

Supplemental Information

The Synovium Attenuates Cartilage Degeneration in KOA through Activation of the Smad2/3-Runx1 Cascade and Chondrogenesis-related miRNAs

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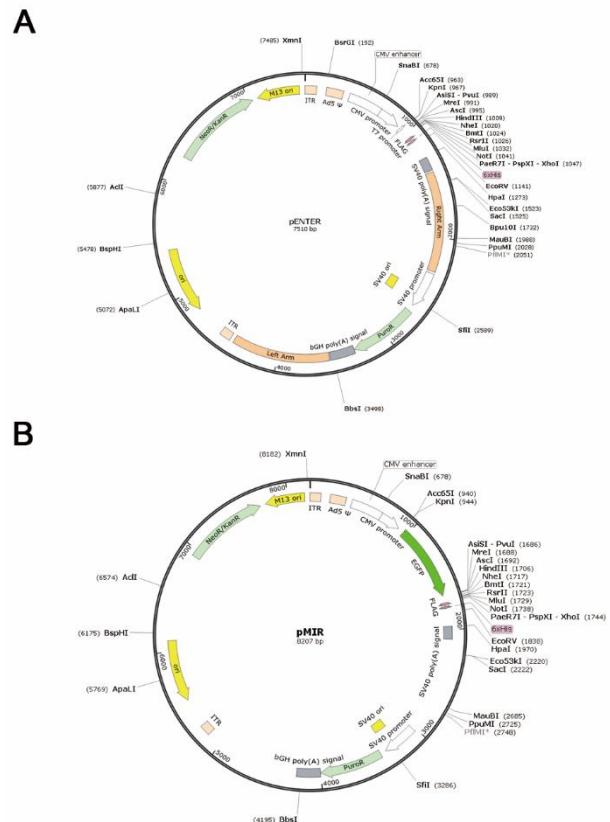


Figure S1. Illustration of the plasmid vectors. Properties of the hsa-RUNX1 overexpression plasmid (A). Properties of the hsa-miR-455 and hsa-miR-210 overexpression plasmid (B)

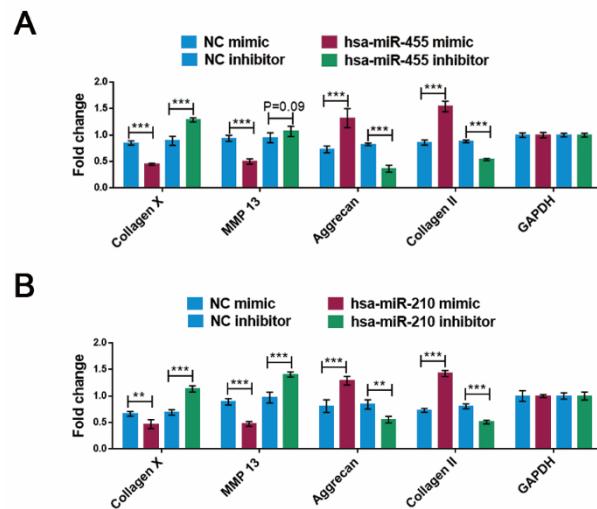


Figure S2. The grayscale values of the blots were quantitated and statistically analyzed. The expression patterns of chondrogenic genes and hypertrophic genes in chondrocytes after transfection with the mimic NC, miRNA mimic, inhibitor NC or miRNA inhibitor for miR-455 (A) and miR-210 (B).

Table S1. The specific siRNA sequences used in the present study.

Gene name	siRNA sequence 5'-3'	
hsa-RUNX1	Sense	CCTCGAAGACATCGGCAGAAA
	Anti-sense	TTTCTGCCGATGTCTCGAGG
hsa-SMAD2	Sense	CGATTAGATGAGCTTGAGAAA
	Anti-sense	TTTCTCAAGCTCATCTAATCG
hsa-SMAD3	Sense	GAGCCTGGTCAAGAAACTCAA
	Anti-sense	TTGAGTTCTTGACCAGGCTC

Table S2. The primer sequences of putative Runx1 binding sites in the promoter regions of miR-455 and miR-210.

Gene name	primer sequence 5'-3'	
Promoter region of hsa-miR-210	Sense	CCCCATGGAGGCTCAAGTTCA
	Anti-sense	CATGTGTGCCCCACTTCCCTTA
Promoter region of hsa-miR-455-01	Sense	TGCAAGCTCCAGGTGGAAAAAT
	Anti-sense	GGAAGGCCTGTATGACTGGA
Promoter region of hsa-miR-455-02	Sense	CTCCCCAATGCCGTGTTCT
	Anti-sense	TGTGAATGTACCTAACGCTATCGAA
Promoter region of hsa-miR-455-03	Sense	AGTGTGGGTGGTGACAATCTG
	Anti-sense	CATTAACTAACAGCGGCTGCG
Promoter region of hsa-miR-455-04	Sense	TCCAGAGGAAAGCTTGGTCC
	Anti-sense	TAGCCTGCTCGTAGAGACCC
Promoter region of hsa-miR-455-05	Sense	ACAGAGATCCCCTCAGCCAT
	Anti-sense	GAGGTAAGCGTGGAAAGCTGT
Promoter region of hsa-miR-455-06	Sense	CAGCCCCCTGTAGAGGTAA
	Anti-sense	TGCTTGTGAGGAGAGGCAG

Table S3. The specific primer sequences of mRNA used in the present study.

