CLAIMS

1. A method of resetting a human stem cell to a more naïve state, the method comprising:

(a) providing a human stem cell to be reset,

(b) inducing a more naïve state by

(i) optionally expressing or introducing one or more heterologous reprogramming factors into the cell,

(ii) culturing the cell in a resetting medium, wherein the resetting medium comprises a MEK inhibitor, and a tankyrase inhibitor and a STAT3 activator, and optionally one or more further inhibitors,

(c) sustaining the cell in a naïve culture medium, wherein the naïve culture medium comprises a MEK inhibitor, a PKC inhibitor and a GSK3 inhibitor, and a STAT3 activator.

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2. A method according to claim 1, wherein step (b)(i) is performed to reprogram the cell to a more naïve state and comprises expressing reprogramming factors in the cell, wherein the reprogramming factors comprise or consist of NANOG and KLF2.

3. A method according to claim 2 of reprogramming a human pluripotent stem cell to a more naïve state, the method comprising expressing reprogramming factors in the cell, wherein the reprogramming factors comprise NANOG and KLF2, and culturing the cell in the resetting medium, wherein the resetting medium comprises a MEK inhibitor, and optionally a GSK3 inhibitor and a STAT3 activator or optionally,
a FGF inhibitor and a STAT3 activator.

4. A method according to any one of claims 1 to 3, wherein expression of the reprogramming factors is transient, which is optionally achieved by introducing into the cell a plasmid preparation which expresses the reprogramming factors in the cell
and wherein heterologous nucleic acid encoding the reprogramming factors is not maintained following reprogramming to the naïve state.

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5. A method according to claim 1, wherein step (b)(i) is not performed and the resetting medium further comprises a HDAC inhibitor.

6. A method according to claim 1, wherein step (b)(i) is not performed and the resetting medium further comprises a PKC inhibitor.

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7. A method according to claim 1, wherein the resetting medium comprises a HDAC inhibitor.

8. A method according to any one of claims 1 to 7, wherein the resetting medium comprises a PKC inhibitor.

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9. A method according to any one of claims 1 to 8, wherein the resetting medium further comprises a GSK3 inhibitor.

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10. A method according to any one of claims 1 to 2, 5 to 6, or 8 to 9, wherein the naive culture medium further comprises a ROCK inhibitor.

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11. A method according to any one of claims 1 to 10 wherein the tankyrase inhibitor is XAV939.

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12. A method according to any one of claims 1 to 11,

(i) wherein the GSK3 inhibitor is CHIR99021; and/or,

(ii) wherein the MEK inhibitor is PD0325901; and/or,

(iii) wherein the PKC inhibitor is Gö6983 or Ro-31-8425; and/or,

(iv) wherein the STAT3 activator is LIF, which is optionally human LIF; and/or

(v) wherein the FGF inhibitor if present, is PD173074.

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13. A method according to any one of claims 1 to 12, wherein (i) the cells are cultured in an FGF supplemented serum replacement medium prior to inducing the more naïve state, and/or (ii) the resetting medium or naïve culture medium is replaced daily and\or wherein the resetting is performed in the presence of 5% oxygen.

s) self-renewal from dissociated single cells with or without Rho associated kinase inhibition (ROCKi);
t) self-renewal in the absence of serum or serum substitutes;
u) self-renewal in the absence of feeder cells;
v) self-renewal in the absence of transgene expression or other genetic perturbation;
w) retention of diploid karyotype without rearrangement in long-term passaging, for example over more than 40 population doublings;
x) differentiation into conventional primed pluripotent phenotype in the presence of growth factor stimulation of Erk/MAP kinase signalling and activin;
y) able to differentiate in vitro into primordial germ cells as well as somatic germ layers;
z) able to establish continuous culture in vitro by transition from a pre-implantation epiblast;
aa) tightly packed domed appearance;
bb) reduction in expression of DNMT3a and DNMT3b.

15. A method according to claim 14, wherein expression of KLF2, KLF4, TFCP2L1, TBX3, REX1, GBX2 and STELLA is induced in the reprogrammed cells.

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16. A method according to any one of the preceding claims wherein the human cells for use in the method are selected from:
(i) induced pluripotent stem cells;
(ii) cells from an embryonic cell line;
(iii) embryonic stem cells obtained by biopsy without destruction of the respective embryo.

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17. A resetting medium as defined in any one of claims 1 to 13, wherein the resetting medium comprises a MEK inhibitor, a tankyrase inhibitor, a PKC inhibitor, and a STAT3 activator, and optionally one or more further inhibitors.

18. A resetting medium according to claim 17, wherein the medium is serum-free.

19. A resetting composition according to claim 17 which comprises a GSK3 inhibitor.