

## **SUPPLEMENTARY MATERIAL**

### **RUNX2 stabilization by long non-coding RNAs contribute to hypertrophic changes in human chondrocytes**

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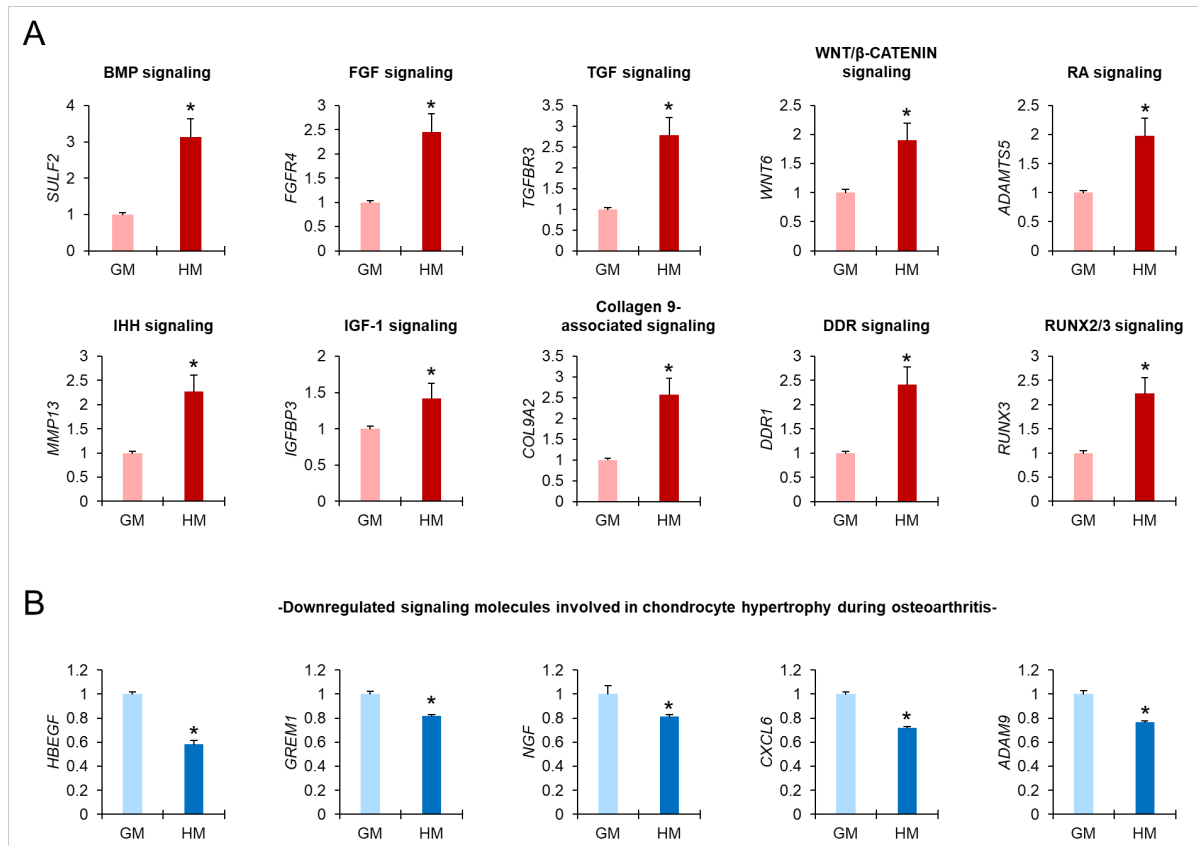
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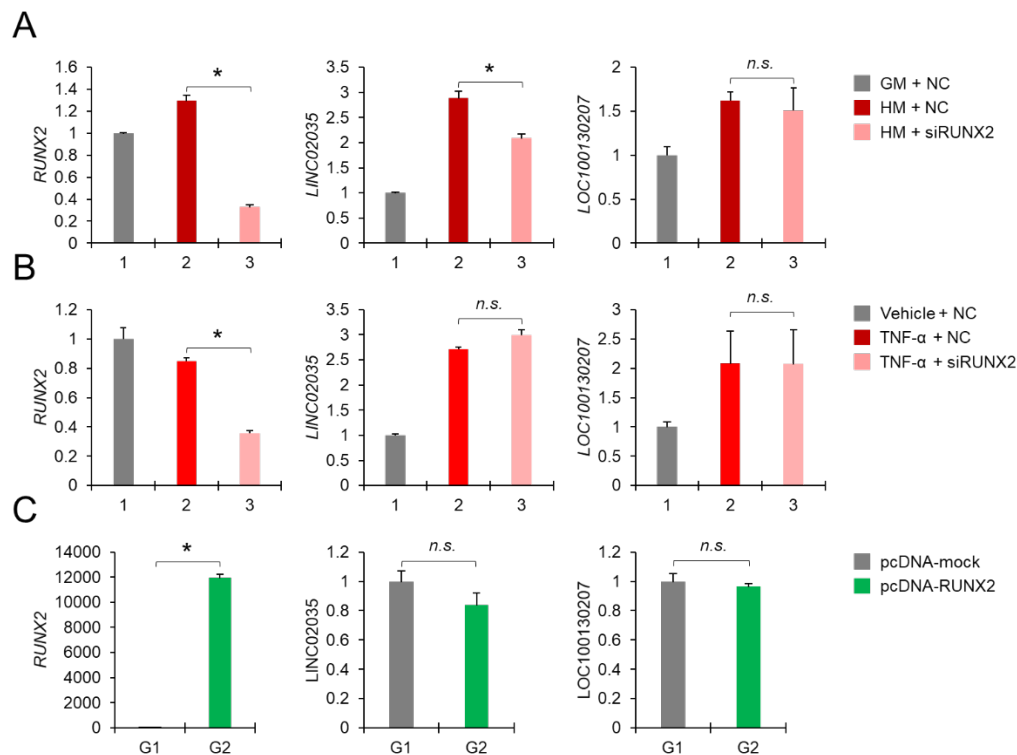
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**Running title:** RUNX2-interacting lncRNAs are involved in chondrocyte hypertrophy