Gene name	mRNA primer sequence 5'-3'	
hsa-ADAMTS4	Sense	GGTCAAGGTCCCATGTGCAAC
	Anti-sense	GAATGCGGCCATCTTGTAC
hsa-ANCA	Sense	GTGCCTATCAGGACAAGGTCT
	Anti-sense	GATGCCTTTCACCACGACTTC
hsa-AR	Sense	ATGGTGAGCAGAGTGCCCTATC
	Anti-sense	ATGGTCCCTGGCAGTCTCCAAA
hsa-COL1A1	Sense	GTGGAAACCCGAGCCCTGCC
	Anti-sense	TCCCTTGGGTCCCTCGACGC
hsa-COL2A1	Sense	GCACCTGCAGAGACCTGAAAC
	Anti-sense	GCAAGTCTGCCAGTCTCCA
hsa-COL5A1	Sense	GCCCGGATGTCGCTTACAG
	Anti-sense	AAATGCAGACGCAGGGTACAG
hsa-COL10A1	Sense	CATAAAAGGCCCACTACCCAAC
	Anti-sense	ACCTTGCTCTCCTTACTGC

hsa-IRF7	Sense	CCACGCTATACCATCTACCTGG
	Anti-sense	GCTGCTATCCAGGGAAGACACA
hsa-JUN	Sense	CCTTGAAAGCTCAGAACTCGGAG
	Anti-sense	TGCTGCCTTAGCATGAGTTGGC
hsa-LOX	Sense	GTGGCCGACCCCTACTACATCC
	Anti-sense	AGCAGCACCTGTGATCATAATCTC
hsa-MEF2C	Sense	TCCACCAGGCAGCAAGAATACG
	Anti-sense	GGAGTTGCTACGGAAACCACTG
hsa-MMP13	Sense	TCCTGATGTGGGTGAATACAATG
	Anti-sense	GCCATCGTGAAGTCTGGTAAAAT
hsa-MYC	Sense	CCTGGTGCTCCATGAGGAGAC
	Anti-sense	CAGACTCTGACCTTTGCCAGG
hsa-PLOD2b	Sense	TTAAAGGAAAGACACTCCGATCAGAGATGA
	Anti-sense	AATGTTCCGGAGTAGGGAGTCTTTT
hsa-RUNX1	Sense	CCACCTACCACAGAGCCATCAA
	Anti-sense	TTCACTGAGCCGCTCGGAAAAG
hsa- RUNX2	Sense	CCCAGTATGAGAGTAGGTGTCC
	Anti-sense	GGGTAAGACTGGTCATAGGACC
hsa- RUNX3	Sense	GGCAATGACGAGAACTACTCCG
	Anti-sense	GATGGTCAGGGTCAAACCTTCC
hsa-SMAD2	Sense	GGGTTTGAGCCGTCTATCAGC
	Anti-sense	CCAACCACGTAGAGGTCCATT
hsa-SMAD3	Sense	TGAGGCTGTCTACCAGTTGACC
	Anti-sense	GTGAGGACCTTGTCAAGCCACT
hsa-SOX9	Sense	AGCGAACGCACATCAAGAC
	Anti-sense	CTGTAGGCGATCTGTTGGGG
hsa-VDR	Sense	CGCATCATTGCCATACTGCTGG
	Anti-sense	CCACCATCATTCACACGAAC
hsa-GAPDH	Sense	GCACCGTCAAGGCTGAGAAC
	Anti-sense	ATGGTGGTGAAGACGCCAGT

Table S4. The specific primer sequences of miRNA used in the present study.

Gene name	miRNA primer sequence 5'-3'	
hsa-miR-193b	Sense	AACUGGCCCUAAAGUCCCGCU
hsa-miR-199a	Sense	ACAGUAGUCUGCACAUUGGUUA
hsa-miR-455	Sense	GCAGUCCAUGGGCAUAUACAC
hsa-miR-210	Sense	AGCCCCUGCCCACCGCACACUG
hsa-miR-381	Sense	AGCGAGGUUGCCUUUGUAAU
hsa-miR-92a	Sense	UAUUGCACUUGUCCGGCCUGU
hsa-miR-320c	Sense	AAAAGCUGGGUUGAGAGGGU
hsa-miR-136	Sense	ACUCCAUUUGUUUUGAUGAUGGA
cel-miR-39	Sense	UCACCGGGUGUAAAUCAGCUUG
hsa-U6	Sense	CTCGCTTCGGCAGCACA
	Anti-sense	AACGCTTCACGAATTGCGT

Table S5. miRNA-specific probes for in situ hybridization analysis

Gene name		miRNA probe sequence 5'-3'
hsa/mmu-miR-455	Probe	DIG-GTGTATATGCCCATGGACTGC-DIG
hsa/mmu-miR-210	Probe	DIG-TCAGCCGCTGTCACACGCACA-DIG
hsa-miR-320c	Probe	DIG-ACCCTCTCAACCCAGCTTT-DIG