(FBS) and/or feeder cells with non-defined components for the derivation of MSCs, which greatly compromise the clinical application of the MSCs derived. Current methods of the differentiation of iPSCs into MSCs are also time-consuming and inefficient. Thus, there is the need for an alternative method of deriving MSCs.

SUMMARY

[0003a] In one aspect, the present disclosure refers to a method of generating mature mesenchymal stem cells from lateral plate mesoderm cells, comprising culturing the lateral plate mesoderm cells on an extracellular matrix in a mesenchymal stem cell culture medium comprising: fibroblast growth factor; platelet-derived growth factor (PDGF); epidermal growth factor (EGF) family protein; and ascorbic acid, wherein the method further comprises, before culturing the lateral plate mesoderm cells: culturing primitive streak mesendoderm cells on an extracellular matrix to obtain lateral plate mesoderm cells, wherein culturing the primitive streak mesendoderm further comprises: (i) culturing the primitive streak mesendoderm on an extracellular matrix in a first lateral plate mesoderm cell culture medium; and (ii) subsequently culturing the primitive streak mesendoderm from (i) on an extracellular matrix in a second lateral plate mesoderm cell culture medium, wherein the first lateral plate mesoderm cell culture medium comprises: fibroblast growth factor; bone morphogenetic protein; Rho-associated protein kinase (ROCK) inhibitor; and Follistatin, and wherein the second lateral plate mesoderm cell culture medium comprises: fibroblast growth factor; bone morphogenetic protein; and follistatin.

[0003b] In another aspect, the present disclosure refers to a method of generating mature mesenchymal stem cells from lateral plate mesoderm cells, comprising culturing the lateral plate mesoderm cells on an extracellular matrix in a mesenchymal stem cell culture medium comprising: fibroblast growth factor; platelet-derived growth factor (PDGF); epidermal growth factor (EGF) family protein; and ascorbic acid, wherein the method further comprises, before culturing the lateral plate mesoderm cells: culturing primitive streak mesendoderm cells on an extracellular matrix to obtain lateral plate mesoderm cells; and before culturing the primitive streak mesendoderm cells: culturing pluripotent stem cells on an extracellular matrix to obtain primitive streak mesendoderm cells, wherein culturing the pluripotent stem cells on an extracellular matrix further comprises: (i) culturing the pluripotent stem cells on an extracellular matrix in a first primitive streak mesendoderm cell culture medium; and (ii)

subsequently culturing the pluripotent stem cells from (i) on an extracellular matrix in a second primitive streak mesendoderm cell culture medium, wherein the first primitive streak mesendoderm cell culture medium comprises: activin; and WNT-signaling activator; and wherein the second primitive streak mesendoderm cell culture medium comprises: activin; WNT-signaling activator; and fibroblast growth factor.

[0004] In a first example, there is provided a method of generating mature mesenchymal stem cells from lateral plate mesoderm cells, comprising culturing the lateral plate mesoderm cells on an extracellular matrix in a mesenchymal stem cell culture medium comprising: (i) fibroblast growth factor; (ii) platelet-derived growth factor (PDGF); (iii) epidermal growth factor (EGF) family protein; and (iv) ascorbic acid.

[0005] In a second example, there is provided a mesenchymal stem cell culture obtained by the method of the present disclosure.

[0006] In a third example, there is provided a method of treating cartilage or bone disease or injury in a patient, comprising transplanting a composition of cells comprising mature mesenchymal stem cells into the patient with cartilage defects, wherein the composition is obtained by the method of the present disclosure.

[0007] In a fourth example, there is provided a culture medium for deriving primitive streak mesendoderm cells from pluripotent stem cells, comprising: (a) activin; (b) WNT-signaling activator; and (c) fibroblast growth factor.

[0008] In a fifth example, there is provided a culture medium for deriving lateral plate mesoderm cells from primitive streak mesendoderm cells, comprising: (1) fibroblast growth factor; (2) bone morphogenetic protein; (3) follistatin; and (4) optionally Rho-associated protein kinase (ROCK) inhibitor.

