

Supplementary information

Periodic heat stress licenses EMSC differentiation into osteoblasts via YAP signaling pathway activation

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Wentao Shi, Zhe Wang and Lu Bian contributed equally to this work.

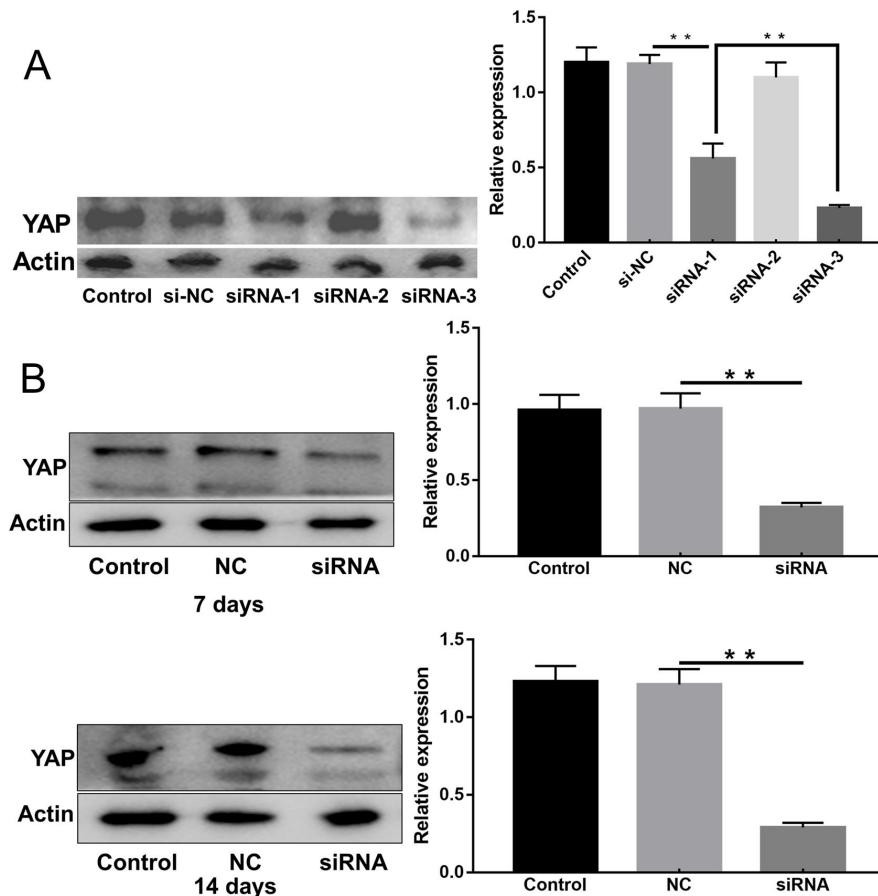


Fig. S1 (A) Western blot showing the effectiveness of YAP knockdown in EMSCs. (B) Expression

level of YAP was suppressed in siRNA-transfected EMSCs in each time point (Day 7, Day 14).

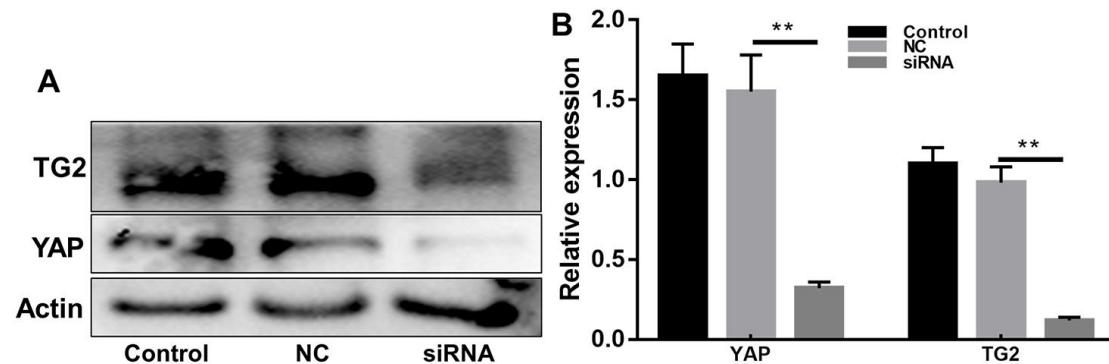


Fig. S2 The expression of YAP and TG2 was analyzed using Western blotting.

