only promote HE development, but also on methods to identify these progenitors as they are specified. In an earlier study, we used T cell potential to map the onset of definitive hematopoiesis in mouse ESC differentiation cultures, and demonstrated that this program initiates from a Flk-1$^+$ *Sox17*$^+$ progenitor that emerged 48 hours following the onset of primitive hematopoiesis (Irion et al., 2010). Several studies have demonstrated that it is possible to generate T lymphocytes from hESCs (Galic et al., 2006; Timmermans et al., 2009).

SUMMARY

There is described herein, use of T-cell potential to map the onset of the definitive hematopoietic program from hESCs and hiPSCs induced with specific morphogens in serum-free cultures.

In one aspect, the present disclosure refers to a method of enriching a population of stem cells for hematopoietic progenitors, the method comprising: a. inducing hematopoietic differentiation in a population of human embryonic stem cells or human induced pluripotent stem cells, wherein the inducing hematopoietic differentiation comprises: (i) culturing the population with BMP4, between days 0 and 4 of differentiation; (ii) culturing with bFGF, between days 1 and day 8 of differentiation; (iii) culturing with at least one of VEGF, DKK, IL-6 and IL-11, and/or combinations thereof, between days 3 and 13, or between days 4 and 9 of differentiation; (iv) culturing the population with at least one of SCF and EPO, between days 4 and 13, or between days 6 and 9 of differentiation; and (v) culturing the population with at least one of TPO, Flt-3 and IL-3, and/or combinations thereof, between days 6 and 13, or between days 8 and 9 of differentiation; b. sorting the population based on expression of CD43 and at least one of CD31 and CD144; and c. selecting a fraction that is CD31$^+$CD43$^-$, CD144$^+$CD43$^-$, or CD31$^+$CD43$^-$ and CD144$^+$CD43$^-$ wherein inducing hematopoietic differentiation is conducted serum free, inducing hematopoietic differentiation is conducted stroma free, the inducing hematopoietic differentiation comprises inhibiting activin/nodal signaling between days 2 and day 4 of differentiation; and the sorting is performed between day 6 and day 13 of differentiation.

In an example, a candidate definitive hematopoietic progenitor population was identified that emerges between days 6 and 9 of culture, expresses surface markers and genes indicative of HE/definitive hematopoietic progenitors, and displays T-cell potential following culture on OP9-DL4 stromal cells. In addition to T-cell progenitors, this population also gives rise to erythroid and myeloid progenitors following co-culture with

CLAIMS:

1. A method of enriching a population of stem cells for hematopoietic progenitors, the method comprising:

a. inducing hematopoietic differentiation in a population of human embryonic stem cells or human induced pluripotent stem cells, wherein the inducing hematopoietic differentiation comprises:

(i) culturing the population with BMP4, between days 0 and 4 of differentiation;

(ii) culturing with bFGF, between days 1 and day 8 of differentiation;

(iii) culturing with at least one of VEGF, DKK, IL-6 and IL-11, and/or combinations thereof, between days 3 and 13, or between days 4 and 9 of differentiation;

(iv) culturing the population with at least one of SCF and EPO, between days 4 and 13, or between days 6 and 9 of differentiation; and

(v) culturing the population with at least one of TPO, Flt-3 and IL-3, and/or combinations thereof, between days 6 and 13, or between days 8 and 9 of differentiation;

b. sorting the population based on expression of CD43 and at least one of CD31 and CD144; and

c. selecting a fraction that is $CD31^{+}CD43^{-}$, $CD144^{+}CD43^{-}$, or $CD31^{+}CD43^{-}$ and $CD144^{+}CD43^{-}$

wherein

inducing hematopoietic differentiation is conducted serum free,

inducing hematopoietic differentiation is conducted stroma free,

the inducing hematopoietic differentiation comprises inhibiting activin/nodal signaling between days 2 and 4 of differentiation; and

the sorting is performed between day 6 and day 13 of differentiation.

2. The method of claim 1, wherein inhibiting activin/nodal signaling comprises culturing the population with an activin/nodal inhibitor.

3. The method of claim 2, wherein the inhibitor is SB-431542.

4. The method of any one of claims 1-3, Wherein the sorting is performed at a time selected between days 7 and 10 of differentiation.

5. The method of any one of claims 1-4, wherein the population is sorted based on expression of CD43 and CD31 and the selected fraction is $CD31^{+}CD43^{-}$.

6. The method of any one of claims 1-4, wherein the population is sorted based on expression of CD43 and CD144 and the selected fraction is $CD144^{+}CD43^{-}$.

7. The method of any one of claims 1-6, wherein the fraction comprises at least 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90% or 100% hematopoietic progenitors.