CLAIMS

1. A method for producing a hematopoietic lineage cell, comprising the steps of:

(a) encapsulating a stem cell core in a hydrogel; and

(b) exposing the encapsulated stem cell core to a culture condition which promotes differentiation of the stem cell core into the hematopoietic lineage cell, such that the hematopoietic lineage cell is extruding or extruded from the hydrogel capsule;

in which the culture condition does not comprise any of a conditioned medium, serum, or a ROCK inhibitor; and

in which the stem cell core is a clump of stem cells or a spheroid.

2. A method according to Claim 1, further comprising the steps of:

(a) changing the culture condition to a different culture condition which further promotes differentiation of the stem cell into the hematopoietic lineage cell; and optionally

(b) repeating step (a) with further different culture conditions which further promote differentiation of the stem cell into the hematopoietic lineage cell.

3. A method according to Claim 1 or 2, in which the hematopoietic lineage cell is a hemogenic precursor cell, a multipotent hematopoietic stem cell, a multipotent hematopoietic progenitor cell, a common myeloid precursor, a common lymphoid precursor, a megakaryocyte, an erythrocyte, a T-lymphocyte, a B-lymphocyte, a natural killer cell, a monocyte, a macrophage, or a platelet.

4. A method according to Claim 1, 2 or 3, comprising an initial culture condition, and first, second, and third different culture conditions.

5. A method according to Claim 4, in which the initial culture condition comprises a medium comprising:

(a) a hematopoietic stem cell expansion medium; and

(b) 50ng/ml of each of BMP4, VEGF, and bFGF;

the first different culture condition comprises a medium comprising:

(a) a hematopoietic stem cell expansion medium;

(b) 10ng/ml of each of BMP4, VEGF, and bFGF;

(c) 284μM ascorbic acid; and

(d) 0.1mM βME;

the second different culture condition comprises a medium comprising:

(a) serum-free expansion medium (SFEM);

(b) 50ng/ml of each of Thrombopoietin (TPO), SCF, and Flt3;

(c) 20ng/ml of IL6;

(d) 10ng/ml of IL9;

(e) 5U/ml of heparin; and

(f) 10μM of Valproic acid;

and the third different culture condition comprises a medium comprising:

(a) SFEM;

(b) 50ng/ml of each of TPO and SCF;

(c) 10ng/ml of each of IL6 and IL9;

(d) 5μM of Arachidoic acid;

(e) 0.5μM of Eltrombopag (SB-497115-GR) – TPO receptor agonist (ETP); and

(f) 2.5mM nicotinamide.

6. A method according to Claim 4, in which the initial culture condition comprises a medium comprising:

(a) a hematopoietic stem cell expansion medium; and

(b) 50ng/ml of each of BMP4, VEGF, and bFGF;

the first different culture condition comprises a medium comprising:

(a) a hematopoietic stem cell expansion medium;

(b) 50ng/ml of each of BMP4 and VEGF;

(c) 20ng/ml of each of bFGF, TPO, and SCF; and

(d) 10μM Valproic acid;

the second different culture condition comprises a medium comprising:

(a) a hematopoietic stem cell expansion medium;

(b) 20ng/ml of SCF;

(c) 500nM of PPARg agonist L-165,041;

(d) 100nM of GPR40ag;

(e) 2mM of Metformin; and

(f) 5U/ml of heparin;

and the third different culture condition comprises a medium comprising:

(a) SFEM;

(b) 50ng/ml of each of TPO and SCF;

(c) 10ng/ml of each of IL6 and IL9;

(d) 5μM of Arachidoic acid;

(e) 0.5μM of ETP; and

(f) 2.5mM nicotinamide.

7. A method according to Claim 4, 5 or 6, in which the day on which the encapsulated stem cell is exposed to the initial culture condition is day 1, and the encapsulated stem cell is exposed to the first different culture condition on day 3 or day 4, to the second different culture condition on day 7, and to the third different culture condition on day 11.

8. A method according to any preceding claim, further comprising the step of separating one or more hematopoietic lineage cells from the encapsulated stem cells.

9. A method according to any preceding claim, in which at least one culture condition is feeder-free.

10. A method according to any preceding claim, in which the hydrogel comprises an alginate.

11. A method according to any preceding claim, in which the stem cell core is a spheroid.

12. A composition comprising:

(a) a hydrogel capsule comprising a stem cell; and

(b) a hematopoietic lineage cell;

in which the hematopoietic lineage cell is extruding or extruded from the hydrogel capsule.

13. A composition according to Claim 12, in which the hematopoietic lineage cell is a hemogenic precursor cell, a multipotent hematopoietic stem cell, a multipotent hematopoietic progenitor cell, a common myeloid precursor, a common lymphoid precursor, a megakaryocyte, an erythrocyte, a T-lymphocyte, a B-lymphocyte, a natural killer cell, a monocyte, a macrophage, or a platelet.

14. A vessel, preferably a bioreactor, containing a composition according to Claim 12 or 13.

15. Use of a vessel according to Claim 14 in a method for producing a hematopoietic lineage cell, in which the method is a method according to any of Claims 1 to 11.