US 20230294097A1

(19) **United States**

(12) **Patent Application Publication**

(10) **Pub. No.: US 2023/0294097 A1**

(43) **Pub. Date: Sep. 21, 2023**

Hasty et al.

(54) **ELECTROCHEMICAL CELLULAR CIRCUITS**

(71) Applicant: **The Regents of the University of California**, Oakland, CA (US)

(72) Inventors: **Jeff Hasty**, Encinitas, CA (US); **Muhammad Omar Din**, San Diego, CA (US); **Aida Martin Galan**, La Jolla, CA (US)

(21) Appl. No.: **17/923,129**

(22) PCT Filed: **May 4, 2021**

(86) PCT No.: **PCT/US2021/030687**

§ 371 (c)(1),
(2) Date: **Nov. 3, 2022**

Related U.S. Application Data

(60) Provisional application No. 63/019,664, filed on May 4, 2020.

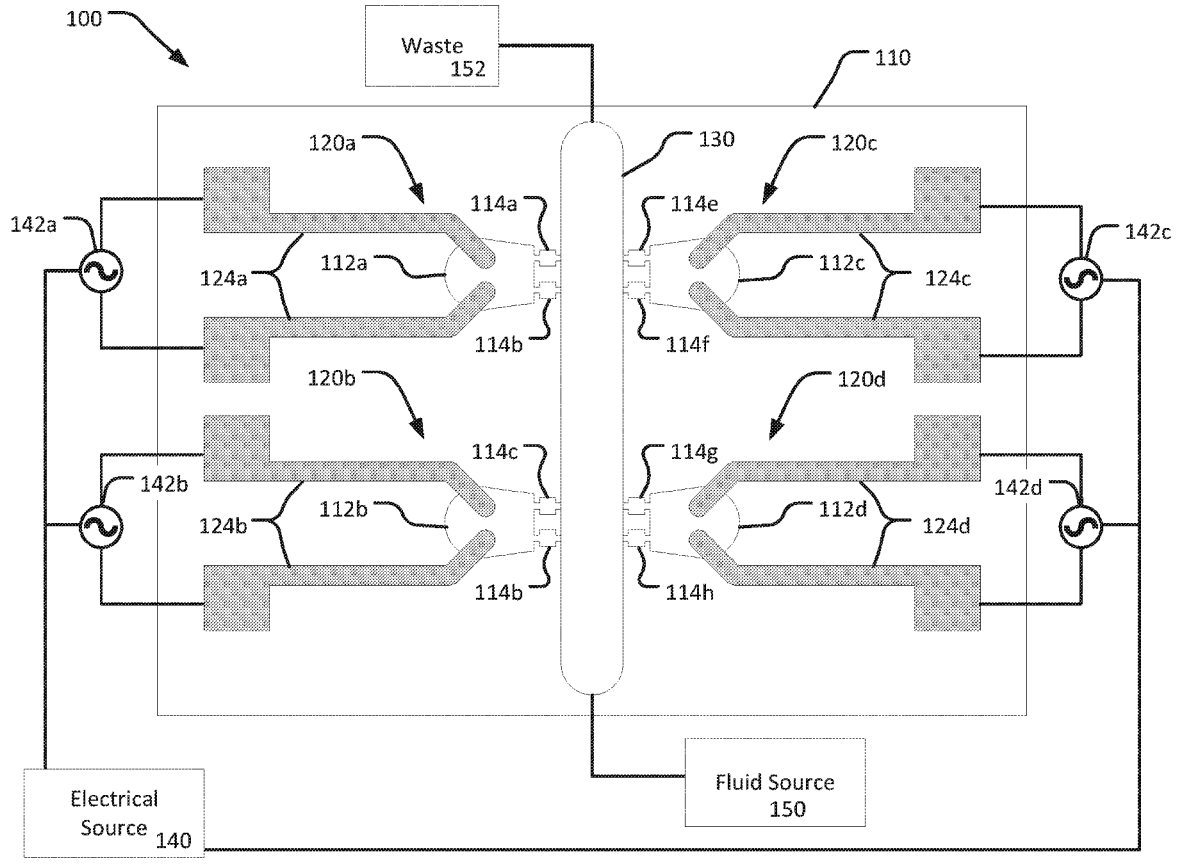
Publication Classification

(51) **Int. Cl.**
B01L 3/00 (2006.01)

(52) **U.S. Cl.**
CPC ... ***B01L 3/502761*** (2013.01); *B01L 2200/0652* (2013.01); *B01L 2300/0645* (2013.01); *B01L 2300/0877* (2013.01)

(57) **ABSTRACT**

Disclosed herein is a method of measuring a cell population, the method including culturing a cell population in a growth medium, wherein the cell population includes a cell strain including a control gene and a promoter sequence; positioning first and second electrodes in contact with the growth medium, wherein the first and second electrodes are connected to different terminals of an electrical source; applying an electrical signal between the two electrodes; measuring an electrical parameter of the growth medium as a function of time; and determining information about the cell population based upon the measured electrical parameter.



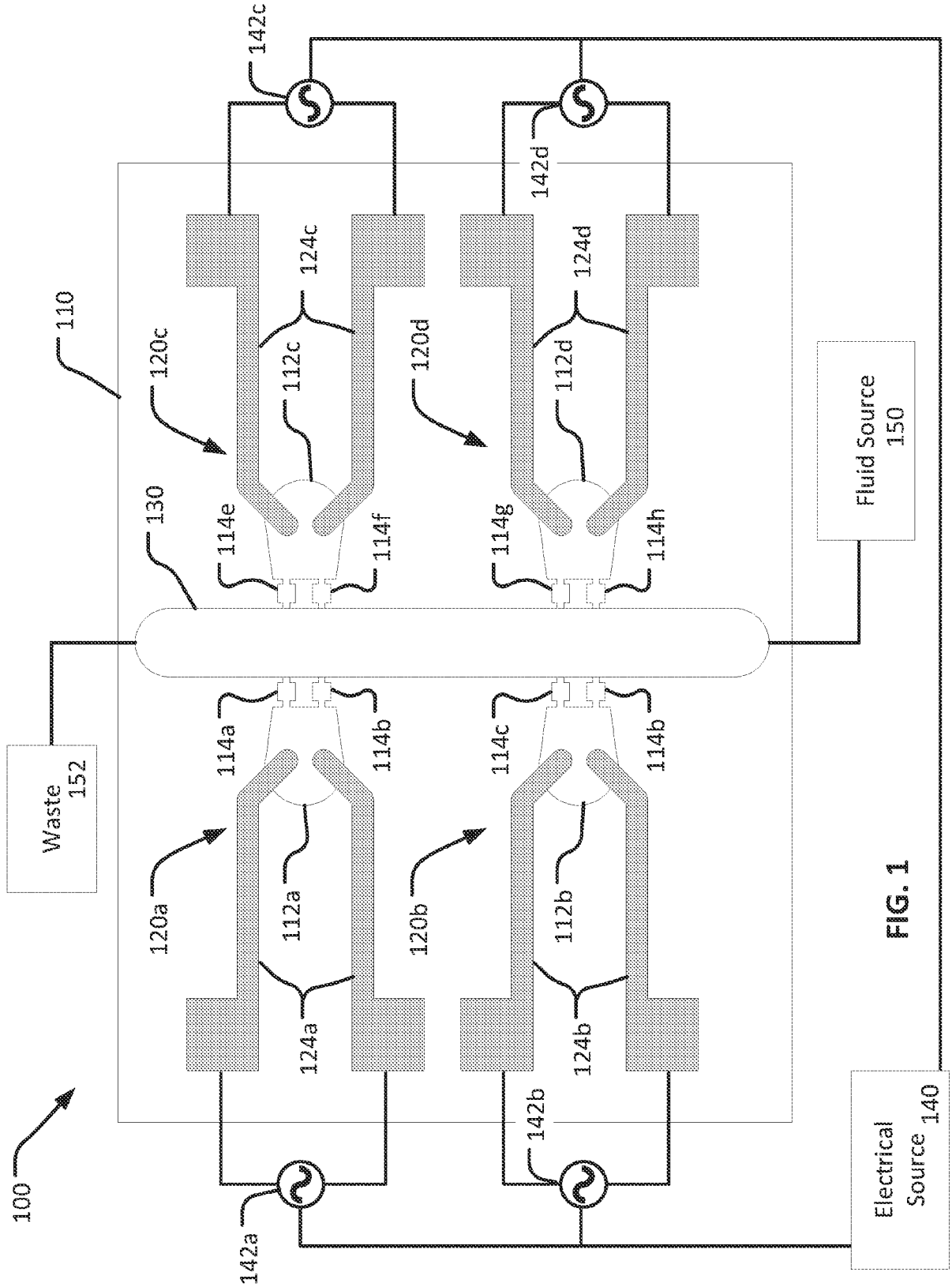

100
110
120a
120b
120c
120d
112a
112b
112c
112d
114a
114b
114c
114e
114f
114g
114h
124a
124b
124c
124d
130
142a
142b
142c
142d
Waste 152
Fluid Source 150
Electrical Source 140

