46,XX (38/51)

Figure S1. Representative normal karyotype of iRSC derived from female TIG1 fibroblasts, Related to Figure 1.
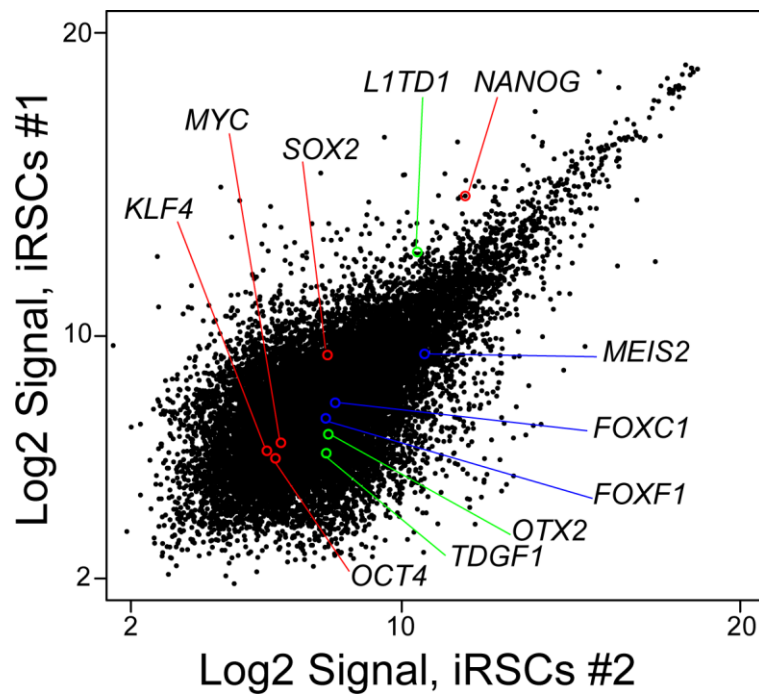
Figure S2. Comparison of global gene expression profile between iRSC lines. Related to Figure 2. Scatter plot of gene expression in iRSC-line #1 and #2. Red circle; endogenous OCT4, SOX2, KLF4, MYC and NANOG, Blue circle; somatic genes, Green circle; pluripotency genes.
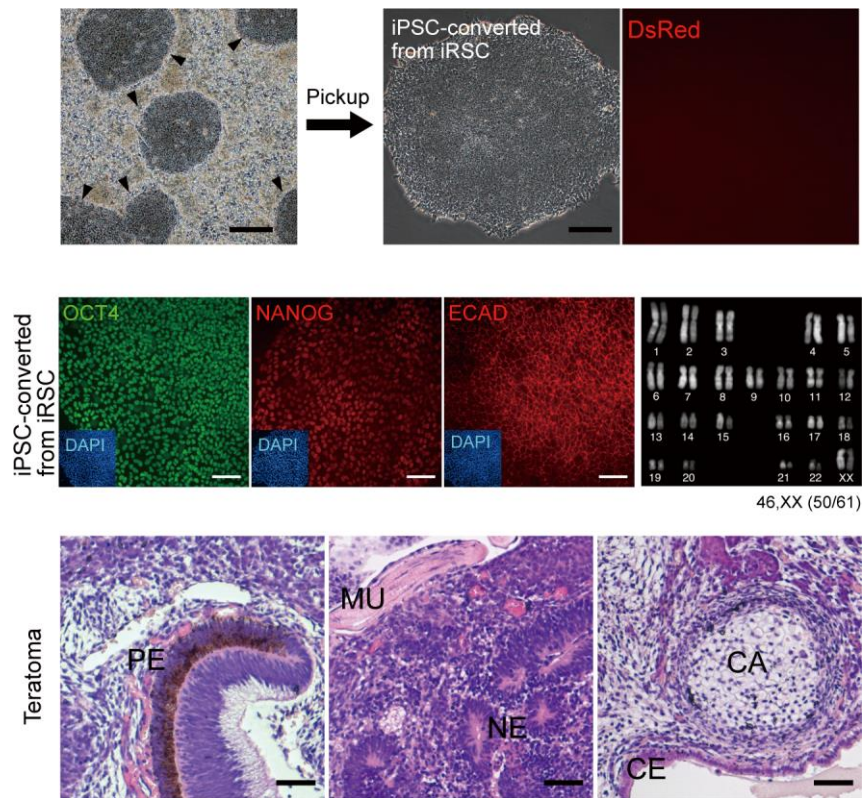
Figure S3. Properties of iPSC-derived iRSCs, Related to Figure 3.

