



Supplementary Figure 1. Derivation and morphology of bovine embryonic cells (ECs). (A) Strategy for derivation of ECs. By injecting the nuclei of adult fibroblasts (AFs) into enucleated oocytes, reconstructed oocytes were obtained. When the cloned embryos developed to the stage of hatching or hatched blastocysts, the zona pellucida (ZP) was removed and 2-3 blastocysts per culture insert were placed in the 8-well IbiTreat μ -plates. Outgrowths from the attached blastocysts were seen and left to expand until day 5–7. ECs were morphologically large cuboidal cells. (B) Intact oocytes. The MII is pointed by an arrow. (C) Enucleated oocyte. (D–E) The procedure of injecting the nuclei of AFs and cytoplasm into enucleated oocytes. (F) Fused embryos. (G) Hatched embryo. (H–I) Proliferating outgrowths from one hatched blastocyst on the plates. (J) AFs-derived ECs. (K) DAPI staining in the amplified ECs.

Supplementary table 1. Primer sequences for RT-qPCR.

Genes	Primer sequence	Accession no.	Product size (bp)
<i>NANOG</i>	F: CGTGTCTTGCAAACGTCAT R: CTGTCTCTCCTCTTCCCTCCTC	DQ069776	66
<i>OCT4</i>	F: CAACAACGAGAATCTGCAGG R: ATGTGGCTAATTTGCTGCAG	AY490804.1	153
<i>SOX2</i>	F: GGTTGACATCGTTGGTAATTTATAATAGC R: CACAGTAATTTTCATGTTGGTTTTTCA	NM_001105463	88
<i>CDX2</i>	F: GCAAAGGAAAGGAAAATCAACAA R: GGGCTCTGGGACGCTTCT	XM_871005	120
<i>p53</i>	F: CCTCCCAGAAGACCTACCCCT R: CTCCGTCATGTGCTCCAACCT	X81704.1	221
<i>Bax</i>	F: GGCTGGACATTGGACTTCCTTC R: TGGTCACTGTCTGCCATGTGG	NM_173894.1	112
<i>Caspase 3</i>	F: TACTTTTCCTGGCGAAATGC R: TTGCATGAAAAGCAGAATCG	NM_001077840	169
<i>BCL2</i>	F: TGGATGACCGAGTACCTGAA R: CAGCCAGGAGAAATCAAACA	NM_001166486.1	120
<i>DNMT1</i>	F: AGGGAGACGTGGAGATGCTG R: CATGGAGCGCTTGAAGGAG	AY244709	194
<i>DNMT3a</i>	F: AGACATGTGGGTGAACCCG R: GGCTCCCACAAGAGATGCAG	AY271298	188
<i>DNMT3b</i>	F: CAGGATGGGAAGGAGTTTGGA R: CACCAAACCACTGGACCCAC	AY244710	151
<i>HDAC1</i>	F: ACCTTTATCCCACAACCCTTCA R: TCCCTTTTACCCAGTACCCATT	NM001037444	201
<i>HDAC3</i>	F: AAGTTTGAGGCTTCTGGTTT R: GACTCGGTCAGTGAGGTAGA	NM0012062431	150
<i>GAPDH</i>	F: CCCAGAATATCATCCCTGCT R: CTGCTTCACCACCTTCTTGA	NM_001034034	185

Supplementary Table 2. Characterization of day 8 bovine blastocysts of three groups.

Groups	No. of blastocysts	No. of total cells	No. of TE cells	No. of ICM cells	ICM:TCN cell ratio
IVF	10	191.7 ± 3.9	144.4 ± 4.4	49.2 ± 1.8a	34.8 ± 1.6a
AF-CICT	10	177.3 ± 0.9	141.2 ± 3.9	30.8 ± 0.9c	21.0 ± 0.7c
EC-CICT	10	187.9 ± 4.9	145.4 ± 5.2	42.5 ± 1.2b	29.7 ± 1.6b

^{a-c} Values with different superscripts denotes significantly different within same column ($p < 0.05$).
Data are mean ± SEM of at least five independent replicates.

Supplementary Table 3. Apoptotic analysis of day 8 bovine blastocysts of three groups.

Groups	No. of blastocysts	No. of total cells	No. of apoptotic cells	Apoptotic cells rate
IVF	10	194.0 ± 9.9	5.4 ± 0.5 ^a	2.9 ± 0.3 ^a
AF-CICT	10	172.1 ± 7.5	4.7 ± 0.4 ^a	2.8 ± 0.4 ^a
EC-CICT	10	190.5 ± 5.2	3.1 ± 0.3 ^b	1.6 ± 0.2 ^b

^{a-c} Values with different superscripts denotes significantly different within same column ($p < 0.05$). Data are mean ± SEM of at least five independent replicates.

Supplementary Table 4. Primary antibodies used for immunofluorescence.

Primary Antibody	REF no.	Company
CDX-2	Am-392	Biogenex
H3K56ac	ab71956	Abcam
H3K9me2	ab1220	Abcam
NF- κ B	sc-271908	Santa Cruz Biotechnology
CDK-2	NU-906	Biogenex
CDK-4	PA5-27827	Invitrogen
DAPI	62248	Thermo Fisher scientific
FITC	F-2765	Invitrogen
TRITC	A16071	Invitrogen