FIG. 1

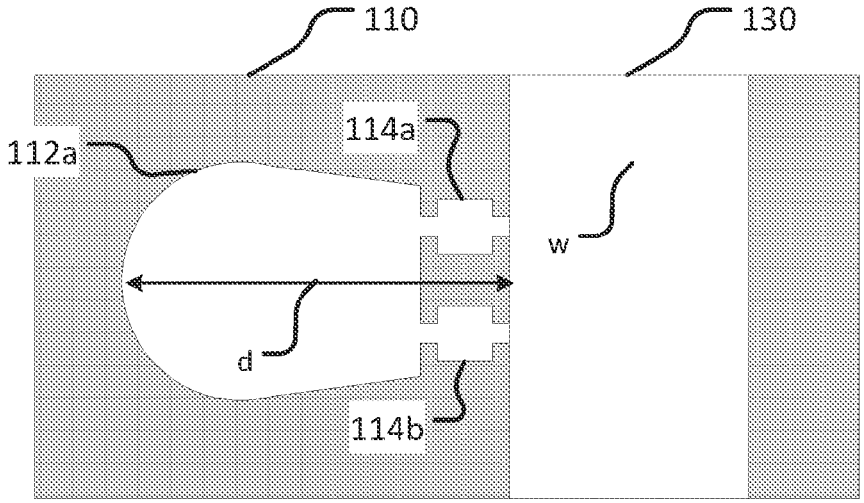


FIG. 2A

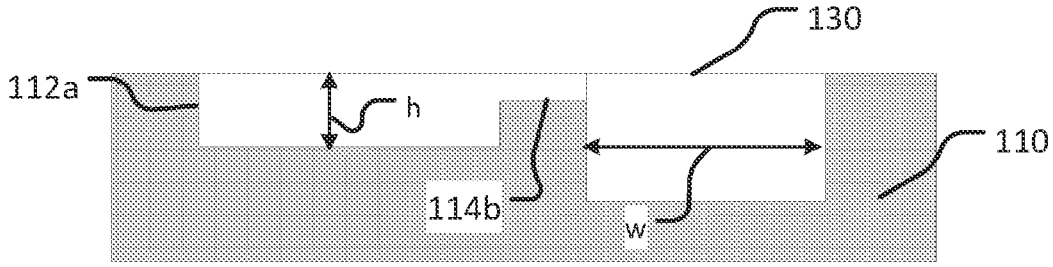


FIG. 2B

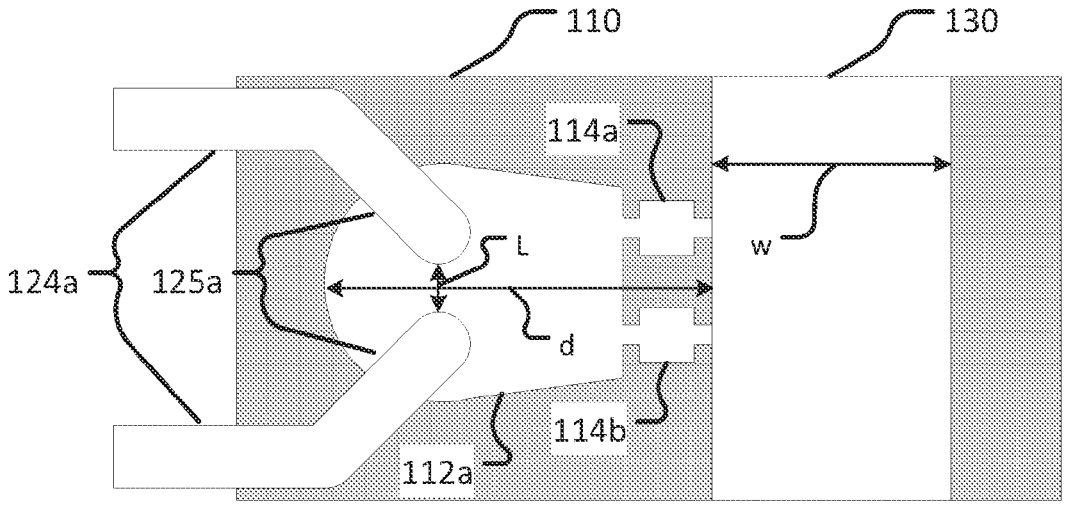


FIG. 2C

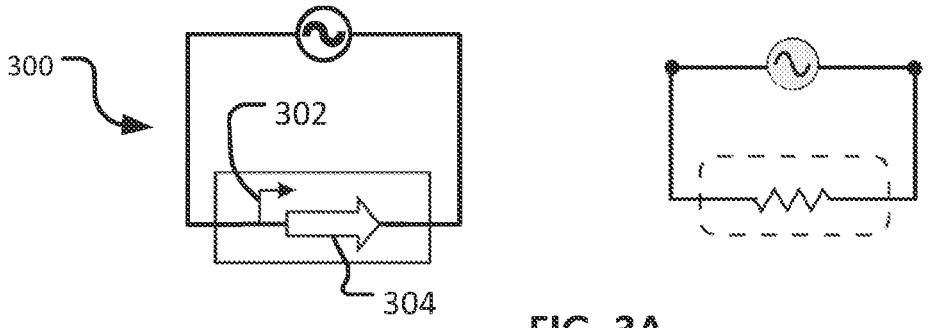


FIG. 3A

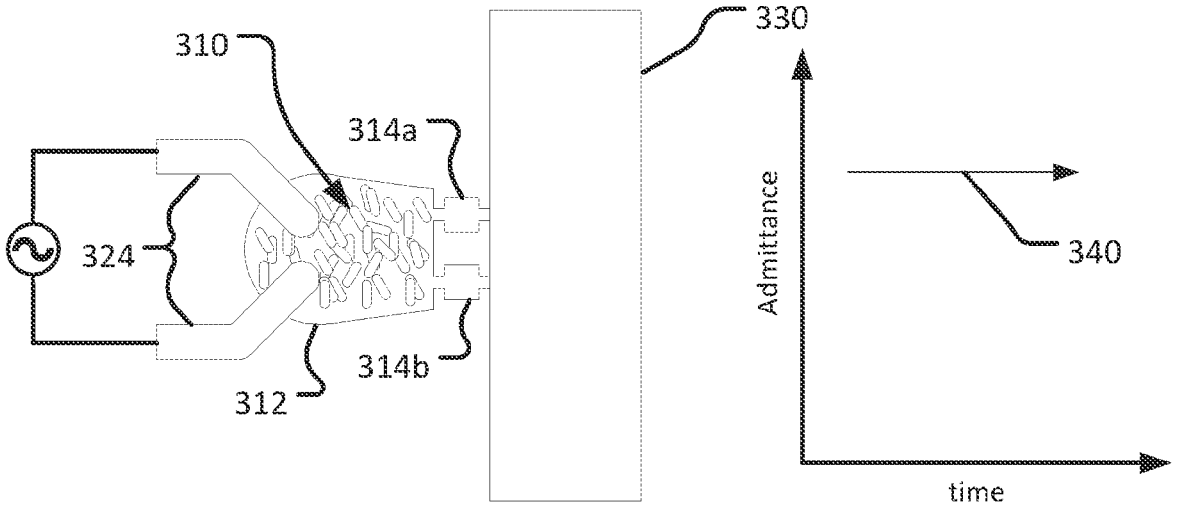


FIG. 3B

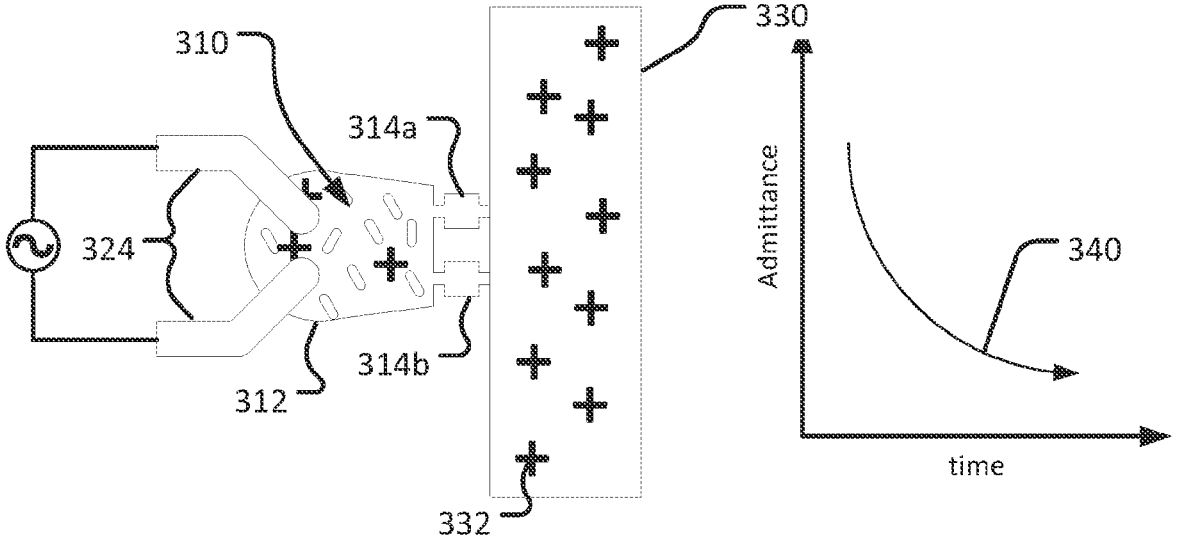


FIG. 3C

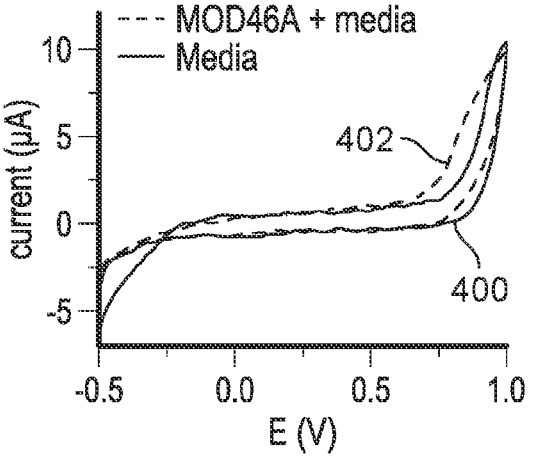


FIG. 4A

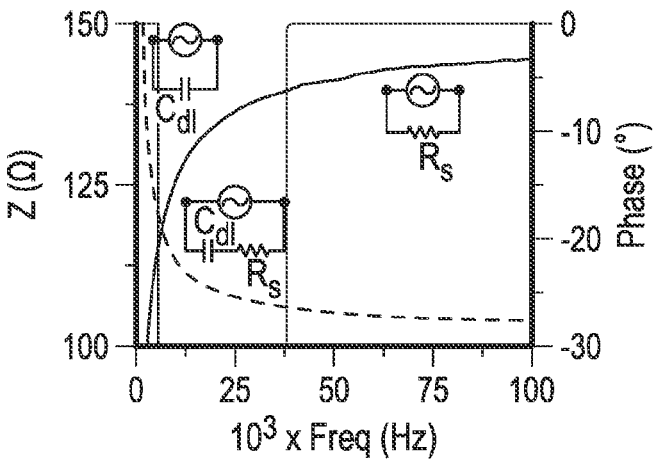