Several colonies of iPSCs (black arrow heads) are observed in iRSCs at day 10 after culture at high cell density. Isolated iPSC colony is negative for the exogenous DsRed reporter gene. Scale bar: 300μm (upper row). Expression of pluripotency-associated proteins is visualized by immuno-staining. Scale bar: 100μm (middle row). iRSC-converted iPSC has normal karyotype, 2n=46, XX (right in middle row). Teratomas are generated by transplantation of iRSCs into kidney capsule. Scale bar: 50μm. PE; pigmented epithelial cells, MU; muscle, NE; neuronal ectoderm, CA; cartilage, CE; cilia-epithelial cell.
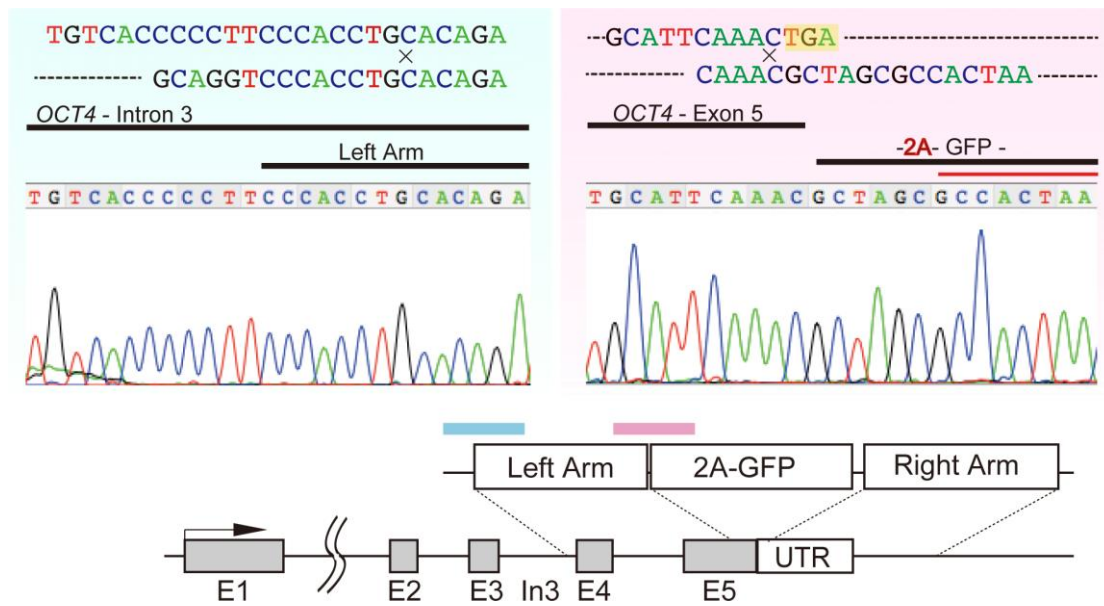
Figure S4. Correct insertion of OCT4-GFP. Related to Figure 4.

