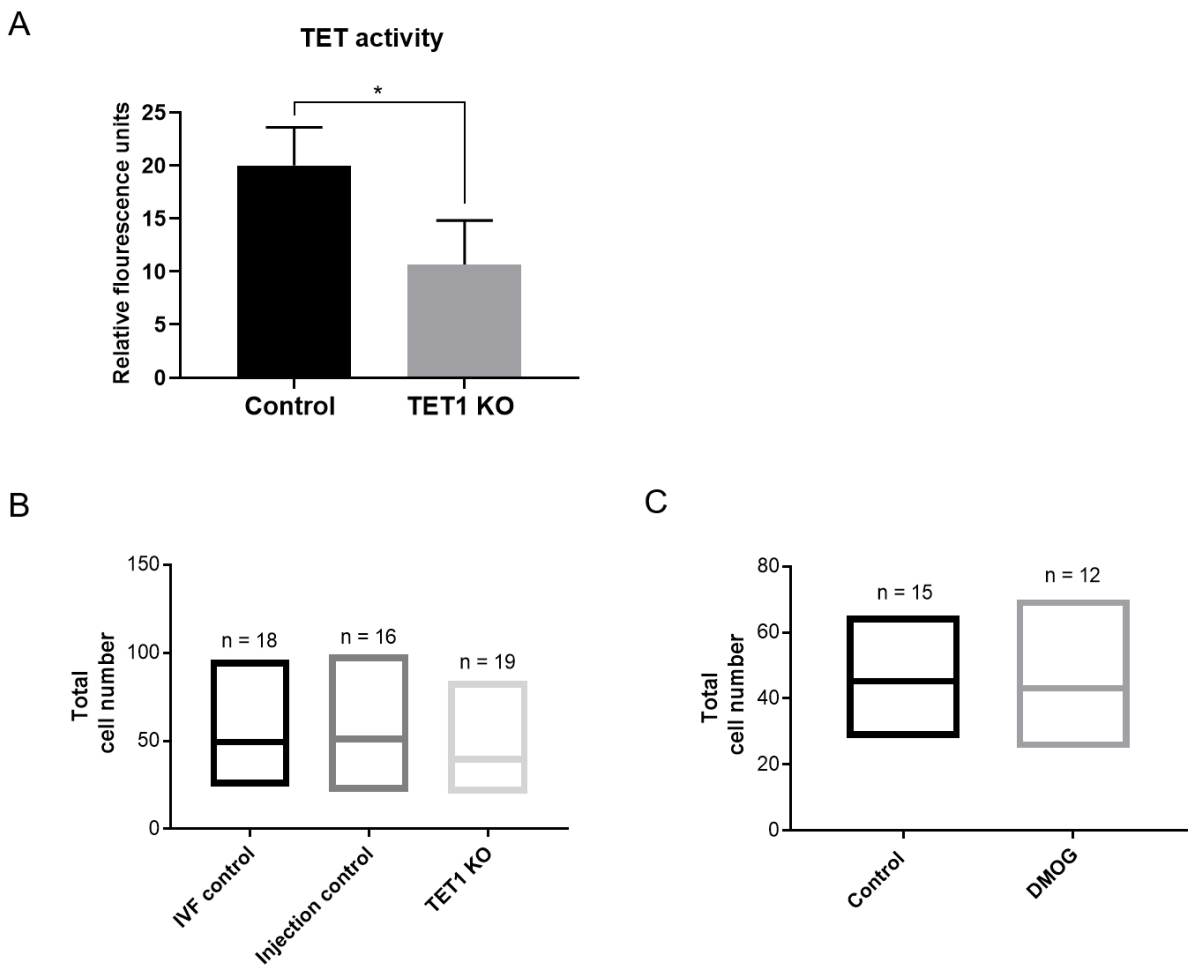

Fig. S1. TET activity and total cell number in TET1-disrupted and DMOG-treated blastocysts. (A) Measurement of TET enzyme activity in *TET1* KO blastocysts. Total TET enzyme activity in *TET1* KO blastocysts was decreased compared to that in control blastocysts. Error bars represent standard deviation. A p-value < 0.05 was considered statistically significant. (B) There was no significant difference of total cell number between IVF control, injection control (Cas9 only), and *TET1* KO blastocysts. (C) Total cell number of blastocysts incubated with DMOG was not different compared to that of control blastocysts. Top horizontal line and bottom horizontal line of the box indicate maximum and minimum values, respectively. Middle horizontal line indicates average value. A p-value < 0.05 was considered statistically significant.