FIG. 4B

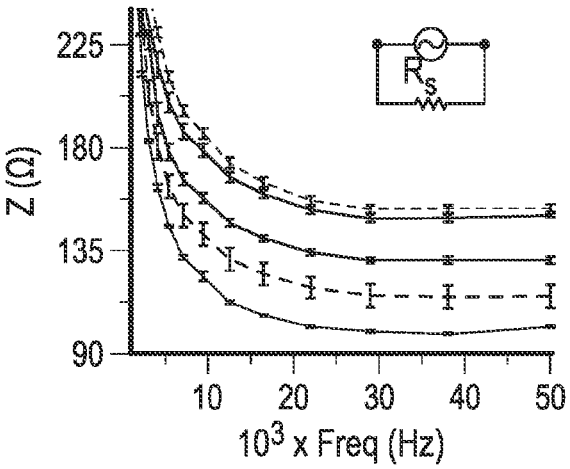


FIG. 4C

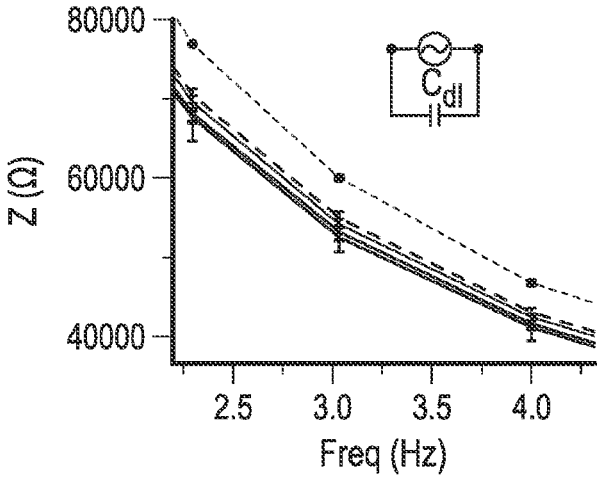


FIG. 4D

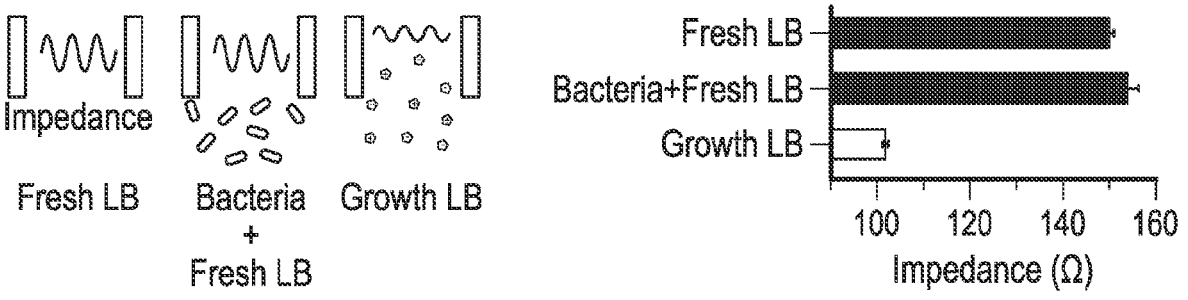


FIG. 4E

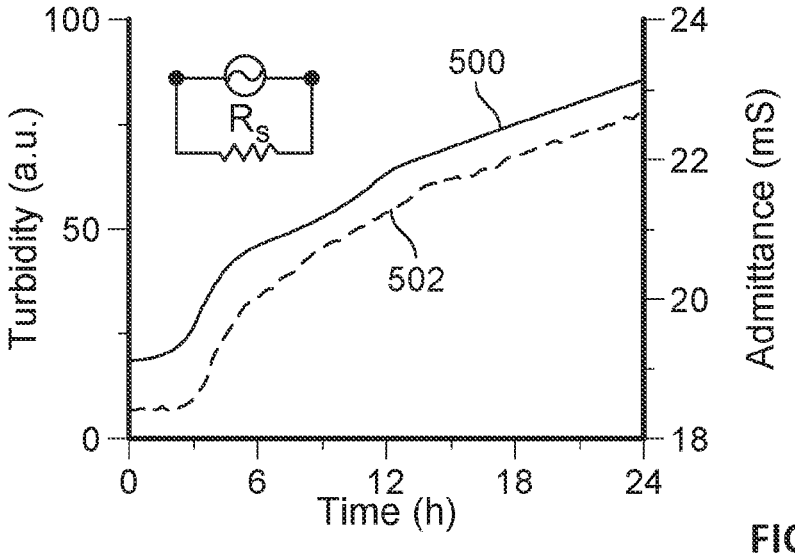


FIG. 5A

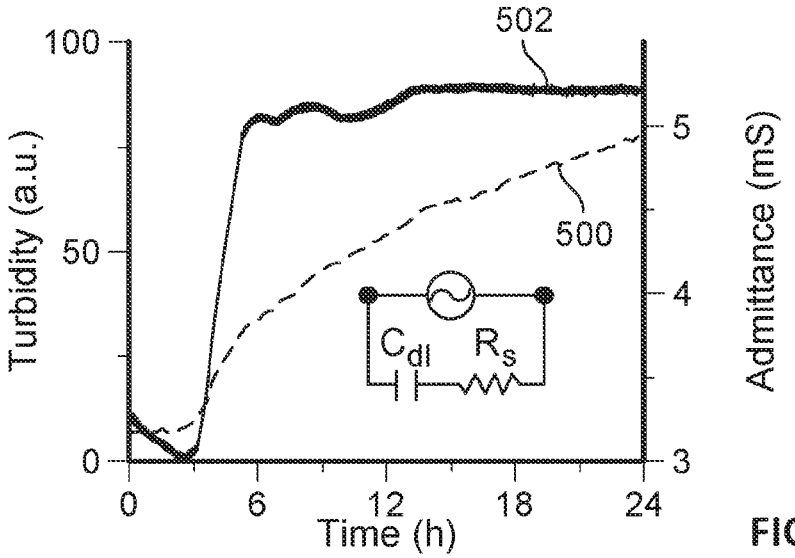


FIG. 5B

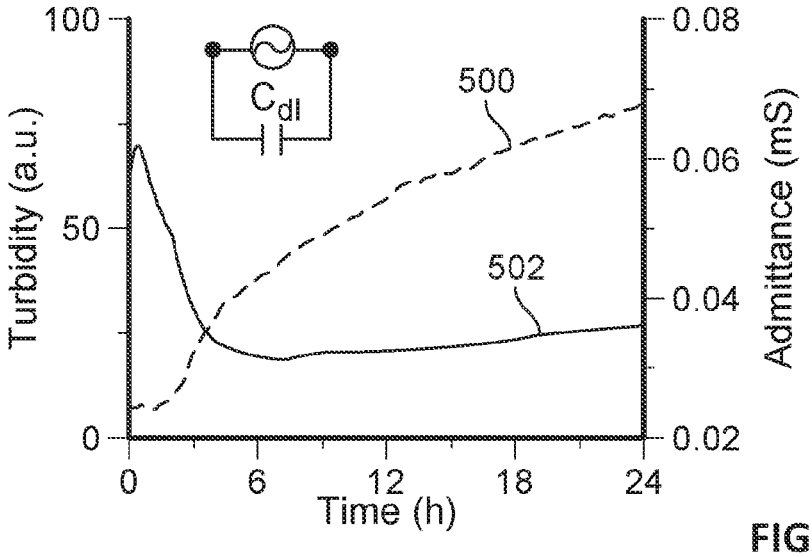


FIG. 5C

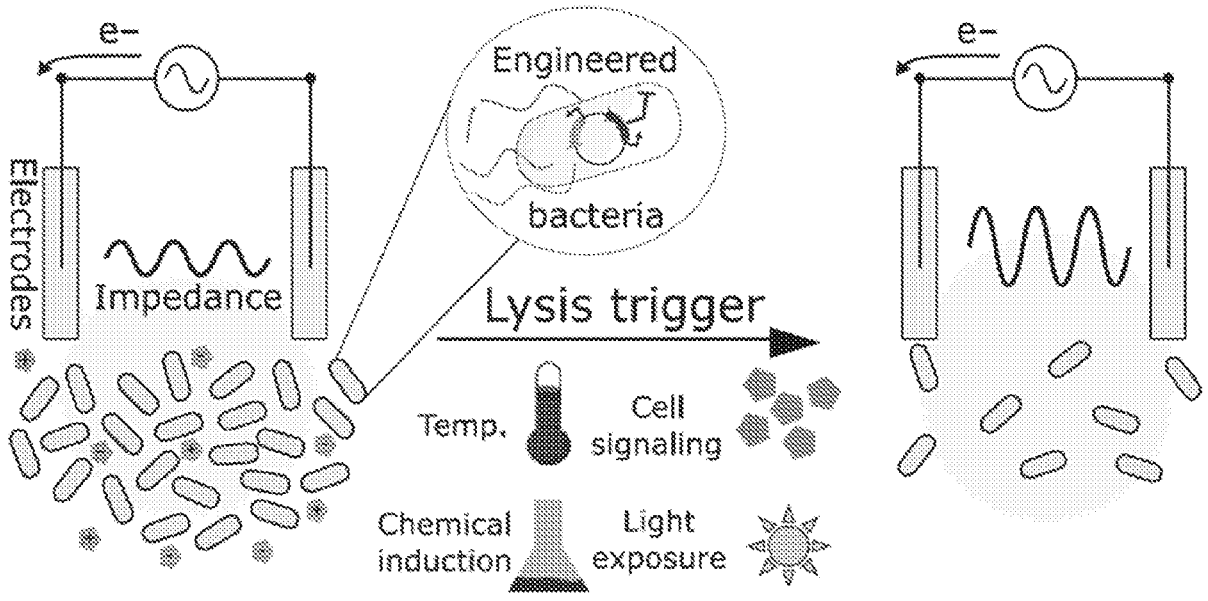

e−
Electrodes
Impedance
Engineered
bacteria