DNA sequence analysis shows that OCT4-GFP was inserted into the correct site. Left (blue) figure corresponds to the blue bar (around intron 3 of OCT4) and Right (pink) figure corresponds to the pink bar (around of 2A-GFP) in the bottom diagram.   E: Exon. In: Intron.
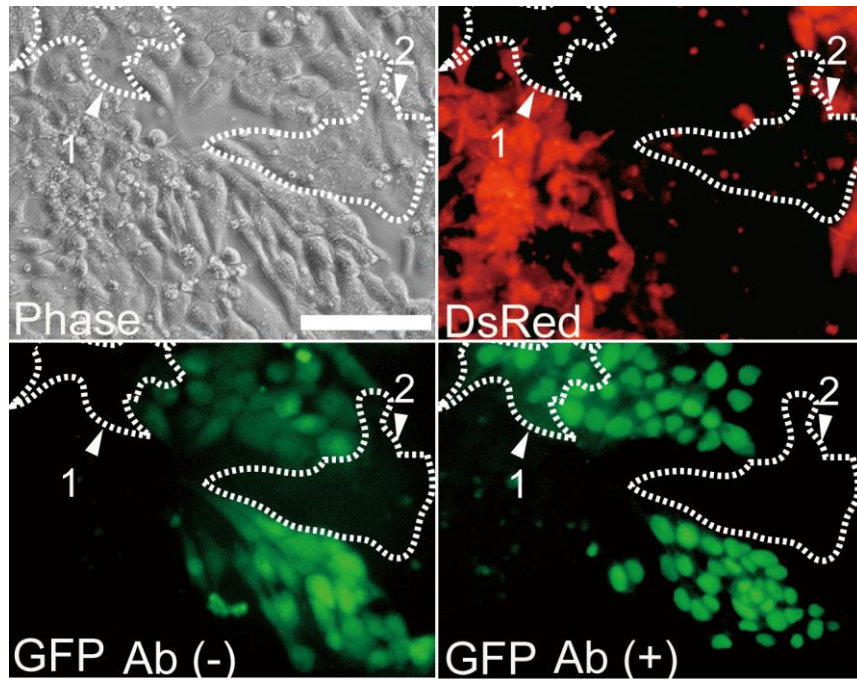
Figure S5. Instability of OCT4-GFP detected with anti-GFP antibody. Related to Figure 6.
DsRed, GFP without antibody, and GFP with antibody were detected in the same microscopic field five days after high-density culture of