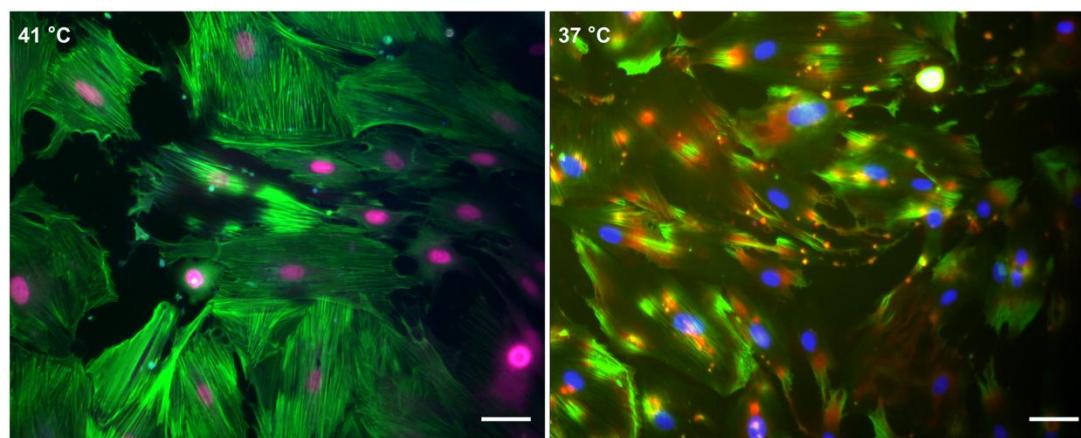


Fig. S3 Merged images showing YAP (red), TRITC (phalloidin) and UV (DAPI) fluorescence.

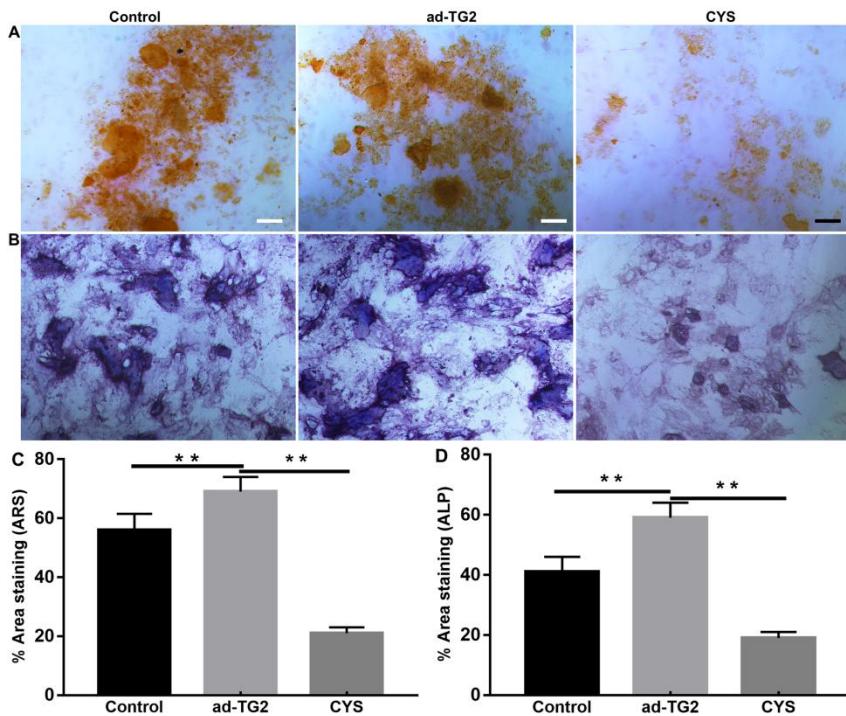


Fig. S4 Overexpression of TG2 promoted EMSC osteogenesis. On day 14 postinduction, the calcified nodules and ALP activity were visualized using ARS and ALP staining, respectively, and the results showed that overexpression of TG2 promoted EMSC osteogenesis.

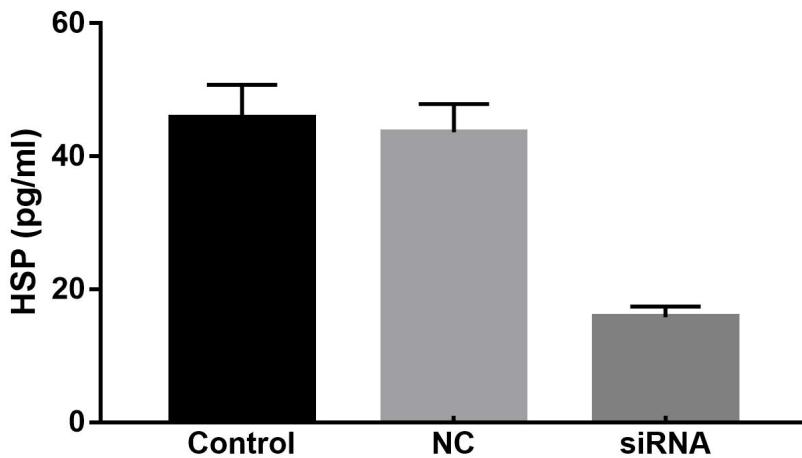


Fig. S 5 Cell supernatants were monitored for HSP secretion using ELISA.

Table S1. Primer sequence for qRT-PCR.

Gene	Forward primer sequence (5'-3')	Reverse primer sequence (5'-3')
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OPN	TGTGAAACTCGTGGCTCTGA	GAACCAAGCGTGGAAACACA
RUNX2	TCATGGCCGGGAATGATGAG	CGCTCCGGCCTACAAATCTC
OCN	ACAGTGACCTGAGTGAGGGT	GGGACTCCTGGCTGTTCATC
COL 1	CCTGGTGCTGATGGACAACC	TTTAGCCCCAAGGGTGAAGGG
GAPDH	ACATTGGGGTAGGAACACG	GCCATCACTGCCACTCAGAA
YAP	ACCATAAGAACAAAGACCACATCC	TTCAATCGCAGCCTCTCCTT
