Therefore, there remains a need in the art for cell cultivation methods that provide one or more of the following: mimics internal environment, eliminates the need for sub-culturing of cells, enables continuous cell growth while optionally simultaneously harvesting grown cells, and enables simultaneous multi-cell cultivation where interfaces between different cells of interest may be produced for further use and/or analysis.

SUMMARY

One or more examples of the disclosure may address one or more of the aforementioned problems. Certain examples according to the disclosure provide a method of cultivating cells. In one aspect, the present disclosure refers to a method of cultivating cells, the method may comprise the following steps: (i) providing an initial scaffolding comprising a first interlocking porous hydrogel block (IPHB), wherein the first IPHB comprises a three-dimensional (3D) macrostructure defined by a continuous polymeric matrix material and a network of microporous channels and/or chambers extending throughout the continuous polymeric matrix material, and wherein the 3D macrostructure comprises a top surface, a bottom surface, and a thickness defined by at least one side edge extending from the top surface to the bottom surface, and wherein the 3D macrostructure structure includes at least one interlocking-male component and at least one interlocking-female component; (ii) seeding the first IPHB with one or more cells of interest; (iii) feeding the one or more cells of interest with a first culture media, and allowing the one or more cells of interest to propagate throughout the network of microporous channels and/or chambers; (iv) expanding the initial scaffolding by interlocking a second IPHB to the first IPHB, wherein the at least one interlocking-male component or at least one interlocking-female component of the first IPHB is joined to a corresponding interlocking-male component or corresponding interlocking-female component of the second IPHB; and (v) allowing the one or more cells of interest to propagate from the first IPHB into the second IPHB, and feeding the one or more cells of interest located inside the second IPHB with the first culture media or a second culture media.

BRIEF DESCRIPTION OF THE DRAWING(S)

The invention now will be described more fully hereinafter with reference to the accompanying drawings, in which some, but not all embodiments of the invention are shown. Indeed, this invention may be embodied in many different forms and should not be construed as limited to the embodiments set forth herein; rather, these embodiments are provided so that

THAT WHICH IS CLAIMED:

1. A method of cultivating cells, comprising:

(i) providing an initial scaffolding comprising a first interlocking porous hydrogel block (IPHB), wherein the first IPHB comprises a three-dimensional (3D) macrostructure defined by a continuous polymeric matrix material and a network of microporous channels and/or chambers extending throughout the continuous polymeric matrix material, and wherein the 3D macrostructure comprises a top surface, a bottom surface, and a thickness defined by at least one side edge extending from the top surface to the bottom surface, and wherein the 3D macrostructure structure includes at least one interlocking-male component and at least one interlocking-female component;

(ii) seeding the first IPHB with one or more cells of interest;

(iii) feeding the one or more cells of interest with a first culture media, and allowing the one or more cells of interest to propagate throughout the network of microporous channels and/or chambers;

(iv) expanding the initial scaffolding by interlocking a second IPHB to the first IPHB, wherein the at least one interlocking-male component or at least one interlocking-female component of the first IPHB is joined to a corresponding interlocking-male component or corresponding interlocking-female component of the second IPHB; and

(v) allowing the one or more cells of interest to propagate from the first IPHB into the second IPHB, and feeding the one or more cells of interest located inside the second IPHB with the first culture media or a second culture media.

2. The method of claim 1, wherein the one or more cells of interest grow and propagate through the network of microporous channels and/or chambers and define a winding pattern wherein the one or more cells of interest wind back-and-forth in a 3D configuration across at least a portion of individual microporous channels as the one or more cells of interest

propagate from an interior portion of the first IPHB towards one or more exterior surfaces, such as the top surface, the bottom surface, and/or or the at least one side edge.

3. The method of claim 1, wherein the one or more cells of interest grow and propagate through the network of microporous channels and/or chambers and define a spiral pattern adhered to walls of the network of microporous channels and/or chambers as the one or more cells of interest propagate from an interior portion of the first IPHB towards one or more exterior surfaces, such as the top surface, the bottom surface, and/or or the at least one side edge.