OG-iRSCs. White dotted lines (regions 1 and 2) surround DsRed-negative reprogramming OG-iRSC colonies. Cells in region 1, which were GFP negative without antibody became GFP positive with antibody, while cells in region 2 were GFP negative even with antibody, verifying the emergence of OCT4-negative post-MET iRSCs. Phase: Phase contrast. Scale bar: 100μm. Ab: Antibody. (-) No antibody. (+) with antibody.
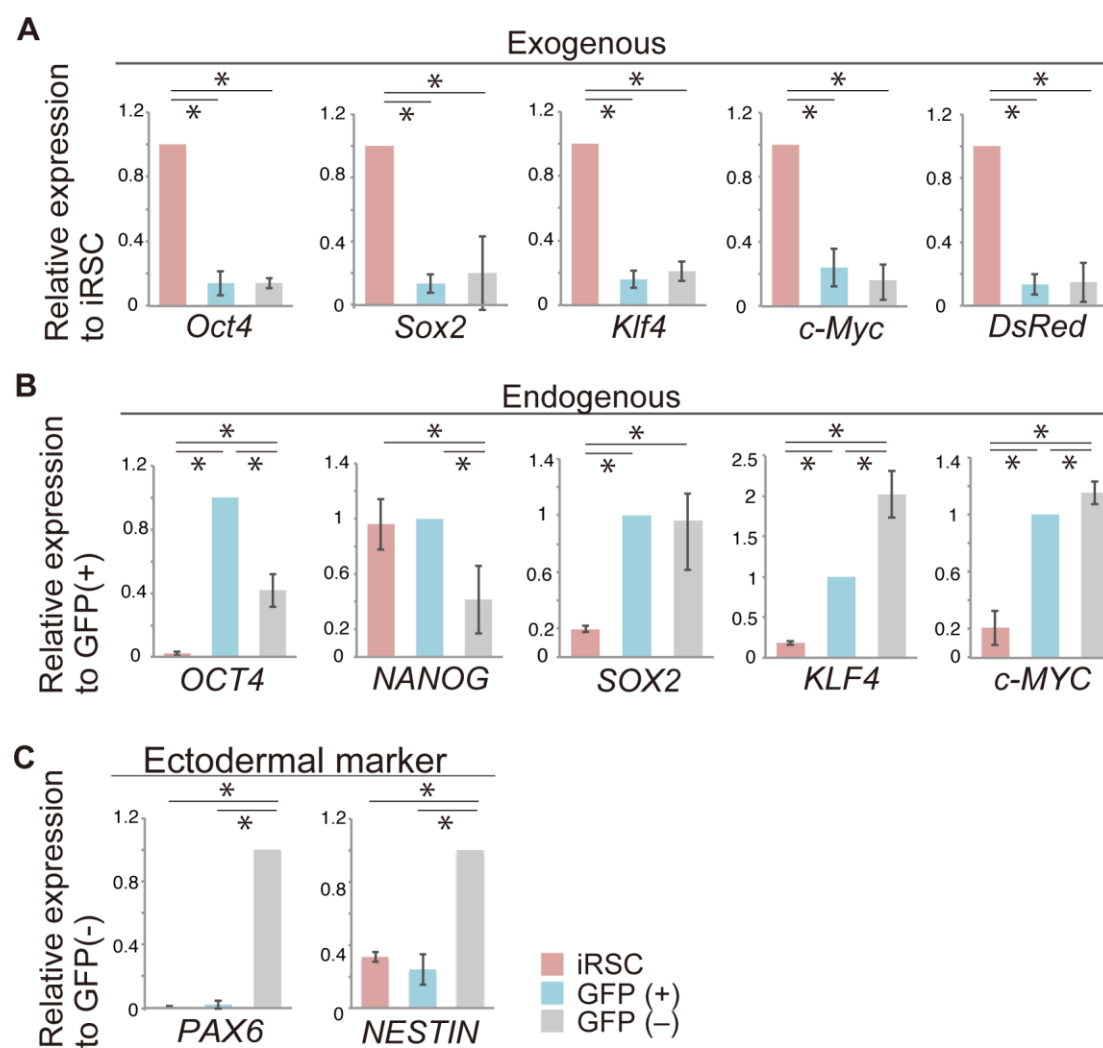
Figure S6. Gene expression in DsRed(-)/GFP(-) post-MET cells. Related to Figure 6.

