

Supplementary Material

Maternal *DDB1* regulates apoptosis and lineage differentiation in porcine preimplantation embryos

Biao Ding^A, Di Gao^B, Xuegu Wang^A, Lei Liu^A, Junpei Sun^A, Meng Liang^C, Fengrui Wu^D, Yong Liu^D, Yunhai Zhang^B, Xiang Li^{A,}, and Wenyong Li^{D,*}*

^AReproductive Medicine Center, The First Affiliated Hospital of Bengbu Medical College, Bengbu 233004, China.

^BCollege of Animal Science and Technology, Anhui Agricultural University, Hefei 230036, China.

^CSchool of Life Science, Bengbu Medical College, Bengbu 233030, China.

^DAnhui Province Key Laboratory of Embryo Development and Reproductive Regulation, Fuyang Normal University, Fuyang 236041, China.

*Correspondence to: Xiang Li Reproductive Medicine Center, The First Affiliated Hospital of Bengbu Medical College, Bengbu 233004, China Email: xiangli@bbmc.edu.cn; Wenyong Li Anhui Province Key Laboratory of Embryo Development and Reproductive Regulation, Fuyang Normal University, Fuyang 236041, China Email: liwenyong@fynu.edu.cn

Supplementary figures

Figure S1

Fig.S1. Injection efficiency, survival rate and interference efficiency after siRNA injection. (a) Representative images of MII oocytes recovered 30 minutes after FAM-siRNA injection. BF: bright field. FAM: green fluorescence. Scale bar: 200 μ m. (b) The rate of MII oocytes with green fluorescence after FAM-siRNA injection and recovered 30 minutes (at least 10 oocytes per group; n = 5). (c) The survival rate of MII oocytes recovered 30 minutes after FAM-siRNA injection (at least 10 oocytes per group; n = 5). (d) Interference efficiency of *DDB1*-siRNA1 at different concentrations (10 embryos per pool, n = 3). All data are presented as mean \pm SEM and different letters on the column indicate significant differences ($p < 0.05$).