4. The method of claim 1, wherein the one or more cells of interest grow and propagate through the network of microporous channels and/or chambers and define a network of nodes and branches defining open regions therebetween as the one or more cells of interest propagate from an interior portion of the first IPHB towards one or more exterior surfaces, such as the top surface, the bottom surface, and/or or the at least one side edge.

5. The method of claim 1, wherein the one or more cells of interest grow and propagate through the network of microporous channels and/or chambers and define a continuous cell sheet adhered to walls of the network of microporous channels and/or chambers as the one or more cells of interest propagate from an interior portion of the first IPHB towards one or more exterior surfaces, such as the top surface, the bottom surface, and/or or the at least one side edge.

6. The method of claim 1, wherein the one or more cells of interest grow and propagate through the network of microporous channels and/or chambers and define a spheroid.

7. The method of claim 1, wherein the one or more cells of interest grow and propagate through the network of microporous channels and/or chambers and define a comet-like

structure having a spheroid structure and a spiral structure emanating from the spheroid structure as the one or more cells of interest propagate from an interior portion of the first IPHB towards one or more exterior surfaces, such as the top surface, the bottom surface, and/or or the at least one side edge.

8. The method according to any one of claims 1-7, further comprising a step of harvesting at least a portion of the one or more cells located throughout the network of microporous channels and/or chambers of the first IPHB.

9. The method of claim 8, wherein the step of harvesting at least a portion of the one or more cells located throughout the network of microporous channels and/or chambers of the first IPHB comprises (i) flushing them out of the first IPHB with a fluid medium, or (ii) degrading the 3D macrostructure of the first IPHB.

10. The method according to any one of claims 1-9, wherein the method comprises a chain-cultivation method comprising the sequential addition of a plurality of secondary blocks to the first IPHB, wherein the plurality of secondary blocks includes the second IPHB and a third IPHB interconnected directly to the second IPHB such that the second IPHB is located directly between the first IPHB and the third IPHB, and wherein the plurality of secondary IPHBs are initially devoid of cells.

11. The method of claim 10, wherein the cells of interest located in the first IPHB are harvested after cell propagation from the first IPHB to the second IPHB, and the cells of interest located in the second IPHB are harvested after cell propagation from the second IPHB to the third IPHB.

12. The method according to any one of claims 1-11, wherein the method comprises a multi-cell cultivation method, wherein the one or more cells of interest seeded in the first

IPHB comprises a first cell type and the second IPHB is seeded with a second cell type, wherein the first cell type is different than the second cell type.

13. The method of claim 12, wherein cells of the first cell type and cells of the second cell type are allowed to propagate towards each other and form a first interface between the first cell type and the second cell type.

14. The method of claim 13, further comprising a step of degrading each of the IPHBs to expose each cell type and the first interface.

15. The method of claim 14, further comprising interlocking a third IPHB directly together with the second IPHB, wherein the second IPHB is located directly between the first IPHB and the third IPHB, and seeding the third IPHB with a third cell type that is different from the first cell type and the second cell type.

16. The method of claim 15, wherein the cells of the second cell type and cells of the third cell type are allowed to propagate towards each other and form a second interface between the second cell type and the third cell type.

17. The method of claim 16, further comprising a step of degrading each of the IPHBs to expose each cell type, the first interface, and the second interface.

18. The method according to any one of claims 1-17, wherein the one or more cells of interest produce or secrete a therapeutic of interest, such as a biologic.

19. The method of claim 18, wherein the therapeutic comprises exosomes, extracellular vesicles, growth factors, monoclonal antibodies, peptides, proteins, viral particles, oligonucleotides, organelles, or combinations thereof.

20. The method according to any one of the preceding claims, further comprising a step of coating an interface between the continuous polymeric matrix material and the network of microporous channels and/or chambers extending throughout the continuous polymeric matrix material with a compatibilizer, wherein the compatibilizer is selected to promote adhesion of a primary cell of interest to the IPHB.

21. The method of claim 1, wherein the cultivation method enhances the secretory activity of Mesenchymal Stem Cells (MSCs), improving their regenerative potential for therapeutic applications.