(A)   Expression levels of exogenous Oct4, Sox2, Klf4, c-Myc and DsRed in DsRed-negative/GFP-negative cells by Q-PCR. Data are represented as mean ± standard deviation, Oct4, Klf4, c-Myc, DsRed: n = 4. Sox2: n=8

(B)   Expression levels of endogenous OCT4, NANOG, SOX2, KLF4 and c-MYC in DsRed-negative/GFP-negative cells by Q-PCR. Data are represented as mean ± standard deviation, OCT4, NANOG, SOX2, c-MYC: n=4. KLF4: GFP(+) n=6, GFP(-) n=5.

(C)   Expression levels of ectodermal marker genes, PAX6 and NESTIN in DsRed-negative/GFP-negative cells by Q-PCR. Data are represented as mean ± standard deviation, n=4.

iRSC (red bar): OCT4-GFP iRSC, GFP(+) (blue bar): DsRed-negative/GFP-positive cells, GFP(-) (gray bar): DsRed-negative/GFP-negative cells.
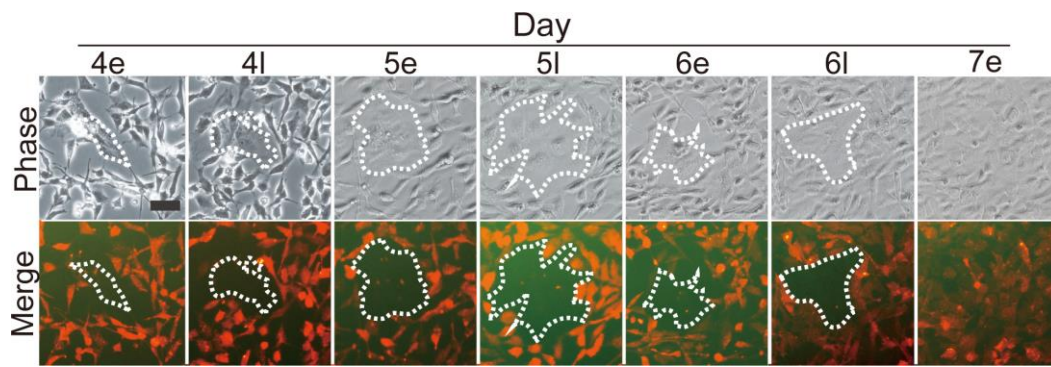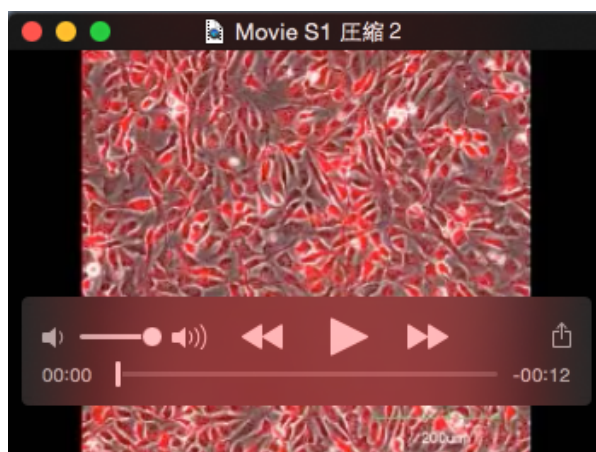
**Figure S7. No conversion of OCT4-GFP(-) cells to (+) cells. Related to Figure 6.