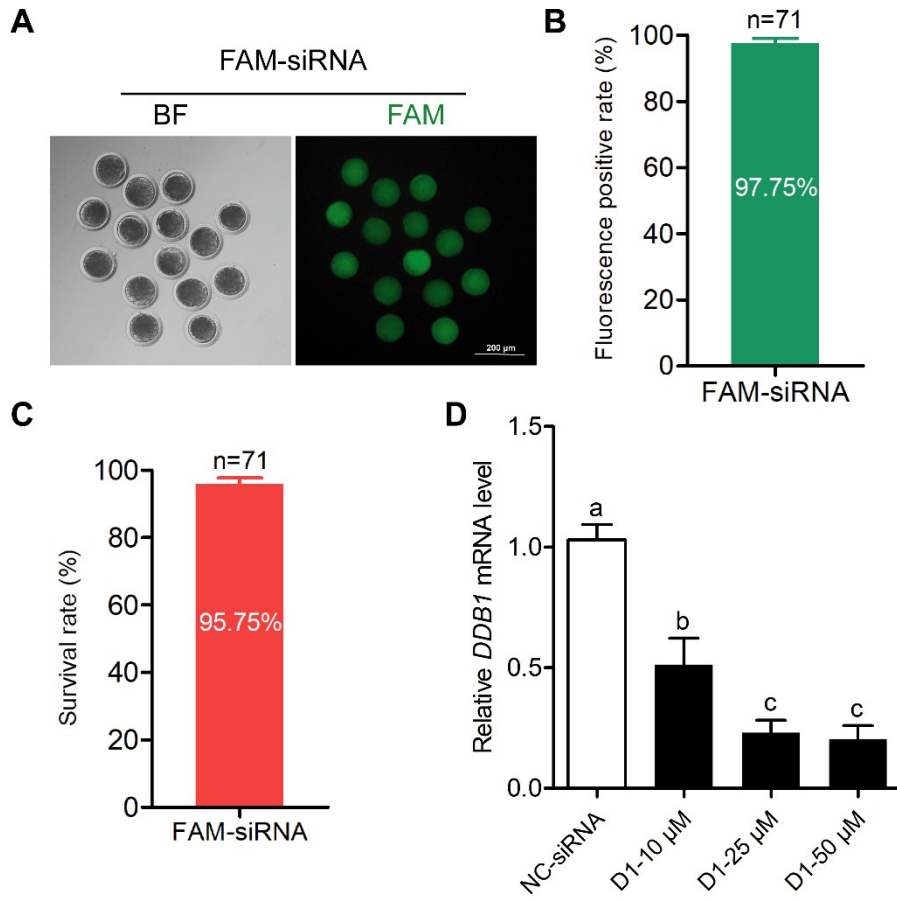


Figure S2

Fig.S2. Effect of *DDB1* deletion on the in vitro development of embryos derived from PA. (a) Representative images of PA embryos at different developmental stages. Scale bar: 100 μm . (b) Developmental rate of embryos at different stages. The number of survived oocytes after PA was regarded as the initial data and the embryos of all other stages were compared with it (at least 20 oocytes per group; $n = 4$). Data are presented as mean \pm SEM, and the different letters indicate significant differences ($p < 0.05$). (c) Sectional fluorescence staining images of the *DDB1*-KD and the control. Nuclei (left panel, blue), CDX2 (middle panel, green), and the merged images between the DNA and CDX2 (right panel, cyan). Scale bar: 100 μm . (d) Detection of the quality of the blastocysts by an analysis of the number of total cells, TE cells, ICM cells, and the ratio of the ICM to TE (at least 10 blastocysts per group; $n = 3$). Data are shown as mean \pm SEM and the different letters indicate the significant differences ($p < 0.05$).