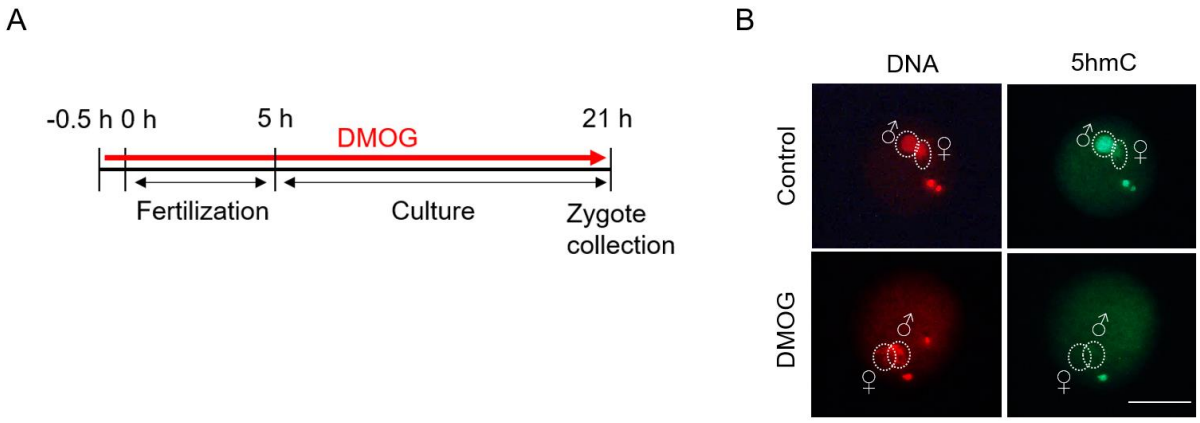

Fig. S2. The effect of DMOG treatment on TET3 inhibition in zygotes. (A) Timeline of DMOG treatment to inhibit TET3 activity in zygotes. Oocytes were incubated with 1mM DMOG for 30 min before fertilization and the treatment was maintained during fertilization and embryo culture for further 21 h, then zygotes were collected for immunocytochemistry analysis. (B) Inhibition of TET3 activity by DMOG treatment. In control zygotes, formation of 5hmC was detected in both paternal and maternal pronuclei. 5hmC was disappeared in pronuclei when zygotes were incubated with DMOG.

Table S1. Validation of targeting efficiency by CRISPR/Cas9 at *TET1* gene locus. All blastocysts genotyped carried mutations at the three target sites indicating 100% targeting efficiency.

Embryos genotyped (n=10)	Target site 1	Target site 2	Target site 3
Embryo #1	Homozygous	Homozygous	Mosaic w/ wildtype
Embryo #2	Homozygous	Homozygous	Mosaic w/ wildtype
Embryo #3	Homozygous	Biallelic	Mosaic w/ wildtype
Embryo #4	Biallelic	Biallelic	Mosaic w/o wildtype
Embryo #5	Biallelic	Biallelic	Mosaic w/o wildtype
Embryo #6	Biallelic	Homozygous	Mosaic w/o wildtype
Embryo #7	Biallelic	Homozygous	Mosaic w/ wildtype
Embryo #8	Biallelic	Biallelic	Mosaic w/o wildtype
Embryo #9	Homozygous	Biallelic	Mosaic w/ wildtype
Embryo #10	Biallelic	Biallelic	Mosaic w/ wildtype

Table S2. Development of *TET1* KO embryos to blastocyst stage. Frequency of blastocyst formation at day 7 decreased in *TET1* KO embryos compared to that in IVF control and injection control (Cas9 only) embryos. Different letters indicate statistical difference.

	Number of blastocysts at day 7 (%)	Number of oocytes used
IVF control	111 (28.8)[a]	385
Injection control (Cas9)	164 (18.5)[b]	885
TET1 KO	119 (12.4)[c]	958

Table S3. Development of embryos treated with DMOG. Frequency of blastocyst formation at day 7 decreased in embryos treated with DMOG compared to that in control embryos. Different letters indicate statistical difference.

	Number of blastocysts at day 7 (%)	Number of oocytes used
Control	304 (21.3)[a]	1429
DMOG	261 (17.6)[b]	1484

Table S4. Information of sgRNAs used for depletion of *TET1* gene.

sgRNAs	Sequence (5’ → 3’)
*TET1*_sgRNA1	TGTCTCGATCTCGCCATGCA
*TET1*_sgRNA2	GTGCTCATCATGGTATGGGA
*TET1*_sgRNA3	AGTCGAACCTGTACATGTCA

Table S5. Primers used for RT-qPCR analysis.