**

Sequential change of the same OCT4-GFP-negative colony observed at 12 hourly intervals between day 4-7. Phase: phase contrast, Merge; merge of DsRed and GFP. Region enclosed with white dotted line: OCT4-GFP negative colony. Phase: Phase contrast. Scale bar: 50µm. e: early. l: late.

Movie 1. Conversion of iRSCs toward iPSCs between days 1-3, Related to Figure 3. Time-lapse images are captured from one day after reseeding, taken at a rate of 18 frames per 5 hours.
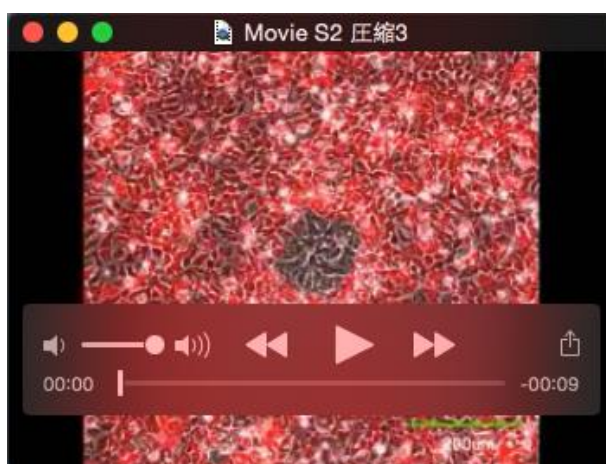
Movie 2. Conversion of iRSCs toward iPSCs between days 3-6, Related to Figure 3. Time-lapse images captured from three day after reseeding, taken at a rate of 18 frames per 5 hours.
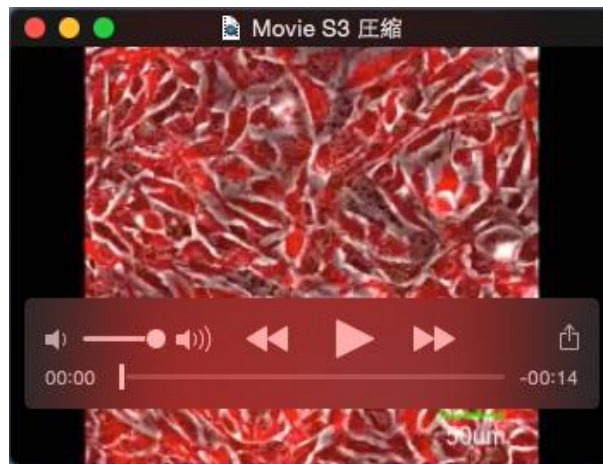
Movie 3. High frame rate images in initial stages of iRSC-to-iPSC conversion during entry into MET, Related to Figure 3. Time-lapse images captured between days 1-2 at a rate of 11 frames per hour.
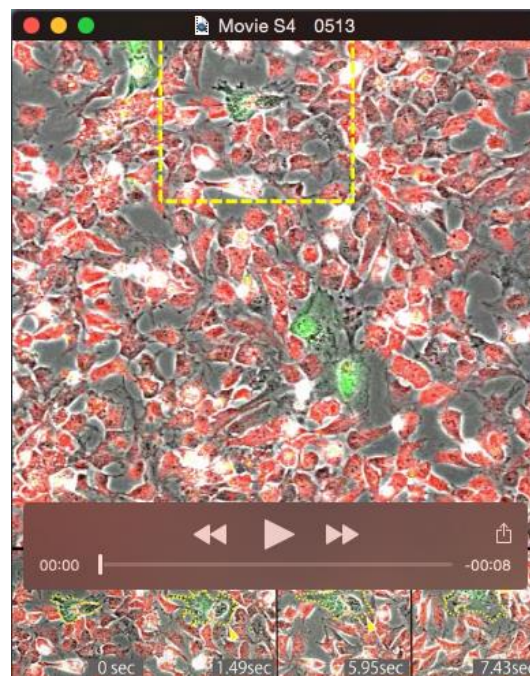
Movie 4. Generation of GFP-positive and negative cells through asymmetric cell division, Related to Figure 6. Time-lapse images are captured between days 2-4 at a rate of 3 frames per hour.

