

alternative solutions to a particular problem, and/or a plurality of different uses of one or more systems and/or articles. In one aspect, the present invention refers to a bioreactor system, comprising a reactor chamber having a volume of equal to or less than about 50 milliliters and containing a liquid growth medium including at least one biological cell and a buffer and a gaseous headspace containing oxygen and carbon dioxide above the liquid growth medium; a first inlet connecting (i) a gas manifold connected to a source(s) of carbon dioxide gas and oxygen to (ii) the gaseous headspace; a second inlet connecting a first reservoir comprising sterile water to the liquid growth medium; a third inlet connecting a second reservoir comprising an alkaline liquid to the liquid growth medium; a dissolved carbon dioxide sensor disposed in the liquid growth medium; an oxygen sensor disposed in the liquid growth medium; a pH sensor disposed in the liquid growth medium; and optionally, a gas-permeable membrane that separates the gaseous headspace and the liquid growth medium, and/or a fourth inlet connecting a third reservoir comprising a feed culture medium,, wherein the bioreactor system achieves a K_{La} of carbon dioxide between the gaseous headspace and the liquid growth medium of at least about 0.1 hour⁻¹ to less than 15 hours⁻¹.

In one example, a bioreactor system is provided. In certain examples, the bioreactor system comprises a reactor chamber having a volume of equal to or less than about 50 milliliters and containing a liquid growth medium including at least one biological cell and a buffer and a gaseous headspace containing carbon dioxide above the liquid growth medium, a first inlet connecting a source of carbon dioxide gas to the gaseous headspace, and a second inlet connecting a source of an alkaline liquid to the liquid growth medium.

In some examples, the bioreactor system comprises a reactor chamber having a volume of equal to or less than about 50 milliliters and containing a liquid growth medium including at least one biological cell and a buffer and a gaseous headspace containing carbon dioxide above the liquid growth medium, a first inlet connecting a source of carbon dioxide gas to the gaseous headspace, and a sensor within the reactor chamber configured to determine the concentration of carbon dioxide and/or pH within the liquid growth medium.

According to certain examples, a method of operating a bioreactor is described. In some examples, the method comprises providing a reactor chamber having a volume of equal to or less than about 50 milliliters and containing a liquid growth medium including at least one biological cell and a gaseous headspace containing carbon dioxide above the liquid growth medium, and operating the reactor such that the k_{La} of carbon dioxide between the headspace and the bulk of the liquid medium is at least about 0.1 hours⁻¹ and less than about 15 hours⁻¹. In one aspect, the present invention refers to a method of operating a bioreactor system, comprising providing a bioreactor system of the present invention; and operating the reactor such that the k_{La} of carbon

dioxide between the headspace and the bulk of the liquid medium is at least about 0.1 hours⁻¹ and less than about 15 hours⁻¹.

5 In certain examples, the method comprises providing a reactor chamber having a volume of equal to or less than about 50 milliliters. The reactor chamber contains, in some examples, a liquid growth medium including at least one biological cell, and a gaseous headspace containing carbon dioxide above the liquid growth medium. In some examples, the method comprises transporting a gas containing carbon dioxide to the gaseous headspace, and transporting an alkaline liquid to the liquid growth medium.

10 Other advantages and novel features of the present invention will become apparent from the following detailed description of various non-limiting embodiments of the invention when considered in conjunction with the accompanying figures. In cases

CLAIMS

1. A bioreactor system, comprising:

5 a reactor chamber having a volume of equal to or less than about 50 milliliters and containing a liquid growth medium including at least one biological cell and a buffer and a gaseous headspace containing oxygen and carbon dioxide above the liquid growth medium;

a first inlet connecting (i) a gas manifold connected to a source(s) of carbon dioxide gas and oxygen to (ii) the gaseous headspace;

10 a second inlet connecting a first reservoir comprising sterile water to the liquid growth medium;

a third inlet connecting a second reservoir comprising an alkaline liquid to the liquid growth medium;

a dissolved carbon dioxide sensor disposed in the liquid growth medium;

15 an oxygen sensor disposed in the liquid growth medium;

a pH sensor disposed in the liquid growth medium; and

optionally, a gas-permeable membrane that separates the gaseous headspace and the liquid growth medium, and/or a fourth inlet connecting a third reservoir comprising a feed culture medium,

20 wherein the bioreactor system achieves a K_La of carbon dioxide between the gaseous headspace and the liquid growth medium of at least about 0.1 hour^{-1} to less than 15 hours^{-1} .