[0009] In a sixth example, there is provided a culture medium for deriving and maintaining mature mesenchymal stem cells from lateral plate mesoderm cells, comprising: (i) fibroblast growth factor; (ii) platelet-derived growth factor (PDGF); (iii) epidermal growth factor (EGF) family protein; and (iv) ascorbic acid.

CLAIMS

1. A method of generating mature mesenchymal stem cells from lateral plate mesoderm cells, comprising culturing the lateral plate mesoderm cells on an extracellular matrix in a mesenchymal stem cell culture medium comprising:

fibroblast growth factor;

platelet-derived growth factor (PDGF);

epidermal growth factor (EGF) family protein; and

ascorbic acid,

wherein the method further comprises, before culturing the lateral plate mesoderm cells:

culturing primitive streak mesendoderm cells on an extracellular matrix to obtain lateral plate mesoderm cells, wherein culturing the primitive streak mesendoderm further comprises:

(i) culturing the primitive streak mesendoderm on an extracellular matrix in a first lateral plate mesoderm cell culture medium; and

(ii) subsequently culturing the primitive streak mesendoderm from (i) on an extracellular matrix in a second lateral plate mesoderm cell culture medium,

wherein the first lateral plate mesoderm cell culture medium comprises:

fibroblast growth factor;

bone morphogenetic protein;

Rho-associated protein kinase (ROCK) inhibitor; and

Follistatin,

and wherein the second lateral plate mesoderm cell culture medium comprises:

fibroblast growth factor;

bone morphogenetic protein; and

follistatin.

2. The method of claim 1, wherein the primitive streak mesendoderm cells to be cultured in the first lateral plate mesoderm cell culture medium are characterized by any one or more or all of the following markers: MIXL1, T, GSC and Wnt3.

3. The method of claim 1 or 2, wherein the fibroblast growth factor is FGF2.

4. The method of any one of claims 1 to 3, wherein the bone morphogenetic protein is BMP4.

5. The method of any one of claims 1 to 4, wherein the ROCK inhibitor is Y27632 (R)-(+)-trans-4-(1-Aminoethyl)-N-(4-Pyridyl)cyclohexanecarboxamide dihydrochloride.

6. A method of generating mature mesenchymal stem cells from lateral plate mesoderm cells, comprising culturing the lateral plate mesoderm cells on an extracellular matrix in a mesenchymal stem cell culture medium comprising:,

fibroblast growth factor;

platelet-derived growth factor (PDGF);

epidermal growth factor (EGF) family protein; and

ascorbic acid,

wherein the method further comprises, before culturing the lateral plate mesoderm cells: culturing primitive streak mesendoderm cells on an extracellular matrix to obtain lateral plate mesoderm cells; and

before culturing the primitive streak mesendoderm cells: culturing pluripotent stem cells on an extracellular matrix to obtain primitive streak mesendoderm cells, wherein culturing the pluripotent stem cells on an extracellular matrix further comprises:

(i) culturing the pluripotent stem cells on an extracellular matrix in a first primitive streak mesendoderm cell culture medium; and

(ii) subsequently culturing the pluripotent stem cells from (i) on an extracellular matrix in a second primitive streak mesendoderm cell culture medium,

wherein the first primitive streak mesendoderm cell culture medium comprises:

activin; and

WNT-signaling activator;

and wherein the second primitive streak mesendoderm cell culture medium comprises:

activin;

WNT-signaling activator; and

fibroblast growth factor.

7. The method of claim 6, wherein the activin is Activin A.

8. The method of claim 6 or 7, wherein the WNT-signaling activator is CHIR-99021 6-[2-[[4-(2,4-dichlorophenyl)-5-(5-methyl-1H-imidazol-2-yl)pyrimidin-2-yl]amino]ethylamino]pyridine-3-carbonitrile.

9. The method of any one of claims 6 to 8, wherein the fibroblast growth factor is FGF2.