SCALABLE BIOREACTOR SYSTEMS AND METHODS FOR TISSUE ENGINEERING

CROSS-REFERENCE

[0001] This application claims the benefit of U.S. Provisional Application No. 62/879,773, filed July 29th, 2019, and U.S. Provisional Application No. 62/991,958, filed March 24th, 2020, both of which are incorporated herein by reference.

BACKGROUND

[0002] Current tissue engineering bioreactors were neither intended for nor directly applicable in large scale commercial manufacturing. A scalable bioreactor system as described herein can address the need for large scale commercial manufacturing.

SUMMARY OF THE DISCLOSURE

[0003] Disclosed herein are devices. In ~~some embodiments~~a first aspect, ~~disclosed~~ the present disclosure provides~~herein is~~ a tissue culturing device comprising a plurality of trays substantially within a device, wherein each tray can be configured to hold at least one cell culturing container, wherein trays can be stacked within a device, and wherein a device can be configured to repeatedly tilt such that an angle between a bottom of a stack and a base of a device can repeatedly cycle between about 0 and about 360 degrees. In some embodiments, at least one tray can comprise at least one cell culture container that contains a liquid medium and cells. In some embodiments, at least one tray can be approximately rectangular on a side. In some embodiments, a device can comprise a pivot and a lifting mechanism configured to tilt stacked trays by angular raising and lowering. In some embodiments, an angular raising and lowering can create a wave in a wave in a cell culture medium within said cell culturing container. In some embodiments, a lifting mechanism can comprise a hydraulic mechanism, an electric mechanism, a spring mechanism, a piston, or a combination thereof. In some embodiments, a device can be jacketed. In some embodiments, a device can be temperature controlled. In some embodiments, a base of a device can comprise a pallet. In some embodiments, a device can be stacked on a pallet rack. In some embodiments, a pallet can be detachable. In some embodiments, a pallet can be non-detachable. In some embodiments, a pallet can comprise a plastic, a metal, or a combination thereof. In some embodiments, a pallet can comprise flats, grids, has an elevated foundation such that air can move underneath, or any combination thereof. In some embodiments, a base of a device can comprise means for moving said device using a forklift truck. In some embodiments, a length of a cell culturing container ranges from about 1 cm to about 1000 cm. In some embodiments, a width of a cell culturing container ranges from about 1 cm to about 1000 cm.

In some embodiments, a height of a cell culturing container ranges from about 1 cm to about 1000 cm. In some embodiments, a wall thickness of a cell culturing container ranges from about 0.01 cm to about 10 cm. In some embodiments, at least one tray can comprise a handle. In some embodiments, at least one tray can slide in and out of a device. In some embodiments, a device can comprise a monitoring system. In some embodiments, a monitoring system can alert a user that an action must be taken. In some embodiments, a monitoring system comprises, a sensor, a camera, or a combination thereof. In some embodiments, a sensor can comprise a thermistor, a thermometer, a pH sensor, a humidity sensor, a pressure sensor, a smoke detector, or any combination thereof. In some embodiments, a device can be jacketed. In ~~some embodiments~~a second aspect, the present disclosure provides a system ~~can~~ comprise~~e~~ing the device according to the first aspect and a cell culture medium reservoir. In some embodiments, a cell culture medium reservoir can comprise a monitoring system. In some embodiments, a monitoring system can alert a user that an action must be taken. In some embodiments, a monitoring system comprises, a sensor, a camera, or a combination thereof. In some embodiments, a sensor can comprise a thermistor, a thermometer, a pH sensor, a humidity sensor, a pressure sensor, a smoke detector, or any combination thereof. In some embodiments, a cell culture medium reservoir can be jacketed. In some embodiments, a cell culture medium reservoir can be temperature controlled. In some embodiments, a cell culture medium reservoir can be maintained at a lower temperature than a device. In some embodiments, a cell culture medium can be warmed before entering a device. In some embodiments, a gas can be bubbled into a cell culture medium. In some embodiments, a gas can comprise carbon dioxide, nitrogen, oxygen, or a combination thereof. In some embodiments, a cell culture medium reservoir can be connected to an inlet in a device. In some embodiments, an inlet can comprise an inlet manifold. In some embodiments, a device can comprise an outlet for waste cell culture medium. In some embodiments, an outlet can comprise an outlet manifold. In some embodiments, a system can comprise racks. In some embodiments, racks can be stackable vertically. In some embodiments, racks can be from about 1m high to about 200m high. In some embodiments, racks can be arranged from approximately floor to ceiling in a warehouse. In some embodiments, a system can comprise a pallet retrieval system. In some embodiments, a system can be configured to fit on a North American 40” x 48” pallet. In some embodiments, a system can be configured to fit on a conveyer system. In some embodiments, at least one container can comprise a tube operatively connected thereto. In some embodiments, a system can further comprise a temperature control system. A temperature control system can be configured to heat or cool at least a portion of a system to a temperature of from about -200 degrees Celsius to about 200 degrees Celsius. In some embodiments, a system can further comprise an intermediate bulk container heating jacket operatively coupled to a temperature control system. An intermediate bulk container heating jacket can be configured to heat a portion of a system. In some embodiments, a system can further comprise an intermediate bulk container cooling jacket