Primers	Sequence (5' → 3')
*GAPDH*_forward	ATGACATCAAGAAGGTGGTGAAGC
*GAPDH*_reverse	CCAGCATCAAAAGTGGAAGAGTGA
*OCT4*_forward	TTTGGGAAGGTGTTCAGCCAAACG
*OCT4*_reverse	TCGGTTCTCGATACTTGTCCGCTT
*NANOG*_forward	AGGACAGCCCTGATTCTTCCACAA
*NANOG*_reverse	AAAGTTCTTGCATCTGCTGGAGGC
*SOX2*_forward	TGTCGGAGACGGAGAAGCG
*SOX2*_reverse	CGGGGCCGGTATTTATAATCC
*KLF2*_forward	CGATCCTCCTTGACGAATTT
*KLF2*_reverse	CAAGCCTCGATCCTCTAGT
*ESRRB*_forward	CTGCAAGGCCTTCTTCAA
*ESRRB*_reverse	CGTTTGGTGATCTCACACTC
*ZFP42*_forward	GGATTCCTTCTCTGACTGTTAC
*ZFP42*_reverse	GCTCTTGTTCTGATCCTTCTT
*DPPA3*_forward	CTCAGGCTTGTCCTCAAATG
*DPPA3*_reverse	CGTCAAGTTACTGAGGTTCTG
*PRDM14*_forward	GAGCCTGCAGGTCATAAAG
*PRDM14*_reverse	CTTGAGATGCTTGTCTCTGTAA
*TCL1A*_forward	GGCAAAGGCTGTGTATGT
*TCL1A*_reverse	CCTGACGCATGAGTACTTG
*SOX17*_forward	CTTCATGGTGTGGGCTAAGG
*SOX17*_reverse	CGGCCGGTACTTGTAGTTG

*GATA6*_forward	GCTGCACAGTCTACAGAGTC
*GATA6*_reverse	AGCGGTTGCACAAGTAGT
*GATA4*_forward	AAGAGATGCGTCCCATCAAG
*GATA4*_reverse	GACTGGCTGACCGAAGATG
*CDX2*_forward	AACCTGTGCGAGTGGATG
*CDX2*_reverse	CCTTTCTCCGAATGGTGATGTA
*TEAD4*_forward	TGTGAGTACATGGTCAACTTCAT
*TEAD4*_reverse	GCTGACACCTCGAAGACATAC
*GATA3*_forward	TACTACGGAAACTCGGTGAGG
*GATA3*_reverse	TGGATGGACGTCTTGGAGAA
*TET1*_forward	TGTCGGCTTGGCAAGAAAGA
*TET1*_reverse	AGACCACTGTGCTGCCATTA
*TET2*_forward	GTGAGATCACTCACCCATCGCATA
*TET2*_reverse	TACTGGCACTATCAGCATCACAGG
*TET3*_forward	TCTTCCGTCGTTCAGCTACTACAG
*TET3*_reverse	GTGGAGGTCTGGCTTCTTAAA

Table S6. Primers used for PCR amplification of bisulfite converted DNA.

Primers	Sequence (5’ → 3’)
NANOG promoter1_forward	AAAATTAGGTAGAGATATTATTAAAAA
NANOG promoter1_reverse	AAATATTCCCTCTATACCCACTTAAC
NANOG promoter2_forward	CTTATATAGGAAGAGAAGAGATTAAATTG
NANOG promoter2_reverse	CCCAACAATACTTACTAAATAAACTTTCC
NANOG gene-body1_forward	CTAATTTAATATGAGTGTGGA
NANOG gene-body1_reverse	GAATATTAAAAAATTCTTACATCTACTAAAA
NANOG gene-body2_forward	GAGAGGTAGAAGTATTTTAGTTTTTAGTAGA
NANOG gene-bopy2_reverse	GTAAAATAATTTAAAAATAAATCCATAATTT

Table S7. PCR conditions for amplification of bisulfite converted DNA

Amplified regions	Primers	PCR conditions
NANOG promoter part 1	*NANOG* promoter1_forward	Initial denaturation for 2 min at 95°C and 40 cycles of denaturation for 30 sec at 95°C, annealing for 30 sec at 50°C, extension for 30 sec min at 72°C, followed by a final extension for 2 min at 72°C.
	NANOG promoter1_reverse	
NANOG promoter part 2	*NANOG* promoter2_forward	Initial denaturation for 2 min at 95°C and 40 cycles of denaturation for 30 sec at 95°C, annealing for 30 sec at 50°C, extension for 45 sec min at 72°C, followed by a final extension for 2 min at 72°C.
	NANOG promoter2_reverse	
NANOG gene-body part 1	*NANOG* gene-body1_forward	Initial denaturation for 2 min at 95°C and 40 cycles of denaturation for 30 sec at 95°C, annealing for 30 sec at 45°C, extension for 30 sec min at 72°C, followed by a final extension for 2 min at 72°C.
	NANOG gene-body1_reverse	
NANOG gene-body part 2	*NANOG* gene-body2_forward	Initial denaturation for 2 min at 95°C and 40 cycles of denaturation for 30 sec at 95°C, annealing for 30 sec at 50°C, extension for 30 sec min at 72°C, followed by a final extension for 2 min at 72°C.
	NANOG gene-bopy2_reverse	