Lysis trigger
Temp.
Cell
signaling
Chemical
induction
Light
exposure
e−

FIG. 6A

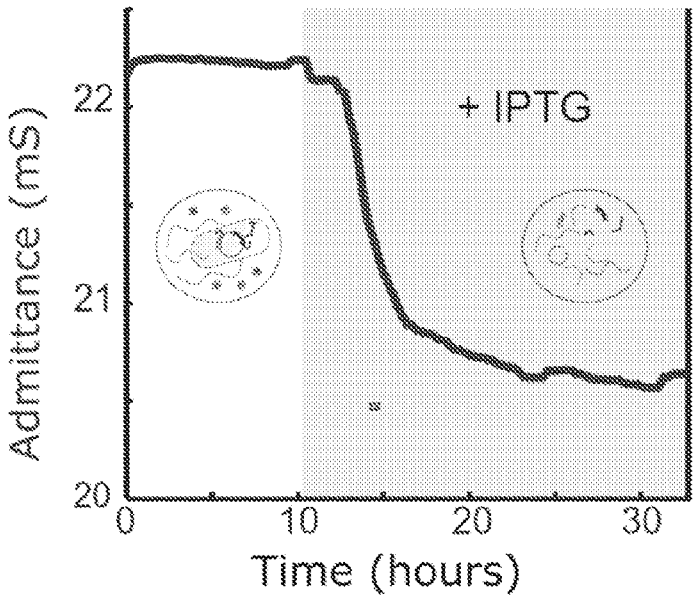


FIG. 6B

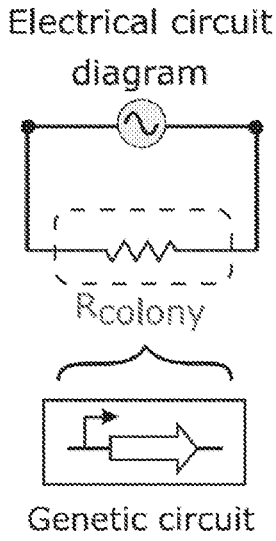


FIG. 6C

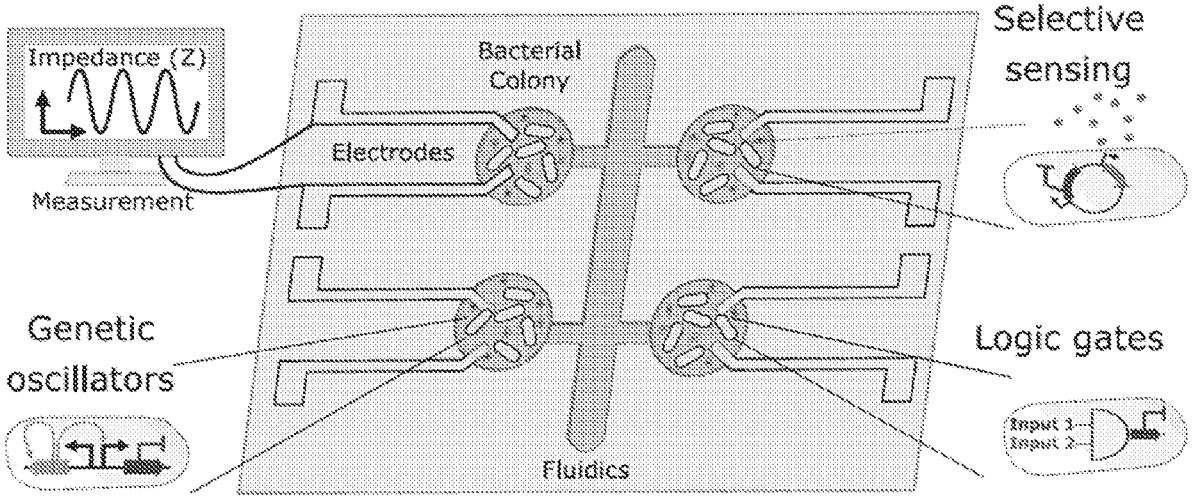


FIG. 6D

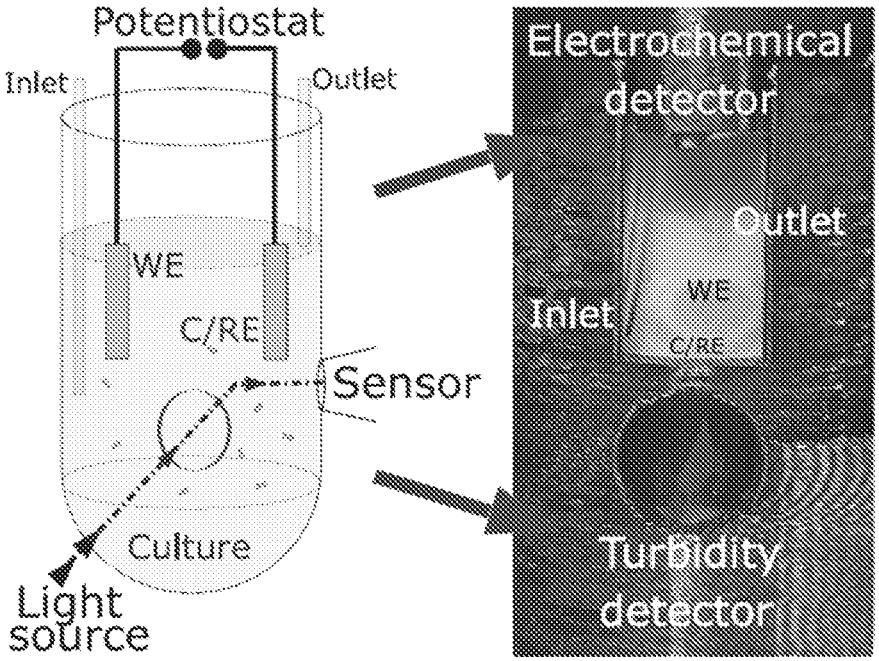

Potentiostat
Inlet
Outlet
WE
C/RE
Sensor
Culture
Light source
Electrochemical detector
Outlet
Inlet
WE
C/RE
Turbidity detector