CLAIMS

WHAT IS CLAIMED IS:

1. A tissue culturing device comprising a plurality of trays substantially within the device, wherein each tray is configured to hold at least one cell culturing container, wherein the trays are stacked within the device, and wherein the device is configured to repeatedly tilt such that the angle between the bottom of the stack and the base of the device can repeatedly cycle between about 0 and about 360 degrees.
2. The device of claim 1, wherein the plurality of trays comprises at least one cell culture container that contains a liquid medium and cells.
3. The device of claim 1, wherein the plurality of trays is approximately rectangular on a side.
4. The device of claim 1, wherein the device comprises a pivot and a lifting mechanism configured to tilt the stacked trays by angular raising and lowering.
5. The device of claim 4, wherein the angular raising and lowering creates a wave in a wave in a cell culture medium within said cell culturing container.
6. The device of claim 1, wherein the lifting mechanism comprises a hydraulic mechanism, an electric mechanism, a spring mechanism, a piston, or a combination thereof.
7. The device of claim 1, wherein the device is jacketed.
8. The device of claim 1, wherein the device is temperature controlled.
9. The device of claim 1, wherein the base of the device can comprise a pallet.
10. The device of claim 9, wherein the device can be stacked on a pallet rack.
11. The device of claim 9, wherein the pallet is detachable.
12. The device of claim 9, wherein the pallet is not detachable.
13. The device of claim 9, wherein the pallet comprises a plastic, a metal, or a combination thereof.
14. The device of claim 9, wherein the pallet comprises flats, grids, has an elevated foundation such that air can move underneath, or any combination thereof.
15. The device of claim 1, wherein the base of the device can comprise means for moving said device using a forklift truck.
16. The device of claim 1, wherein a length of the trays is from about 1 cm to about 1000 cm.
17. The device of claim 1, wherein a width of the trays is from about 1 cm to about 1000 cm.
18. The device of claim 1, wherein a height of the trays is from about 1 cm to about 1000 cm.
19. The device of claim 1, wherein a wall thickness of the trays is from about 0.01 cm to about 10 cm.
20. The device of claim 1, wherein a length of the cell culturing container ranges from about 1 cm to about 1000 cm.

21. The device of claim 1, wherein a width of the cell culturing container ranges from about 1 cm to about 1000 cm.

22. The device of claim 1, wherein a height of the cell culturing container ranges from about 1 cm to about 1000 cm.

23. The device of claim 1, wherein a wall thickness of the cell culturing container ranges from about 0.01 cm to about 10 cm.