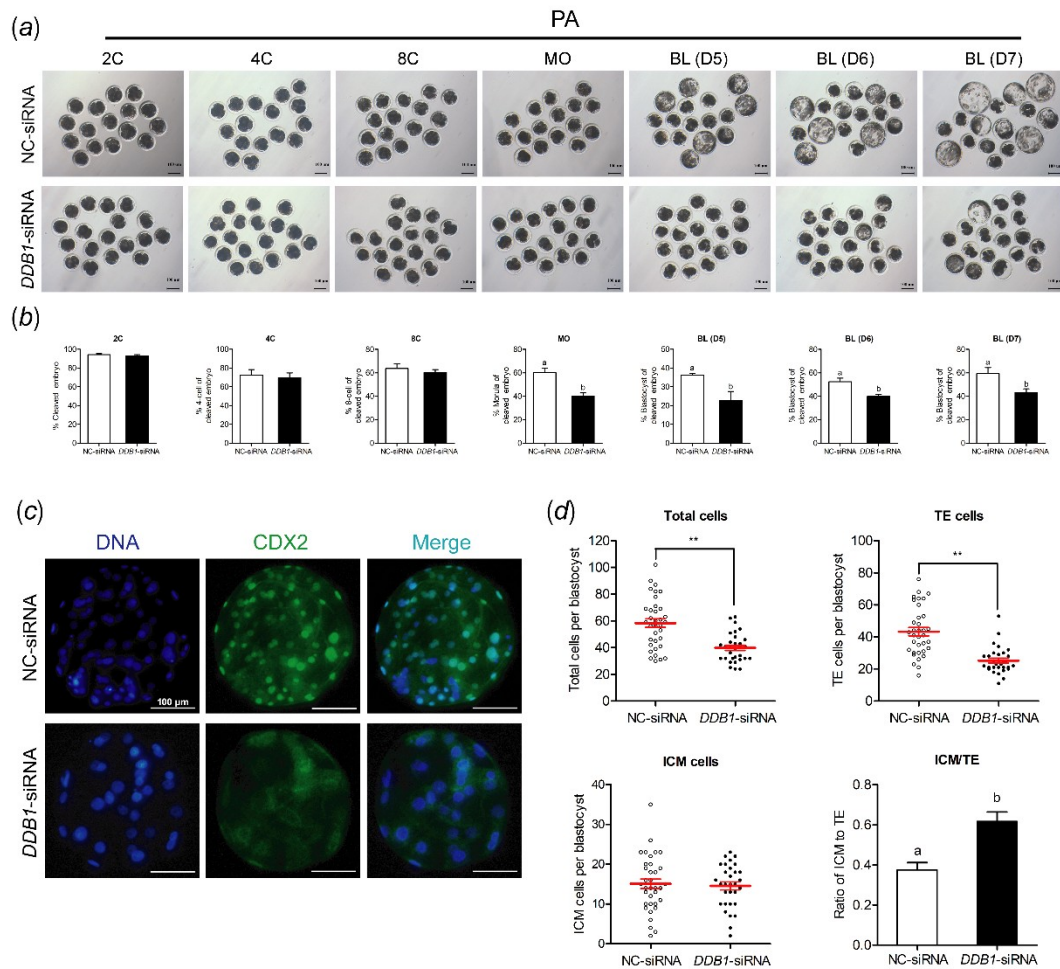
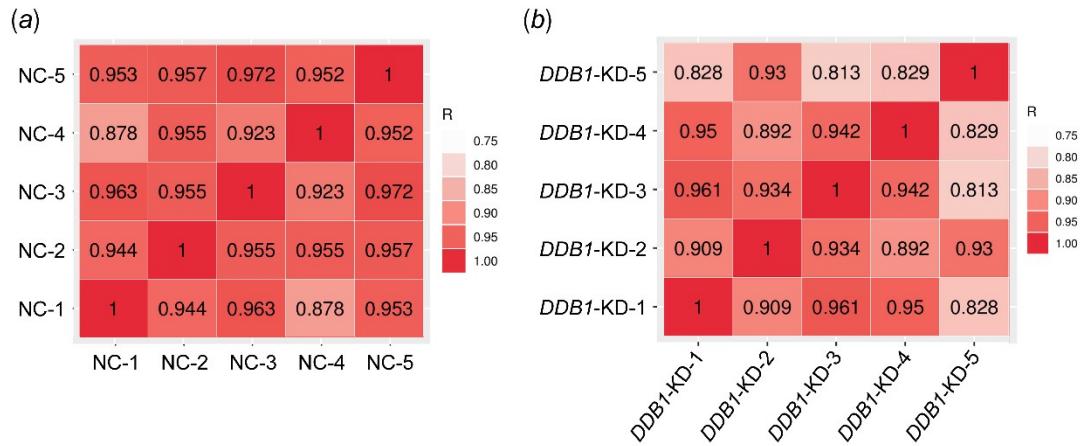


Figure S3

Fig.S3. Correlations of five biological replicates from each group. (a) The transcriptomes of four biological replicates from the NC group. (b) The transcriptomes of four biological replicates from the *DDB1*-KD group. R: Pearson correlation coefficient.



Supplementary tables

Table S1. Component of modified tris-buffered medium

Components	g/100 mL	Con. (mM)
NaCl	0.6611	113.1
KCl	0.0224	3.0
CaCl ₂	0.0833	7.5
Tris	0.2324	2.0
D-Glucose	0.1982	11.0
Na-pyruvate	0.0550	5.0

Table S2 Information on *DDB1* siRNA sequences

No.	Sequence (5'-3')
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	Sense	Antisense
<i>DDB1</i> -siRNA1	GCACAAUUGUGUGUCACAATT	UUGUGACACACAAUUGUGCTT
<i>DDB1</i> -siRNA2	GCAUUGACUUACCAGGCAUTT	AUGCCUGGUAAGUCA AUGCTT