FIG. 7A

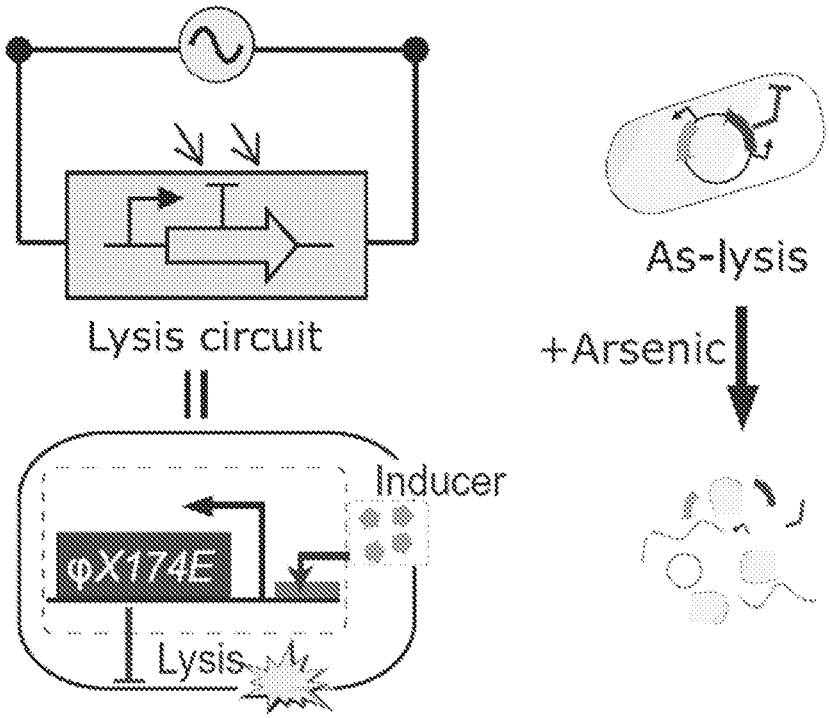

Lysis circuit
Inducer
φX174E
Lysis
As-lysis
+Arsenic

FIG. 7B

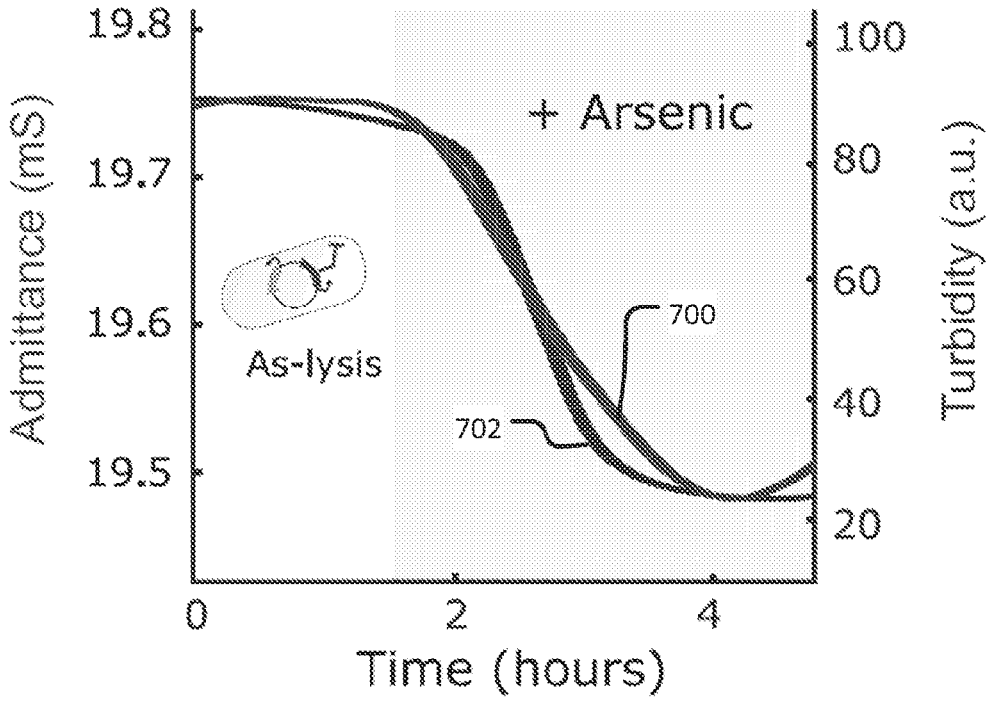

19.8
19.7
19.6
19.5
Admittance (mS)
100
80
60
40
20
Turbidity (a.u.)
+ Arsenic
As-lysis
700
702
0
2
4
Time (hours)

FIG. 7C

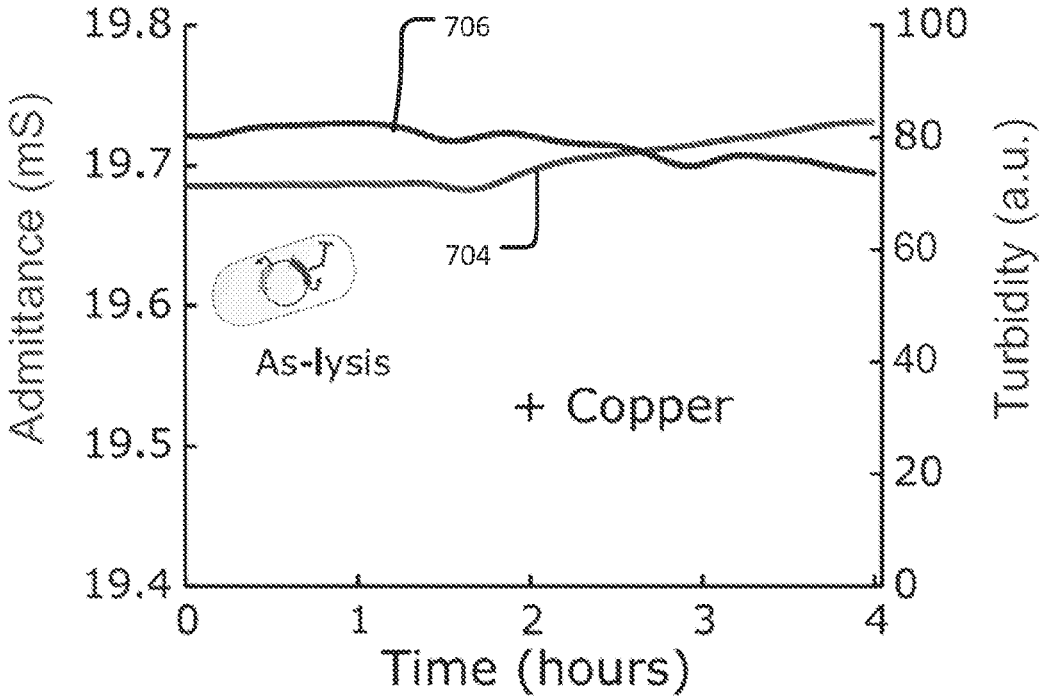

19.8
19.7
19.6
19.5
19.4
Admittance (mS)
100
80
60
40
20
0
Turbidity (a.u.)
706
704
As-lysis
+ Copper
0
1
2
3
4
Time (hours)

FIG. 7D

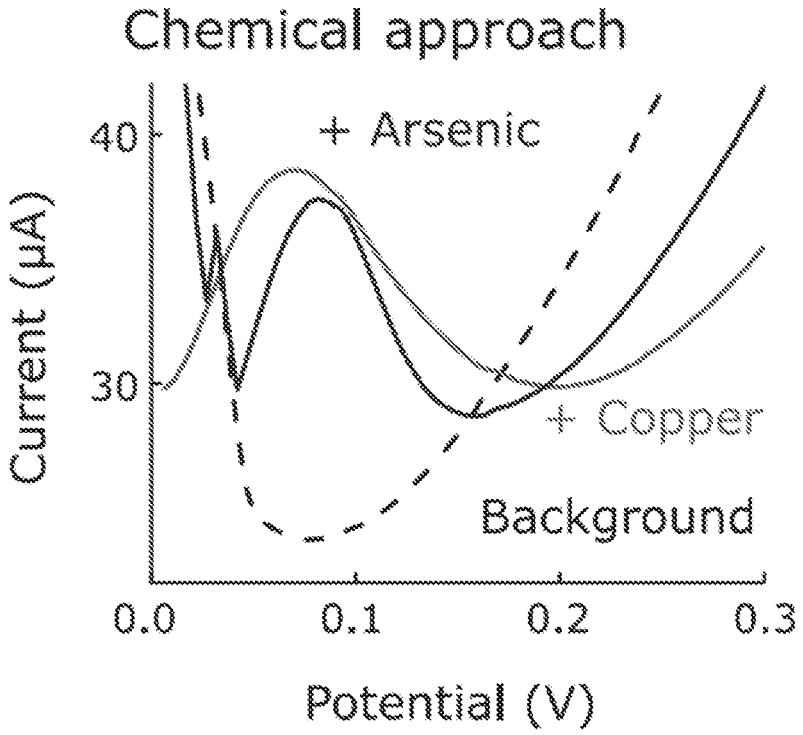

Chemical approach
+ Arsenic
+ Copper
Background
Current (μA)
40
30
0.0
0.1
0.2
0.3
Potential (V)

FIG. 7E

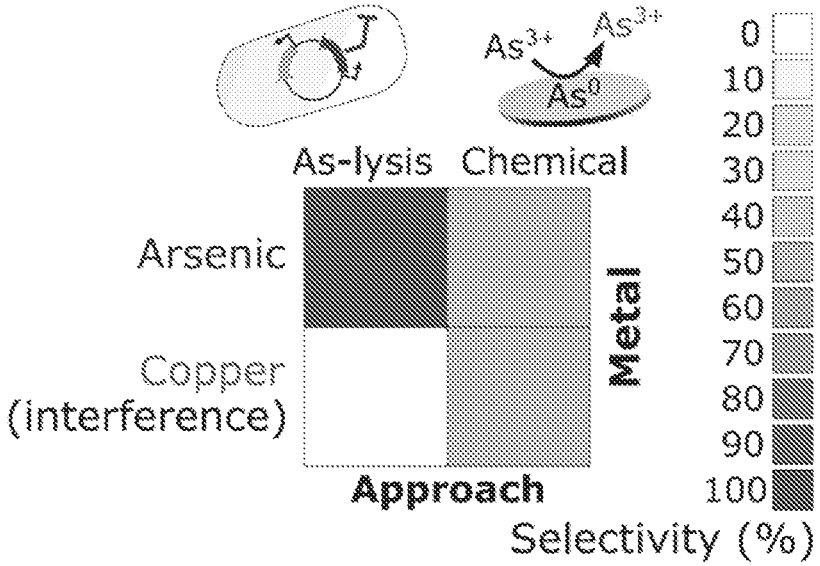

As3+
As3+
As0
As-lysis
Chemical
Arsenic
Copper
(interference)
Metal
Approach
0
10
20
30
40
50
60
70
80
90
100
Selectivity (%)