24. The device of claim 1, wherein at least one of the trays comprises a handle.

25. The device of claim 1, wherein at least one of the trays can slide in and out of the device.

26. The device of claim 1, comprising a monitoring system.

27. The device of claim 26, wherein the monitoring system can alert a user that an action must be taken.

28. The device of claim 26, wherein the monitoring system comprises, a sensor, a camera, or a combination thereof.

29. The device of claim 26, wherein the sensor comprises a thermistor, a thermometer, a pH sensor, a humidity sensor, a pressure sensor, a smoke detector, or any combination thereof.

30. The device of claim 1, wherein the device is jacketed.

31. A system comprising the device of claim 1 and a cell culture medium reservoir.

32. The system of claim 31, wherein the cell culture medium reservoir comprises a monitoring system.

33. The system of claim 32, wherein the monitoring system can alert a user that an action must be taken.

34. The system of claim 32, wherein the monitoring system comprises, a sensor, a camera, or a combination thereof.

35. The system of claim 34, wherein the sensor comprises a thermistor, a thermometer, a pH sensor, a humidity sensor, a pressure sensor, a gas sensor, a CO_2 sensor, an O_2 a smoke detector, or any combination thereof.

36. The system of claim 31, wherein the cell culture medium reservoir is jacketed.

37. The system of claim 31, wherein the cell culture medium reservoir is temperature controlled.

38. The system of claim 31, wherein the cell culture medium reservoir is maintained at a lower temperature than the device.

39. The system of claim 38, wherein the cell culture medium is warmed before entering the device.

40. The system of claim 31, wherein a gas is bubbled into the cell culture medium.

41. The system of claim 40, wherein the gas comprises carbon dioxide, nitrogen, oxygen, or a combination thereof.

42. The system of claim 31, wherein the cell culture medium reservoir is connected to an inlet in the device.

43. The system of claim 42, wherein the inlet comprises an inlet manifold.

44. The system of claim 31, wherein the device comprises an outlet for waste cell culture medium.

45. The system of claim 44, wherein the outlet comprises an outlet manifold.

46. The system of claim 31, wherein the system comprises racks.

47. The system of claim 46, wherein the racks are stackable vertically.

48. The system of claim 47, wherein the racks are from about 1m high to about 200m high.

49. The system of claim 48, wherein the racks can be arranged from approximately floor to ceiling in a warehouse.

50. The system of claim 31, wherein the system comprises a pallet retrieval system.

~~**51.** A system for growing an animal cell culture, comprising:~~

~~(a) at least two containers, wherein the at least two containers are configured for stacking, and wherein the animal cell culture directly contacts a portion of a surface of the at least two containers; and~~

~~(b) a motion control system that, when activated, is configured to rock or tilt the at least two containers at an angle of greater than 0 degrees to about 360 degrees, and wherein the at least two stackable containers are configured to reversibly attach to the motion control system.~~

~~**52.** The system of claim 47, wherein a length of the at least two containers ranges from about 1 cm to about 1000 cm.~~

~~**53.** The system of claim 47, wherein the width of the at least two containers ranges from about 1 cm to about 1000 cm.~~

~~**54.** The system of claim 47, wherein the height of the at least two containers ranges from about 1 cm to about 1000 cm.~~

~~**55.** The system of claim 47, wherein the wall thickness of the at least two containers ranges from about 0.01 cm to about 10 cm.~~

~~**56.** The system of any one of claims 1-51, wherein at least one of the at least two containers comprise a port operatively coupled to at least one of the at least two containers, wherein the port comprises a diameter of from about 0.1 cm to about 1000 cm.~~

~~**57.** The system of any one of claims 1-52, wherein at least one of the at least two containers comprise a lid.~~

~~**58.** The system of claim 53, wherein the lid is gas permeable.~~

~~**59.** The system of claim 53, wherein the lid is gas impermeable.~~

60. The system of any one of claims 1-55, further comprising a rack comprising a frame and a shelf; wherein the rack is configured to reversibly attach to the motion control system, and wherein at least one of the at least two containers are configured to reversibly attach to the shelf.