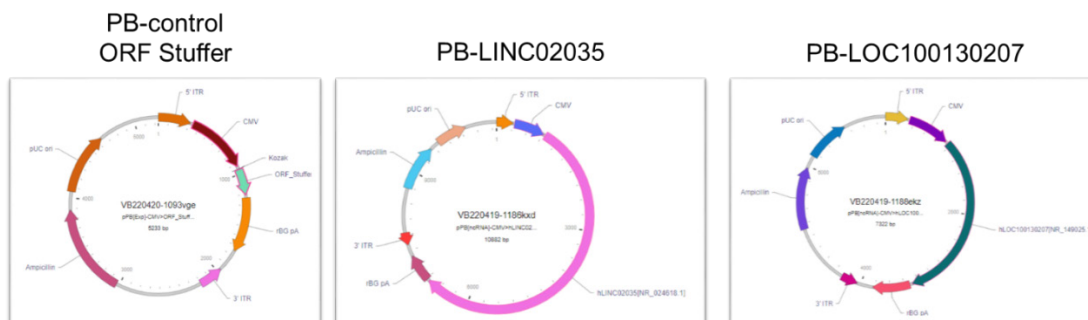
**-Supplementary figures-**



**Supplementary Fig. S1. qPCR confirmation for RNA-sequencing. (A) Validation of 10 selected upregulated mRNAs and (B) 5 selected downregulated mRNAs. For more details, please see Table S1 and S2.**



**Supplementary Fig. S2. Effects of RUNX2 downregulation or overexpression on expression of LINC02035 or LOC100130207 in human primary chondrocytes.** qPCR analysis was performed to determine the effects of RUNX2 knockdown on upregulated expression of LINC02035 or LOC100130207 in (A) hypertrophy-induced medium (HM)- or (B) TNF- $\alpha$ -treated human primary chondrocytes. (C) Conversely, qPCR analysis was performed to determine the effects of RUNX2 overexpression on the expression of both lncRNAs in human primary chondrocytes.