FIG. 7F

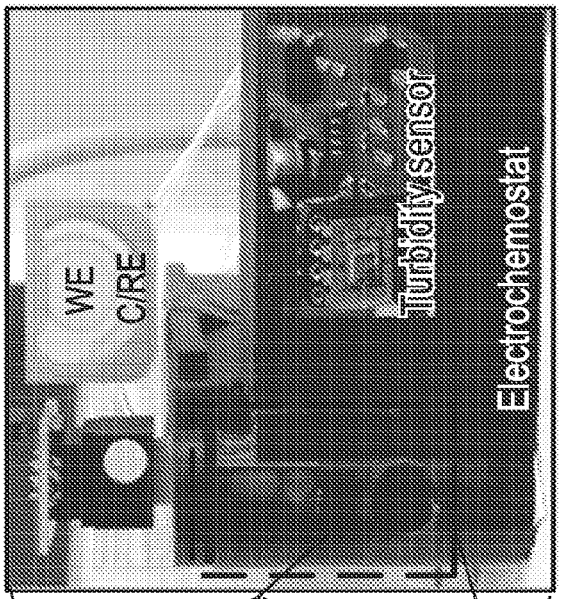

WE
C/RE
Turbidity sensor
Electrochemostat
1 cm
Electrode
Culture

FIG. 8B

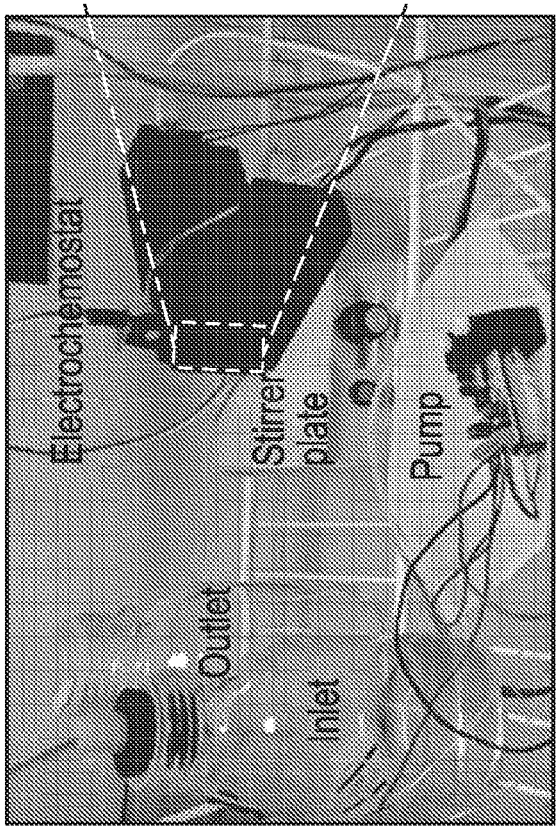

Electrochemostat
Stirrer plate
Pump
Outlet
Inlet

FIG. 8A

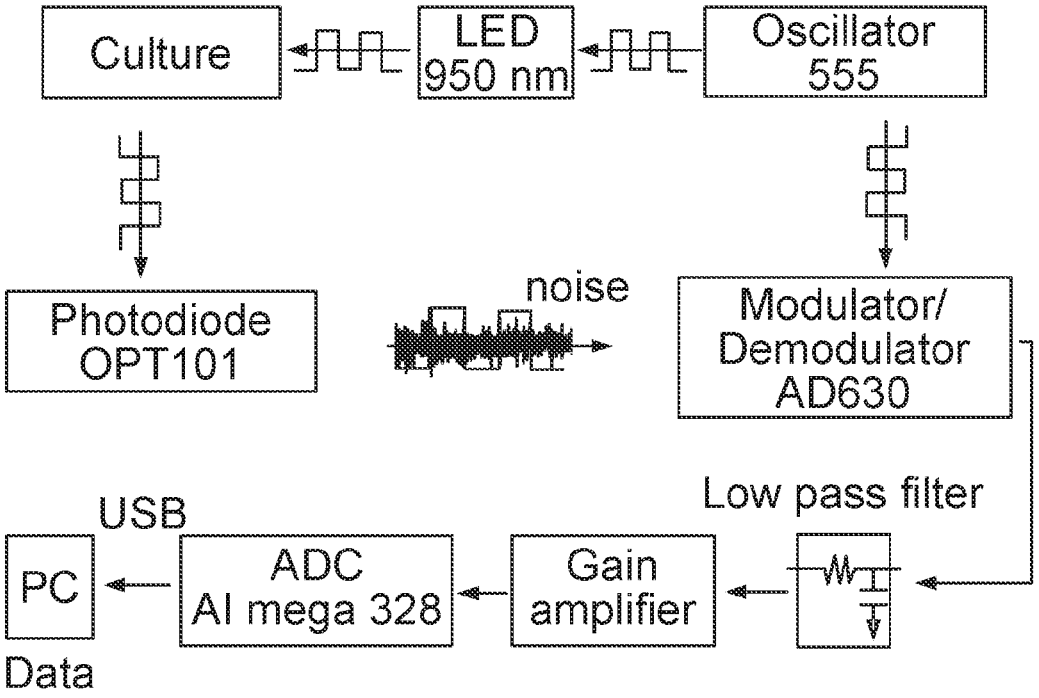

Culture
LED
950 nm
Oscillator
555
Photodiode
OPT101
noise
Modulator/
Demodulator
AD630
Low pass filter
USB
PC
ADC
AI mega 328
Gain
amplifier
Data

FIG. 8C

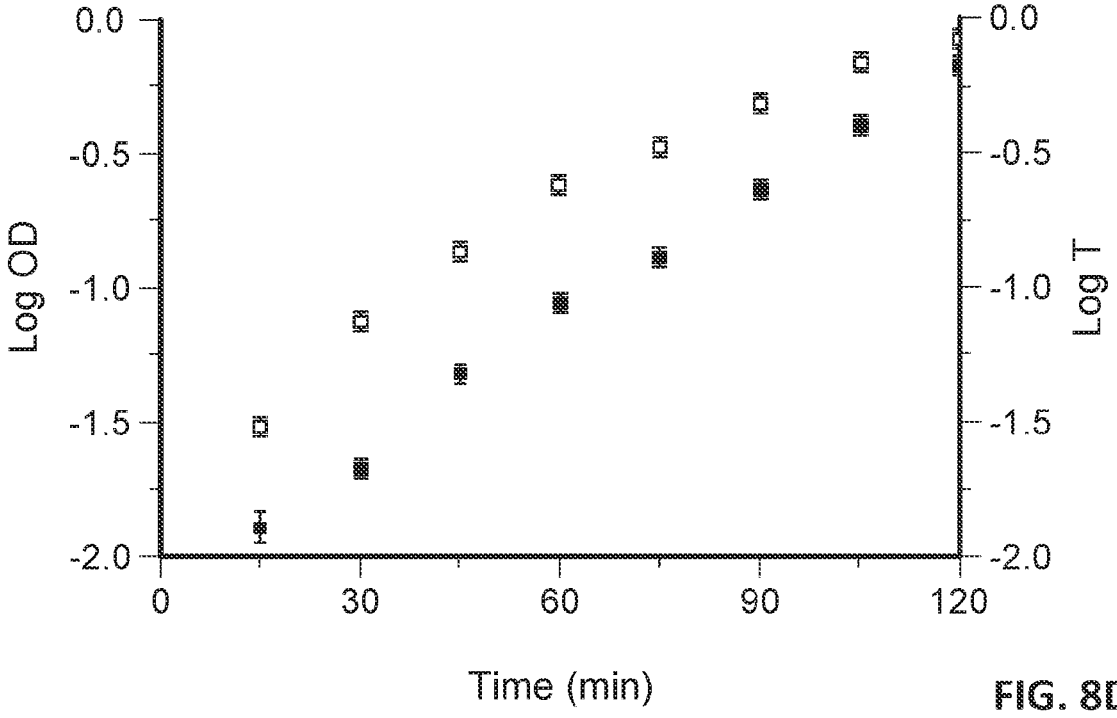

0.0
-0.5
-1.0
-1.5
-2.0
Log OD
Log T
0
30
60
90
120
Time (min)