61. The system of claim 56, wherein the rack, the frame, or the shelf are independently constructed from a material selected from the group consisting of: polypropylene, polypropylene co-polymers, polyethylene, polyester, polystyrene, polycarbonate, polysulfone, polyolefin, polyetherimide, fluorinated ethylene propylene, polyphenylsulfone, polyetheretherketone, perfluoroalkoxy, ethylene tetrafluoroethylene, ethylene chlorotrifluoroethylene, polychlorotrifluoroethylene, polytetrafluoroethylene, polyphenyl sulfide, silicone, silicone co-polymers, stainless steel, steel alloys, aluminum, aluminum alloys, titanium, titanium alloys, glass, ceramic, and any combination thereof.

62. The system of any one of claims 1-57, wherein the system is configured to fit on a North American 40" x 48" pallet.

63. The system of any one of claims 1-58, wherein the system is configured to fit on a conveyer system.

64. The system of any one of claims 1-59, wherein at least one of the at least two containers comprise a tube operatively connected thereto.

65. The system of any one of claims 1-60, further comprising a temperature control system configured to heat or cool at least a portion of the system to a temperature of from about -200 degrees Celsius to about 200 degrees Celsius.

66. The system of claim 61, further comprising an intermediate bulk container heating jacket operatively coupled to the temperature control system, wherein the intermediate bulk container heating jacket is configured to heat the portion of the system.

67. The system of claim 61 or 62, further comprising an intermediate bulk container cooling jacket operatively coupled to the temperature control system, wherein the intermediate bulk container cooling jacket is configured to cool the portion of the system.

68. The system of any one of claims 1-63, further comprising a cell growth substrate.

69. The system of claim 64, wherein the cell growth substrate comprises a synthetic polymer, a natural polymer, a plant-derived material, a microbial-derived material, an animal-derived material, a metal, a ceramic, a glass, a mineral, a rock, a gem, a clay, or any combination thereof.

70. The system of claim 64, wherein the cell growth substrate comprises polystyrene, polyester, polyethylene terephlatate, poly(glycolic acid), poly(lactic acid), poly(lactic-co-glycolic acid), poly(ethylene glycol), polyurethane, poly(glycerol sebacate), polycarbonate, polyetherimide, stainless steel, silver, gold, platinum, palladium, iridium, titanium, tantalum, noble metal,

collagen, fibrin, silk, wool, wood, hemp, linin, cotton, cellulose, grass, leaves, straw, lignin, diamond, sapphire, amethyst, ruby, emerald, opal, topaz, quartz, marble, slate, lava rock, coral, sponge, diatom, sand dollar, sea shell, starfish, seaweed, kelp, algae, or any combination thereof.

71. The system of any one of claims 64-66, wherein the cell growth substrate is positioned inside at least one of the at least two containers.

72. The system of claim 67, wherein the cell growth substrate is positioned using a magnet or a fastener.

73. The system of any one of claims 1-68, wherein the at least two containers are independently rectangular, cylindrical, spherical, or triangular.

74. The system of any one of claims 1-69, wherein the at least two containers are independently constructed from a material that is substantially rigid, semi-rigid, or flexible.

75. The system of any one of claims 1-70, wherein the motion control system is an intermediate bulk container tilter.

76. The system of any one of claims 1-71, wherein at least one of the at least two containers is an intermediate bulk container.

77. The system of claim 72, wherein the intermediate bulk container comprises a volume of at least 275 gallons.

78. The system of any one of claims 1-73, wherein at least one of the at least two containers is a single-use bioprocessing container.

79. The system of any one of claims 1-73, wherein at least one of the at least two containers is a storage tank.

80. The system of any one of claims 1-73, wherein at least one of the at least two containers is a pressure vessel.

81. The system of any one of claims 1-73, wherein at least one of the at least two containers is a drum.

82. The system of any one of claims 1-77, wherein the system is configured to be heated in an incubator.

83. The system of any one of claims 1-78, wherein the system is configured to be heated in a heated space.

84. The system of any one of claims 1-79, wherein the system is configured to be cooled using refrigeration or freezing technology.