**Supplementary Fig. S3. Information of the vectors used for lncRNA overexpression.**

**Table S5. List of primers used in the present study.**

Primers	Sequence or Cat. No. of validated primers
<i>β-ACTIN</i>	5'-GTCCTCTCCCAAGTCCACACA-3' (sense, NM_001101.3) 5'-GGGCACGAAGGCTCATCATTC-3' (antisense)
<i>IHH</i>	Cat. No- P101104 V (RNA accession: NM_002181.3) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>RUNX2</i>	Cat. No- P229954 V (RNA accession: NM_001015051.3) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>COL10A1</i>	Cat. No- P261909 V (RNA accession: NM_000493.3) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>PTGES</i>	Cat. No- P327048 V (RNA accession: NM_004878.4) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>BGLAP</i>	Cat. No- P128146 V (RNA accession: NM_199173.5) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>LINC02593</i>	5'-CCGAGCTTCAGAGGAGAGG-3' (sense, NR_026874.2) 5'-CTCCCAGATCGTGAGCTTTC-3' (antisense)
<i>IGFL2-AS1</i>	5'-GGTGCCGAAGACCTGAGAC-3' (sense, NR_135234.1) 5'-GGCCACTTCTTCATTCTTGTTGG-3' (antisense)

<i>HCG4B</i>	5'-GTCATTCACCGGCCTAGCTC-3' (sense, NR_001317.3) 5'-GAGTATTGGGACCGGAGCAC-3' (antisense)
<i>LINC02035</i>	5'-GAAGCAAGCCAGAAAGCCTG-3' (sense, NR_024618.1) 5'-GTGGTGCAGAGCCATGAAAG-3' (antisense)
<i>LOC100287175</i>	5'-GATCAGTCTCTCTGCGCCTC-3' (sense, NR_149003.1) 5'-GTGTCTCTTGGCCCTGTACC-3' (antisense)
<i>LMNTD2-ASI</i>	5'-CAGGCACTCACCTACCTGAC-3' (sense, NR_147607.1) 5'-CCTGGAGCAGAGGGAATACTTG-3' (antisense)
<i>LOC100287808</i>	5'-ACTGGGAACTCAAGGCACAG-3' (sense, NR_149004.1) 5'-AGATTCGGCTCAAAGGAGGC-3' (antisense)
<i>LOC100130207</i>	5'-GCACAACAGGACAAAGGAAGG-3' (sense, NR_149025.1) 5'-CAGATCGCCGTTTGCTTGG-3' (antisense)
<i>H19</i>	5'-AAGAAATGGTGCTACCCAGC-3' (sense, NR_002196.2) 5'-GTGCAGTGGTTGTAAAGTGC-3' (antisense)
<i>OBSCN-ASI</i>	5'-CTCCTCGATGCCGTACTION-3' (sense, NR_073154.1) 5'-CCACGTTCCCTATGTGAGGTG-3' (antisense)
<i>FOXD2-ASI</i>	5'-CCTCCTACCAATTACACGCC-3' (sense, NR_026878.1) 5'-GTACACACAGGTCGCTATGG-3' (antisense)
<i>LOC101927811</i>	5'-TTAAACGAGCCCTGGATCTG-3' (sense, NR_110119.1) 5'-CCCGAAACAGACTTCTGAAC-3' (antisense)

<i>ARHGAP5-ASI</i>	5'-ATCGCTCGCCAACTACAGAC-3' (sense, NR_027263.1) 5'-TGGCTTCTCGCTCCACTTTC-3' (antisense)
<i>A2M-ASI</i>	5'-GGGTAGCATAGTGCCCAAGG-3' (sense, NR_137424.1) 5'-TTTATTGGAGAGGGCCGCTG-3' (antisense)
<i>LOC100129034</i>	5'-TTGCTGATGACGCAGAGGTC-3' (sense, NR_027406.1) 5'-GGACATTATTGCCGCCGTTC-3' (antisense)
<i>TSC22D1-ASI</i>	5'-GAGAGCGAGCTTCGGAAAGG-3' (sense, NR_038381.1) 5'-TTCCTTTGAACGGCGTCGG-3' (antisense)
<i>LOC90246</i>	5'-CTCCCAACCTCACCTGTGC-3' (sense, NR_026954.1) 5'-TAGCAGGGCTTTGGATGTGC-3' (antisense)
<i>SNAP25-ASI</i>	5'-AGCACCTAACGACAACAGTC-3' (sense, NR_040710.1) 5'-AGAACCCAGCTCCATTCATCC-3' (antisense)
<i>LIFR-ASI</i>	5'-TTACAGGTGTCTGTGCGGTG-3' (sense, NR_103554.1) 5'-TCCAGTCAATCCTTGGCATCC-3' (antisense)
<i>MKLN1-AS</i>	5'-ATCTCAGGGTTTCCCACGTC-3' (sense, NR_125364.1) 5'-ACTGTCCAGGCTTTCAGGAG-3' (antisense)
<i>LINC02015</i>	5'-ACAGCCGGGAAGAAAATTGT-3' (sense, NR_110826.1) 5'-CCAGAAGTTCATGGCAGCAG-3' (antisense)
<i>SOX9</i>	Cat. No- P232240 V (RNA accession: NM_000346.3) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>

