

Supplementary Figure 1

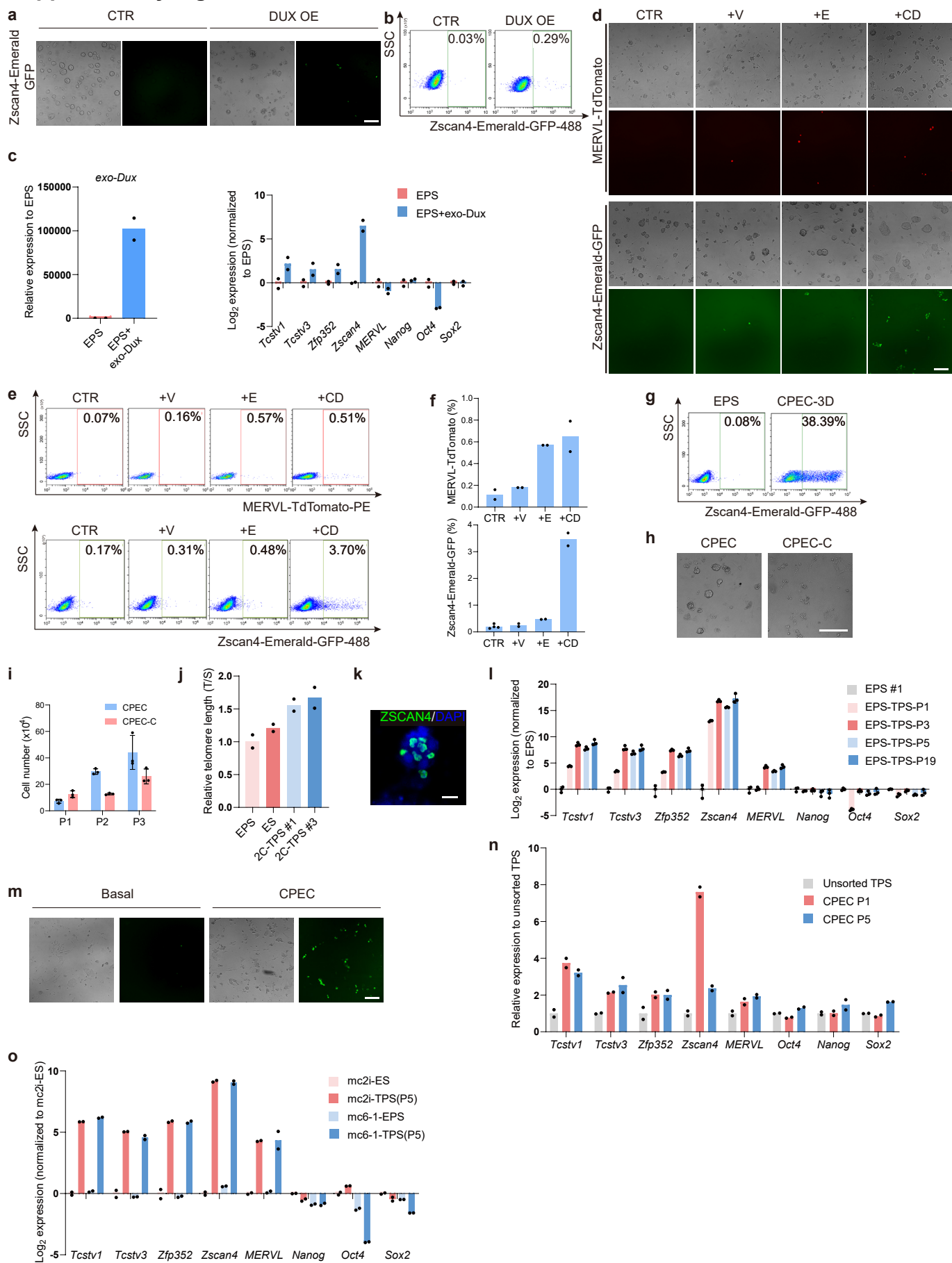


Figure S1. Identification of small molecules that can increase the percentage of cells expressing totipotency marker genes.

- a. Representative images showing the effect of exogenous *Dux* transient overexpression on activating Zscan4 reporter expression in EPS cells on day 3. Scale bar, 200 μ m. CTR, control. DUX OE, exogenous *Dux* overexpression.
- b. Representative flow cytometry analysis of the effect of exogenous *Dux* transient overexpression on activating Zscan4 reporter expression in EPS cells on day 3. The percentage of Zscan4-Emerald-GFP positive cells was shown. CTR, control. DUX OE, exogenous *Dux* overexpression.
- c. Q-PCR analysis of expression of exogenous *Dux* (left panel) and totipotency and pluripotency marker genes (right panel) upon exogenous transient overexpression of *Dux*. exo-*Dux*, exogenous *Dux* overexpression. N = 2 biological replicates.
- d. Representative images showing the effect of VPA, EPZ004777, and CD1530 on activating Zscan4 or MERVL reporter expression in EPS cells. Scale bar, 200 μ m. V, VPA. E, EPZ004777. CD, CD1530. CTR, N2B27 medium supplemented with CHIR 99021.
- e-g. Representative flow cytometry analysis of the percentage of Zscan4 or MERVL positive cells in EPS cells on day 3 after treatment of small molecules. For (f), N = 2 biological replicates. V, VPA. E, EPZ004777. CD, CD1530. CTR, N2B27 medium supplemented with CHIR 99021. EPS, EPS cells. CPEC-3D: EPS cells treated with 4 small molecules (VPA, CHIR 99021, EPZ004777, CD1530) for 3 days.
- h. Representative images showing the effect of CHIR 99021 on cell proliferation during inducing TPS cells. Scale bar, 200 μ m. CPEC-C, CPEC condition without CHIR 99021.
- i. Cell number analysis showing that CHIR 99021 promote cell proliferation during the conversion of TPS cells. N = 3 biological replicates. CPEC-C, CPEC condition without CHIR 99021.
- j. Q-PCR analysis of telomere length in TPS, ES and TPS cells. N = 2 biological replicates.
- k. Representative immunofluorescent analysis of ZSCAN4 expression in TPS cells. Scale bar, 200 μ m.
- l. Q-PCR analysis of the expression of totipotency and pluripotency marker genes during inducing TPS cells from EPS cells at different passages. EPS, EPS cells. EPS-TPS-P1/3/5/19: converted cells at passage 1/3/5/19. N = 3 biological replicates.
- m. Representative images showing sorted Zscan4-Emerald GFP positive cells cultured using the basal medium and CPEC-medium. Scale bar, 200 μ m. Basal, basal medium of CPEC condition.
- n. Q-PCR analysis of expression levels of representative totipotency and pluripotency marker genes in sorted Zscan4 positive cells cultured in CPEC-medium at passage 1 and 5 (CPEC-P1, CPEC-P5). N = 2 technical replicates. Similar results were obtained in at least 2 independent experiments.
- o. Q-PCR analysis of expression levels of representative totipotency and pluripotency marker genes in TPS cells converted from 2i/LIF-ES (mc2i-ES) and LCDM-EPS (mc6-1-EPS) respectively. N = 2 technical replicates. Similar results were obtained in at least 2 independent experiments.