FIG. 8D

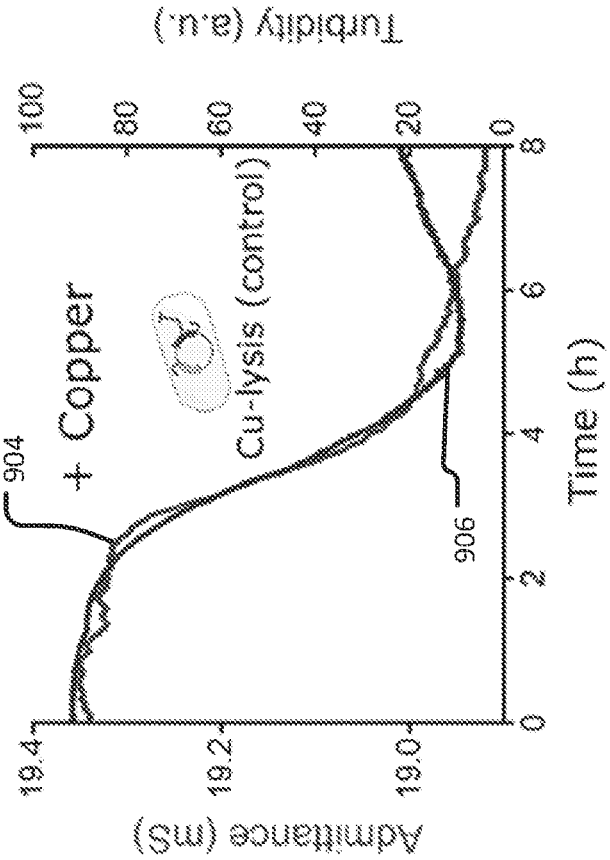

Turbidity (a.u.)
100
80
60
40
20
0
8
6
4
2
0
Time (h)
Admittance (mS)
19.4
19.2
19.0
+ Copper
Cu-lysis (control)
904
906

FIG. 9B

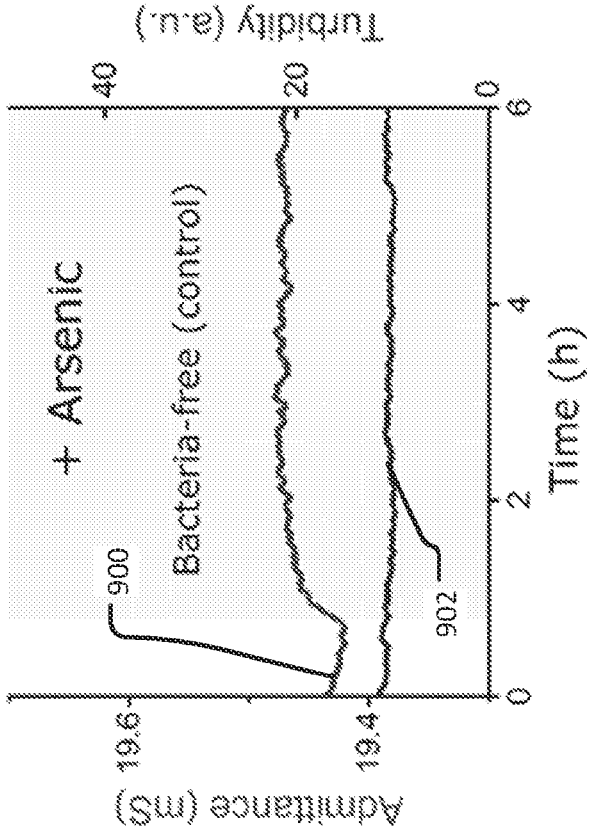

Turbidity (a.u.)
40
20
0
6
4
2
0
Time (h)
Admittance (mS)
19.6
19.4
+ Arsenic
Bacteria-free (control)
900
902