<i>COL2A1</i>	Cat. No- P298511 V (RNA accession: NM_001844.4) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>SULF2</i>	Cat. No- P102960 (RNA accession: NM_001161841.1) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>FGFR4</i>	Cat. No- P164132 (RNA accession: NM_001291980.1) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>TGFBR3</i>	Cat. No- P270889 (RNA accession: NM_001195683.1) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>WNT6</i>	Cat. No- P204423 (RNA accession: NM_006522.3) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>ADAMTS5</i>	Cat. No- P127505 (RNA accession: NM_007038.4) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>MMP13</i>	Cat. No- P221090 (RNA accession: NM_002427.3) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>IGFBP3</i>	Cat. No- P123087 (RNA accession: NM_00598.4) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>COL9A2</i>	Cat. No- P288169 (RNA accession: NM_001852.3) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>DDR1</i>	5'-GCGTCTGTCTGCGGGTAGAG-3' (sense, NM_001202521.1) 5'-ACGGCCTCAGATAAATACATTGTCT-3' (antisense)

<i>RUNX3</i>	Cat. No- P272246 (RNA accession: NM_001031680.2) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>HBEGF</i>	Cat. No- P166323 (RNA accession: NM_001945.2) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>GREM1</i>	Cat. No- P224682 (RNA accession: NM_001191322.1) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>NGF</i>	Cat. No- P319108 (RNA accession: NM_002506.2) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>
<i>CXCL6</i>	5'-GGGAAGCAAGTTTGTCTGGACC-3' (sense, NM_002993.4) 5'-AAACTGCTCCGCTGAAGACTGG-3' (antisense)
<i>ADAM9</i>	Cat. No- P104329 (RNA accession: NM_003816.2) <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a>

**Table S6. siRNAs used in the current study.**

siRNAs	Dose (nM)	Sequence or Cat. No of validated siRNAs
Negative control	100	Sense siRNA targeted sequence 5-CCUACGCCACCAAUUUCGU-3' antisense siRNA targeted sequence 5-ACGAAAUUGGUGGCGUAGG-3'
<i>IGFL2-AS1</i>	100	RNA-GAC UCU CUU CAU AUG GAC U=tt(1-AS) RNA-AGU CCA UAU GAA GAG AGU C=tt(1-AA)
		RNA-CUG CAA AGA AGG AAC ACU A=tt(2-AS)



<i>HCG4B</i>	100	RNA-UAG UGU UCC UUC UUU GCA G=tt(2-AA)
<i>LINC02035</i>	100	RNA-CAG GAU UGA GGA AUC UGU A=tt(3-AS) RNA-UAC AGA UUC CUC AAU CCU G=tt(3-AA)
<i>LOC100287175</i>	100	RNA-GCA GCC UCC UCU UGU UCC U=tt(4-AS) RNA-AGG AAC AAG AGG AGG CUG C=tt(4-AA)
<i>LMNTD2-AS1</i>	100	RNA-CUG ACU UGC UGA AAC UAC A=tt(5-AA) RNA-UGU AGU UUC AGC AAG UCA G=tt(5-AA)
<i>LOC100287808</i>	100	RNA-CAA UUA CCU CCA GCA UAU =tt(6-AS) RNA-UAU AUG CUG GAG GUA AUU G=tt(6-AA)
<i>LINC02593</i>	100	RNA-GAU UAC AGA GUC AUU CAU U=tt(7-AS) RNA-AAU GAA UGA CUC UGU AAU C=tt(7-AA)
<i>LOC100130207</i>	100	RNA-CAG AAC UGC CAG CAG ACU A=tt(8-AS) RNA-UAG UCU GCU GGC AGU UCU G=tt(8-AA)
<i>RUNX2</i>	100	siRNA ID- 860-1 (Bioneer; <a href="https://www.bioneer.co.kr/sirna-customorder.html">https://www.bioneer.co.kr/sirna-customorder.html</a> )