were placed in tissue culture and exposed to adiponectin.

**Results:** Plasma adiponectin (2.5±0.2 μg/ml) correlated positively with serum COMP (r=0.55, p=0.001) and plasma MMP-3 (r=0.34, p=0.046). In tissue culture experiments, adiponectin increased the expression of iNOS, and production of nitric oxide, interleukin-6 (IL-6) and MMP-3 in human OA cartilage. The effects of adiponectin of NO and IL-6 production were mediated through MAP kinases Erk1/2, p38 and JNK, and p38 pathway was involved in the adiponectin-induced MMP-3 production.

**Conclusions:** Circulating adiponectin concentrations correlated positively with the measured biomarkers of cartilage degradation, i.e. COMP and MMP-3 in male OA patients; and adiponectin was found to increase the production of catabolic/proinflammatory mediators MMP-3, nitric oxide and IL-6 in human OA cartilage. The findings introduce adiponectin as a catabolic factor in OA.

186 TOLL-LIKE RECEPTORS ACTIVATE THE ESE-1 PROMOTER IN CHONDROCYTES BY A MECHANISM PARTIALLY DEPENDENT UPON NF-κB

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**Purpose:** Toll-like receptor (TLR)2 is increased in OA cartilage and TLR2 ligands trigger catabolic responses in chondrocytes, including MMP release. We previously showed that Epithelial Specific ETS (ESE)-1 expression is induced by inflammatory factors, including LPS, in non-epithelial tissues and that it acts as a primary mediator of inflammatory responses. In addition, ESE-1 levels are higher in OA chondrocytes, where it mediates IL-1-induced COL2A1 promoter repression, suggesting its involvement in cartilage catabolism. In this study, we aimed to investigate the role of TLR2 in regulating ESE-1 gene expression in chondrocytes.

**Methods:** Immortalized human C28/12 chondrocytes were transfected with TLR2, TLR-4 and MyD88 expression vectors, and ESE-1 messenger RNA (mRNA) was analyzed by real time RT-PCR. Transactivation by TLR2 and TLR4 was analyzed using ESE-1 messenger RNA (mRNA) was analyzed by real time RT-PCR. Transactivation by TLR2 and TLR4 was analyzed using reporter assays. TLR2 and TLR4 luciferase assays were performed in C28/I2 cells. In luciferase reporter assays, TLR2 overexpression, alone or together with MyD88, activated the pXP2/ESE1wt promoter, whereas TLR4 required MyD88 for the activation and the pXP2/ESE1mut was less responsive to TLR2 without or with the MyD88 adaptor. TLR2 alone gave a modest increase in ESE-1 promoter activity, while TLR2 cooperated with NF-κB p65/p50 in transactivating the ESE-1 promoter activity. However, TLR4 required its adaptor MyD88 to cooperate with p65 and p50 in transactivating the ESE-1 promoter activity. The addition of an NF-κB inhibitors, SN50 or CAPE, partially blocked the TLR2-induced ESE1 promoter activity alone or together with MyD88.

**Conclusions:** Our previous findings that ESE-1 is induced by inflammatory factors, including LPS, a TLR4 ligand, prompted us to ask whether ESE-1 participates in cartilage catabolic processes induced by TLRs. These receptors have been detected in OA cartilage, where they are proposed to augment joint destruction. Indeed, TLR2 levels are higher in OA cartilage and its ligands induce chondrocyte catabolic responses, including MMP release and type II collagen degradation. Here, we show that the ESE1 promoter activity and endogenous ESE1 mRNA were up-regulated by TLR2 and that this activation partially relies on the translocation of the NF-κB family member p50 and p65 to the nucleus. Given the role of ESE-1 as a key regulator of responses to inflammatory mediators, our results suggest that ESE-1 may participate in the catabolic responses triggered by TLR2 or TLR4 in OA cartilage.