Table S3. Primers used for RT-qPCR

Gene symbol	Primer sequences (5'-3')	Product size (bp)	Accession number
<i>DDB1</i>	F: CTGTTGAGCGACCGTAAG R: GATAGCCATCTGAGTTGAGG	199	XM_003122651.6
<i>YAP1</i> (<i>YAP</i>)	F: TCAACCAAAGCACCCCTACC R: TCCTCTCCTTCTATGTTCAATTCC	121	XM_021062706.1
<i>EZH1</i>	F: TGGCTCTGTGCTGATTAGTG R: CCGTCCTCCTCCTCATCC	81	NM_001243206.1
<i>JAM3</i>	F: CCTGCTGCTGCTTCTACTG R: TTGTCTGTGAATCCGTAATGATAC	125	XM_021063173.1
<i>PHF1</i>	F: CTGCTGTGTATGTCGCTCTG R: TGTGCCCTCTCCCTCTCC	130	XM_005665891.3
<i>PPP1CB</i>	F: ATGCTGGTGGGAATGATGAGTG R: CGAGGTGGAGTGACAGGAC	127	NM_214184.2
<i>BMP15</i>	F: CTCCCTTCCACCTCTCCTG R: CATCCATCTCCGTCCAAGC	126	NM_001005155.2
<i>KPNA7</i>	F: CACCAGCACATCCAGCAG R: TAGCCACCGTCCACACTG	109	NM_001163411.1

<i>SH2B3</i>	F: ACCACTTCCAGCGTTCAC R: AGCGAGAGAGGGAGAAAGG	135	XM_001929523.5
<i>MAPK9</i>	F: AGAAGTAATGGATTGGGAAGAAAG R: TGATAGATGACGACTGAGAAGG	107	XM_005661441.3
<i>CD3E</i>	F: TTGCTGATGGTGGTGTATTAC R: GGGCTCATAGTCTGGATTGG	141	NM_214227.1
<i>ANXA1</i>	F: AGTTGTTGTGAAGTGTGCTAC R: TCTTATGACGAGTTCCATTACC	89	NM_001163998.1
<i>CSF1R</i>	F: ACTCTGTGGTTGCCTTGC R: TCTGTCTTGCTCTGCTCTTC	80	XM_003124100.6
<i>IL18</i>	F: GGCTCTCCACCTCCTCAC R: TATCTTGTTGTTGCTATCATCTCC	88	XM_021085847.1
<i>PIWIL2</i>	F: GCATCAATCTCACTCTCACAAG R: GGCACAACCTTCAGGCTATCC	87	NM_001194971.1
<i>FKBP6</i>	F: AACCGACCTACGCCTACG R: AACTTGTCCGACTCAGCAG	115	XM_021086319.1
<i>ERBB4</i>	F: ACTCTTCAGCACCATCAACC R: AAGCGACGACAGGACAGG	150	XM_021075968.1
<i>FRMD4B</i>	F: GGAAAGATTTGAAGACACTACCC R: ATACTGAACCACTGCTTGACC	140	XM_021069209.1
<i>DAB2</i>	F: GCACAGCCAGACCAATCG R: GCCCAAAGTAATCAACATCCC	133	XM_021076650.1
<i>TCIRG1</i>	F: CTGGTGGTCCTGGCTCTG R: AGTCTTGTCTCATCCTCTACC	120	XM_005660613.3

<i>OCT4</i>	F: CGAGAACCGAGTGAGAGG	206	NM_001113060.1
<i>(POU5F1)</i>	R: GGAAAGGAGACCCAGCAG		
<i>SOX2</i>	F: CGCAGACCTACATGAACG	103	NM_001123197.1
	R: TCGGACTTGACCACTGAG		
<i>CDX2</i>	F: AGTCGCTACATCACCATTTCGGAG	139	NM_001278769.1
	R: GCTGCTGTTGCTGCAACTTCTTC		
<i>NANOG</i>	F: CTCTCCTCTTCCTTCCTC	139	NM_001129971.1
	R: CTTCTGCTTCTTGACTGG		
<i>TEAD4</i>	F: CATTACTCCTACCGCATCC	151	NM_001142666.1
	R: CCTGTGTGTCTCTGTTGG		
<i>GATA3</i>	F: CACGACACGCTGGAGGAC	106	NM_001044567.1
	R: GGCTGGAGTGGCTGAAGG		
<i>H2AFZ</i>	F: CCAAGACAAAGGCGGTTTCC	94	NM_001123122.1
	R: TGGCTGGTCGTCCTAGATTTC		

F: forward, R: reverse