85. The system of any one of claims 1-80, wherein at least a portion of at least one of the at least two containers is gas permeable.

86. The system of any one of claims 1-80, wherein at least a portion of at least one of the at least two containers is gas impermeable.

87. The system of any one of claims 1-82, wherein at least a portion of an interior of at least one of the at least two containers is shaped or textured to influence fluid motion.

88. The system of any one of claims 1-83, wherein at least a portion of an interior of at least one of the at least two containers is shaped or textured to influence cell or tissue attachment.

89. The system of any one of claims 1-84, wherein at least one of the at least two containers is sterile.

90. The system of any one of claims 1-84, wherein at least one of the at least two containers is non-sterile.

91. A method of growing an animal cell culture comprising culturing the cell culture in the system of any one of claims 1-86.

92. The method of claim 87, wherein the culturing produces a cell-based tissue.

93. The method of claim 88, wherein the cell-based tissue is tanned, retanned, dyed, or fatliquored.

94. The method of claim 87, wherein the animal cell culture comprises a transgene, a heterologous RNA, or an epigenetically-modified base.

95. A cell-based tissue produced by the method of any one of claims 87-89.

96. The cell-based tissue of claim 91, wherein the cell-based tissue is treated for the purpose of making leather.

97. A composition that comprises:

(a) an acrylated chitosan; and

(b) an acrylate polymer, wherein the acrylate polymer comprises a phosphine oxide or a salt thereof.

98. The composition of claim 93, wherein the acrylated chitosan is a methacrylated chitosan or salt thereof.

99. The composition of claim 93, wherein the acrylated polymer comprises polyethylene glycol.

100. The composition of claim 95, wherein the acrylated polymer is poly(ethylene glycol) dimethacrylate or a salt thereof.

101. The composition of claim 93, wherein the phosphine oxide or salt thereof is 2,4,6-trimethylbenzoyl-diphenylphosphine oxide or a salt thereof.

102. The composition of claim 97, wherein the phosphine oxide comprises the salt of the 2,4,6-trimethylbenzoyl-diphenylphosphine oxide.

103. The composition of claim 98, wherein the salt is a lithium salt.

104. The composition of claim 98, wherein the salt is a sodium salt.

105. The composition of claim 93, that further comprises an organic dye or a salt thereof.

106. The composition of claim 101, wherein the organic dye or salt thereof is Martius yellow or a salt thereof.

107. The composition of claim 102 that comprises the salt of Martius yellow, wherein the salt is sodium.

108. A method comprising contacting an acrylated chitosan with an acrylate polymer, wherein the acrylate polymer comprises a phosphine oxide or a salt thereof.

109. The method of claim 104, wherein the acrylated chitosan is a methacrylated chitosan or salt thereof.

110. The method of claim 104, wherein the acrylated polymer comprises polyethylene glycol.

111. The method of claim 106, wherein the acrylated polymer is poly(ethylene glycol) dimethacrylate or a salt thereof.

112. The method of claim 104, wherein the phosphine oxide or salt thereof is 2,4,6-trimethylbenzoyl-diphenylphosphine oxide or a salt thereof.

113. The method of claim 108, wherein the phosphine oxide is the salt of the 2,4,6-trimethylbenzoyl-diphenylphosphine oxide.

114. The method of claim 109, wherein the salt is a lithium salt.

115. The method of claim 109, wherein the salt is a sodium salt.

116. The method of claim 104, further comprising contacting the acrylated chitosan and acrylate polymer with an organic dye or a salt thereof.

117. The method of claim 112, wherein the organic dye or salt thereof is Martius yellow or a salt thereof.

118. The method of claim 113, wherein the organic dye is the salt of Martius yellow, wherein the salt is sodium.

119. The method of claim 104, wherein the acrylated chitosan is dissolved in an organic acid prior to contacting with the acrylate polymer.

120. The method of claim 104, further comprising printing a scaffold comprising the acrylated chitosan and acrylate polymer.