Table S1. Primers

	Target	Forward	Reverse
RT-PCR analysis	OCT4	GCACTGTACTCCTCGGTCCCTTTCCC	CTTCCCTCCAACCAGTTGCCCCAAAC
	SOX2	GGGAAATGGGGAGGGGTGCAAAAGAGG	TTGCGTGAGTGTGGATGGGATTGGTG
	KLF4	ACTCGCCTTGCTGATTGTCT	GAACGTGGAGAAAGATGGGA
	c-Myc	CGGGCGGGCACTTTG	GGAGAGTCGCGTCCTTGCT
	Tg-Oct4	CCCATGGTGGTGGTACGGGAATTC	AGTTGCTTTCCACTCGTGCT
	Tg-Sox2	CCCATGGTGGTGGTACGGGAATTC	TCTCGGTCTCGGACAAAAGT
	Tg-Klf4	CCCATGGTGGTGGTACGGGAATTC	GTCGTTGAACTCCTCGGTCT
	Tg-c-Myc	CAGAGGAGGAACGAGCTGAAGCGC	GACATGGCCTGCCCGGTTATTATT
	NANOG	AAAGAATCTTCACCTATGCC	GAAGGAAGAGGAGAGACAGT
	TDGF1	CCGCCCCGACTGGGGTTTGT	AAGCAGGAGCAAGGCGTCCAG
	REX1	TTAGCTAGGCCTGGTTGCAT	GGGCTCTTGCTGTTATCCAG
	ECAD	GAGCTTGTCATTGAGCCTGGCA	TGGGCAAATGTGTTCAGCTCAGC
	EPCAM	AATGTGTGTGCGTGGGACGA	GGTAAAGCCAGTTTCAAGCTGC
	OCLN	TCACACCCCAGACGATGTCTTCA	GGGAGGCTGGTAGATCATCACA
	EMP1	GCTGTCCCTCATGGAGACCT	AAGTGGGATAGGCAGGGTCC
	MMP1	CGCTGGGAGCAAACACATCT	TTCATGAGCCGCAACACGAT
	ZEB1	TCTGACTCTCAGCTCCTGCACT	GCCAGGCACCCTGTTAGGCA
	ZEB2	TTGTTACCTTCGCTGTGAATTGAA	GGACACAGCCTACTAGCCCAA
	SNAI2	TTCAAATGCATACCACAAATGCAAT	AGTGGTTTGGTACTAATCATGAAGC
	GAPDH	CTTCTTTTGCGTCGCCAGCCGAG	CAGCCTTGACGGTGCCATGGAA
Quantitative RT-PCR analysis	OCT4	GAGTGAGAGGCAACCTGGAG	ACACTCGGACCACATCCTTC
	SOX2	TAAGTACTGGCGAACCATCT	AAATTACCAACGGTGTCAAC
	KLF4	TGCCAAGGGGGTGACTGGAAGT	TCTTCCCTCCCCCAACTCACGG
	c-Myc	CGGGCGGGCACTTTG	GGAGAGTCGCGTCCTTGCT
	NONOG	TGGGATTGGGAGGCTTTGCT	TGAAACACTCGGTGAAATCAGGG
	Tg-Oct4	ACTAGCATTGAGAACCGTGTG	GGTGTCCCTGTAGCCTCATAC
	Tg-Sox2	GCGCCCAGTAGACTGCACA	ACATGTGCGACAGGGGCAG
	Tg-Klf4	TCCCTAGAGGCCCATTTGAG	GGGGACTTGTGACTGCATCT
	Tg-c-Myc	TGTGGAGAAGAGGCAAACCCC	TCCAAGACGTTGTGTGTCCG
	Tg-DsRed	TACGTGAAGCACCCCGCCGA	GCCGCCGTCCTCGAAGTTCA
	FOXC1	TTCGAGTCACAGAGGATCGG	TAGTTCGGCTTTGAGGGTGT
	FOXF1	TCTCGCTCAACGAGTGCTTC	TCATGCTGTACATGGGCTTG
	L1TD1	TATACTGTTGGGGGAGGGCT	CTTGCCATCTTTTCCCGTGC
	MEIS2	CAGTGTAGCTTCACCTGGTACA	TGGGCTGTACTATTCTTCTTCTGG
	OTX2	GTATGGACTTGCTGCACCCC	AAACCATACCTGCACCCTCG
	PPARG	CCTGCAGGAGCAGAGCAAA	GCCCTCGGATATGAGAACCC
	ZIC2	CAGAACGGCTTCGTTGACTC	AAGTCCCGGGTGGAGTTGAA
	NESTIN	TCCAGGAACGGAAAATCAAG	GCCTCCTCATCCCCTACTTC
	PAX6	GTCCATCTTTGCTTGGGAAA	TAGCCAGGTTGCGAAGAACT
	GAPDH	CTGGCCAAGGTCATCCATGAC	CCATCCACAGTCTTCTGGGTG
Bisulfite sequencing analysis	Bis-OCT4	GAAGGGGAAGTAGGGGATTAATTTT	CAACAACCATAAACACAATAACCAA
Genomic PCR	OCT4-lane1	GTCACAGACCCCTGTGATGC	AAGTCGTGCTGCTTCATGTG
	OCT4-lane2	GTCACAGACCCCTGTGATGC	CAACCAGTTGCCCCAAACTC
	OCT4-lane3	CGTAAACGGCCACAAGTTCA	GGGGTGTTCTGCTGGTAGTG
	GAPDH	GCCTCACTCCTTTTGCAGAC	TGAGCTTGACAAAGTGGTCG

Table S2. Antibodies for Immunocytochemistry

	Target	Dilution ratio	Manufacturer
Primary Antibodies	NANOG	1:200	ReproCELL, Japan
	SSEA4	1:500	Hybridoma Bank, USA
	TRA1-60	1:500	Millipore, USA
	ECAD	1:200	Takara, Japan
	OCT4	1:50	Santa Cruz Biotechnology, USA
	EGFP	1:1000	Nacalai, Japan
Secondary Antibody	Alexa 488	1:500	Molecular Probes, USA