2. The bioreactor system of claim 1, wherein the bioreactor system comprises a gas permeable membrane that separates the gaseous headspace and the liquid growth medium.

25 3. The bioreactor system of claim 1, wherein the bioreactor system comprises a fourth inlet connecting a third reservoir comprising a feed culture medium.

30 4. The bioreactor system of claim 1, wherein the bioreactor system comprises a gas-permeable membrane that separates the gaseous headspace and the liquid growth medium, and a fourth inlet connecting a third reservoir comprising a feed culture medium,

5. The bioreactor system of any one of claims 1-4, wherein an aspect ratio of the reactor chamber is less than about 10.

6. The bioreactor system of any one of claims 1-5, wherein the biological cell is a eukaryotic cell.

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7. The bioreactor system of any one of claims 1-6, wherein the biological cell is selected from the group consisting of single-cell organisms, plant cells, and animal cells.

8. The bioreactor system of claim 7, wherein the single-cell organism is a bacterium, a protozon, a trypanosome, an amoeba, a yeast cell, or algae.

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9. The bioreactor system of any one of claims 1-6, wherein the biological cell is from a multicellular organism.

10. The bioreactor system of any one of claims 1-6 and 9, wherein the biological cell is a mammalian cell selected from the group consisting of primate cells, bovine cells, horse cells, porcine cells, goat cells, dog cells, cat cells, rodent cells, human cells, and a hamster cells.

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11. The bioreactor system of any one of claims 1-6 and 9, wherein the biological cell is a cardiac cell, a fibroblast, a keratinocyte, a hepatocyte, a chondrocyte, a neural cell, an osteocyte, a muscle cell, a blood cell, an endothelial cell, an immune cell, or a stem cell.

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12. The bioreactor system of any one of claims 1-11, wherein the biological cell is a genetically engineered cell.

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13. The bioreactor system of any one of claims 1-12, wherein the gas manifold is connected to a source of carbon dioxide gas, and the source of carbon dioxide gas comprises substantially pure carbon dioxide

14. The bioreactor system of any one of claims 1-13, wherein the alkaline liquid contains bicarbonate ions.

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15. The bioreactor system of any one of claims 1-14, wherein the alkaline liquid has a pH of greater than or equal to 7.5.

16. The bioreactor system of any one of claims 1-15, comprising a fifth inlet connecting a fourth reservoir comprising an acidic material to the liquid growth medium.

5 17. The bioreactor system of claim 16, wherein the acidic material has a pH of less than or equal to 6.5.

18. The bioreactor system of any one of claims 1-17, wherein a partial pressure of carbon dioxide above the liquid growth medium is between about 0% and about 20%.

10 19. The bioreactor system of any one of claims 1-18, wherein a total pressure in the gaseous headspace is from about 0 psi to about 15 psi.

20. The bioreactor system of claim 1, wherein the gaseous headspace and the liquid growth
15 medium are in direct contact.

21. The bioreactor system of claim 1, wherein the cell comprises a Chinese hamster ovary (CHO) cell.

20 22. The bioreactor system of any one of claims 1-21, wherein the reactor chamber is configured to contain a volume of the liquid medium of equal to or greater than 10 microliters and less than about 50 milliliters.

23. A method of operating a bioreactor system, comprising:
25 providing a bioreactor system of any one of claims 1-22; and
operating the bioreactor system such that the k_{La} of carbon dioxide between the gaseous headspace and the liquid growth medium is at least about 0.1 hours⁻¹ to less than about 15 hours⁻¹.