

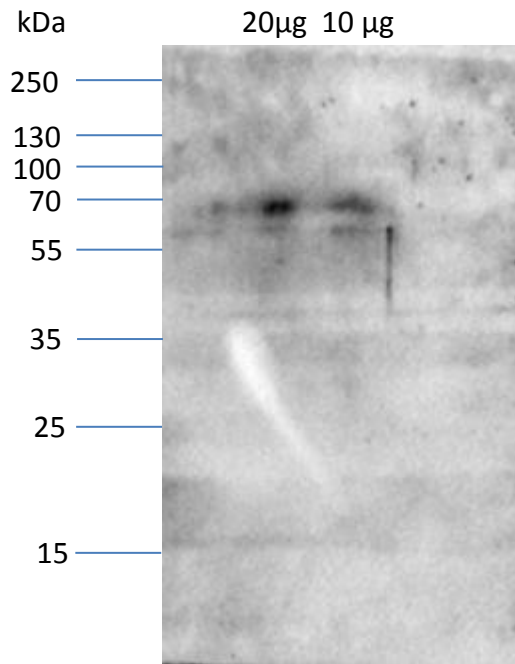
IL-6 secretion in osteoarthritis patients is mediated by chondrocyte-synovial fibroblast cross-talk and is enhanced by obesity

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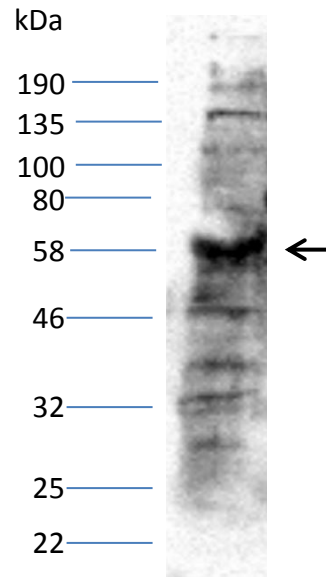
Supplementary Table S1: Cytokine expression in OA synovial fluid

Cytokine	Abundance (pg/ml)		p value
	<i>-leptin</i>	<i>+leptin (500ng/ml)</i>	
IL4	0.41 ± 0.19	0.5 ± 0.25	0.7860
IL6	339.31 ± 6.18	691.19 ± 36.66	0.0007
IL8	566.03 ± 487.67	876.9 ± 755.5	0.7470
IL10	3.16 ± 0.49	3.08 ± 0.88	0.9407
IL17	21.01 ± 7.52	27.75 ± 13.53	0.6856
GM-CSF	55.53 ± 4.07	59.83 ± 3.68	0.4769
TNF α	15.26 ± 3.76	18.37 ± 5.69	0.6727

Statistical significance was determined by unpaired T test.

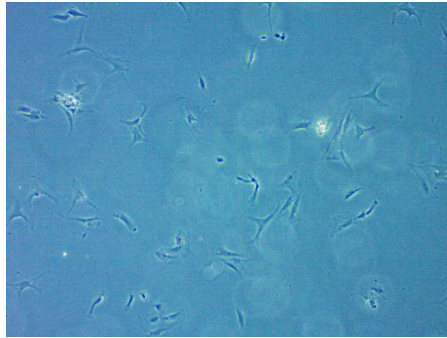


Supplementary Figure 1. Western blot demonstrating positive CD55 expression in primary OA synovial fibroblasts. Total protein RIPA lysates (20 and 10 μ g) were subjected to 10% SDS-PAGE, and immunoprobed with a CD55 antibody (Abcam, ab133684). Blots were developed using ECL. Molecular weight marker used was PageRuler Plus (ThermoFisher).

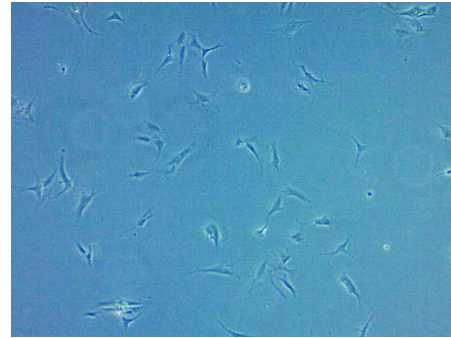


Supplementary Figure 2. Western blot demonstrating positive expression of Type II collagen in primary human OA chondrocytes isolated by collagenase digestion of articular cartilage. Total protein RIPA lysates (10 μ g) were subjected to 10% SDS-PAGE, and immunoprobed with a Col2 antibody (Abcam, ab34712). Blots were developed using ECL. Molecular weight marker used was Colour Prestained Protein Standard, Broad-range (NEB).

Chondrocytes

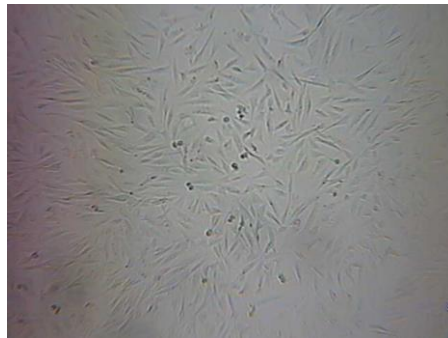


Control

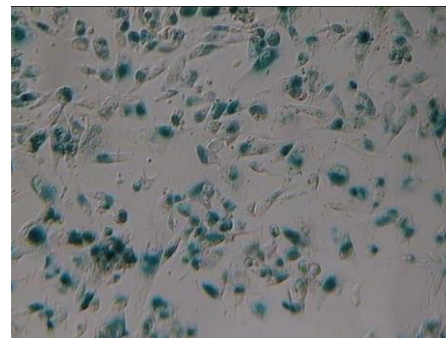


Leptin (500 ng/ml)

HRas
Fibroblasts



Control



4-OHT (333 nM)

Supplementary Figure 3. *In situ* staining for Beta-gal activity as a marker of cellular senescence. Primary human OA chondrocytes cultured in T25 culture flasks were stimulated for 24h with leptin (500 ng/ml), or left unstimulated. Beta-gal staining was performed using a histochemical staining kit (Sigma Aldrich, UK). A HRas fibroblast cell line was used as a positive control, whereby senescence was induced by addition of 4-OHT. Images are at 4x magnification