FIG. 9A

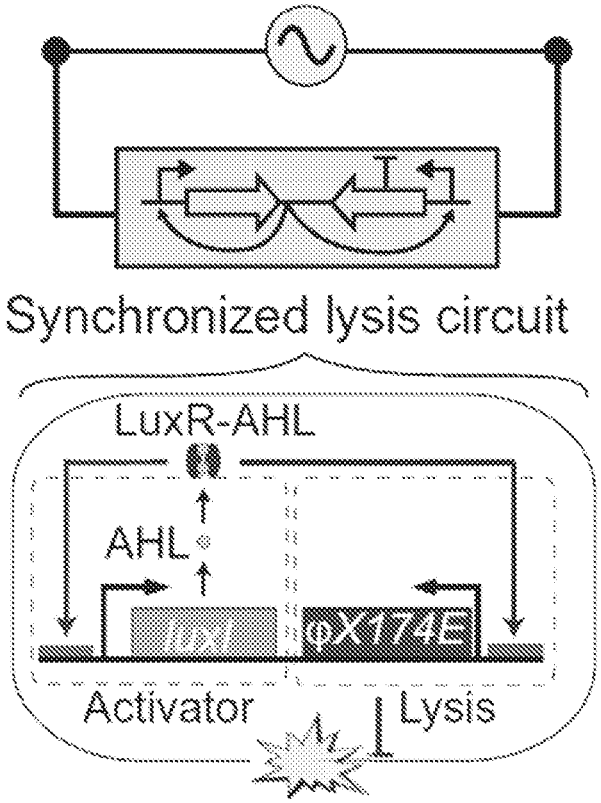


FIG. 10A

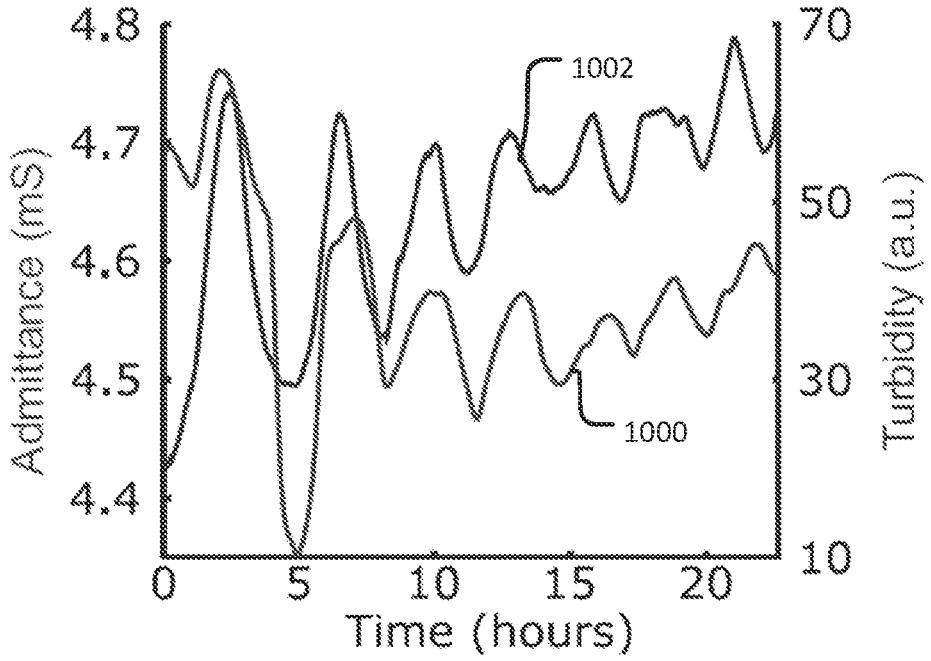


FIG. 10B

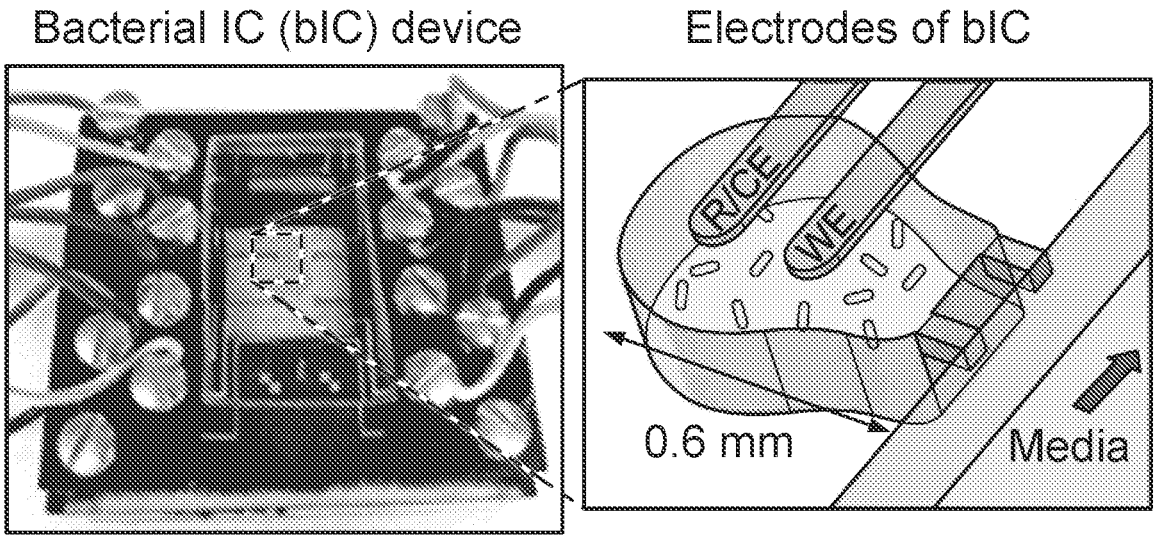


FIG. 11A

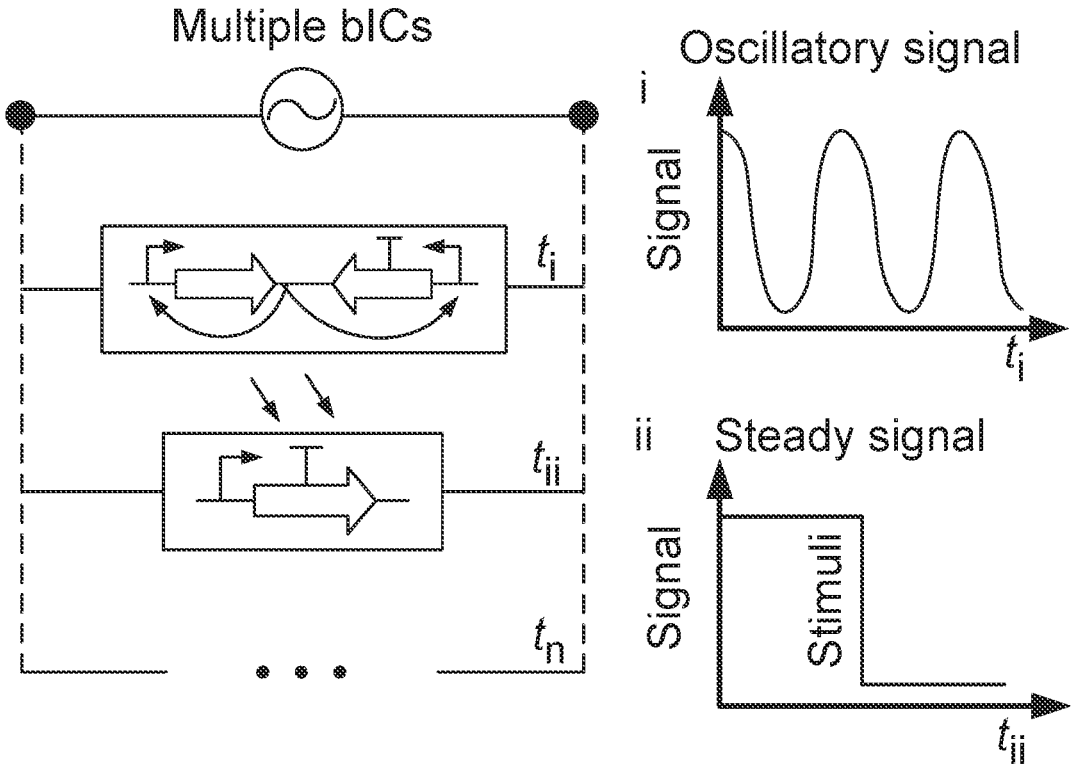


FIG. 11B

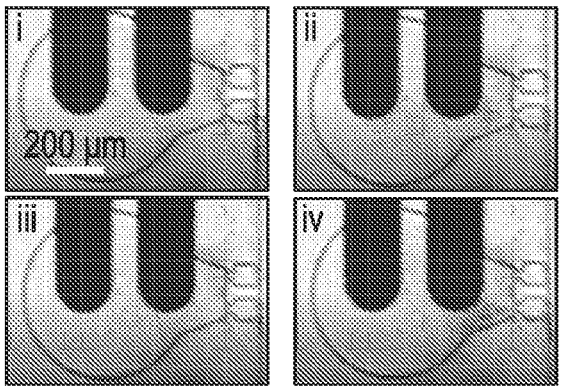


FIG. 11C

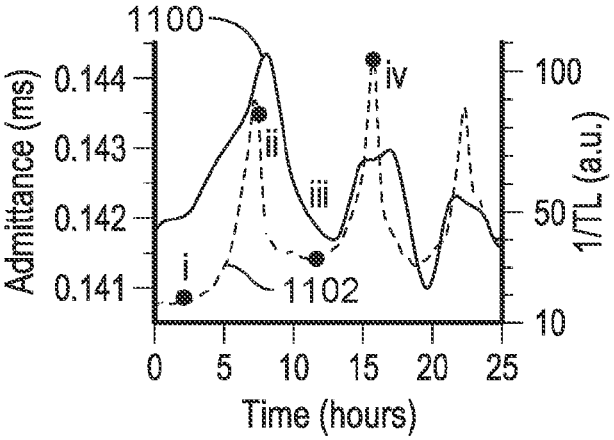


FIG. 11D

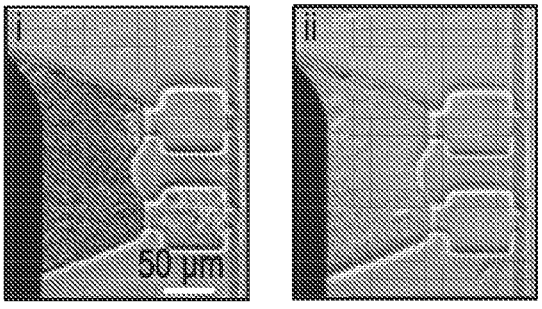


FIG. 11E

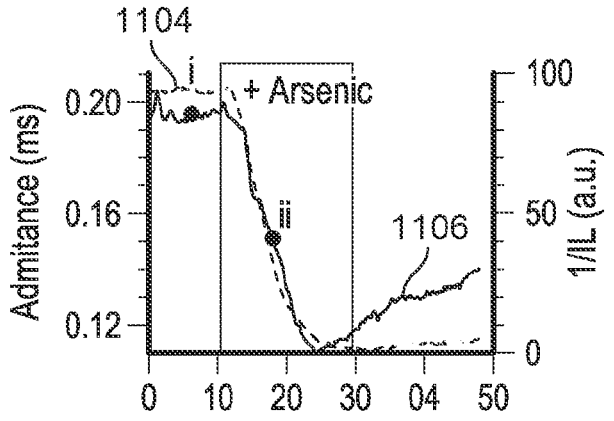


FIG. 11F

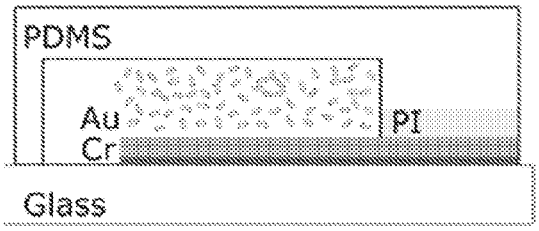


FIG. 12A

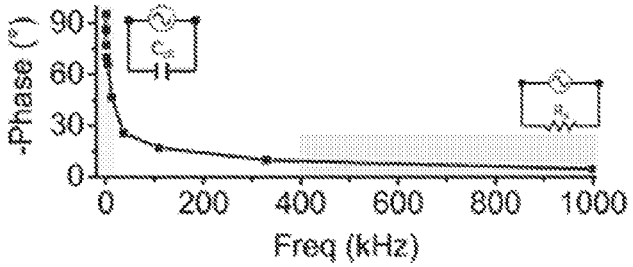


FIG. 12C

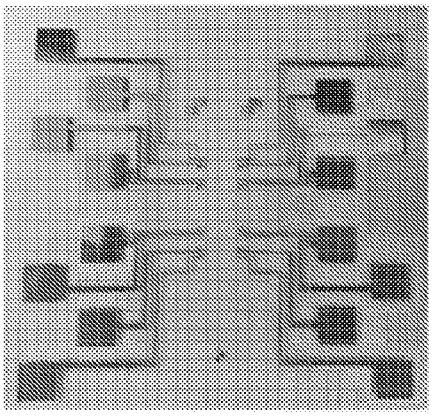


FIG. 12B

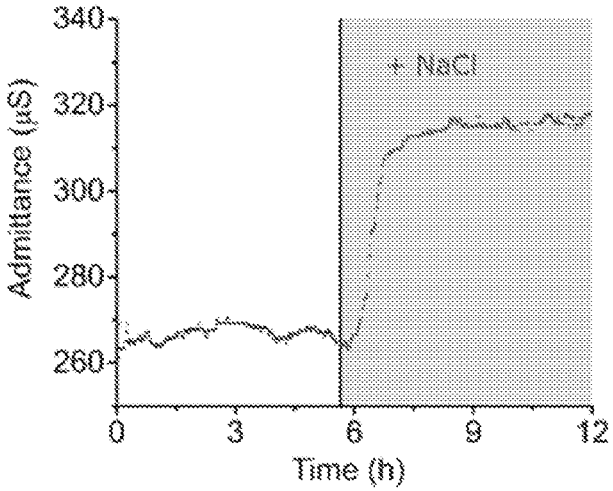
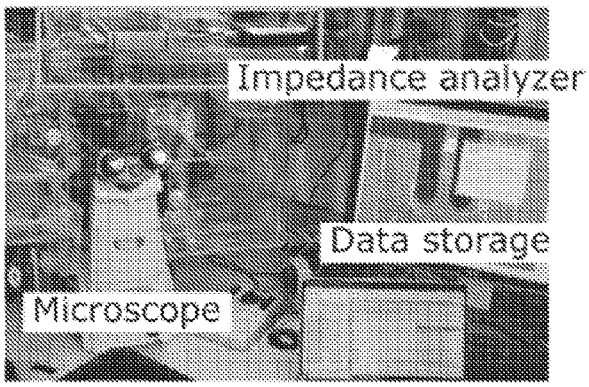


FIG. 12D



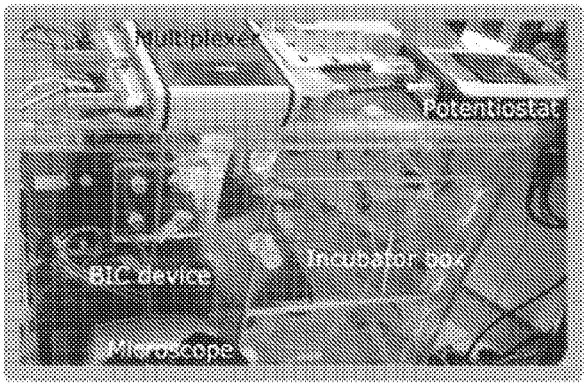


FIG. 12E

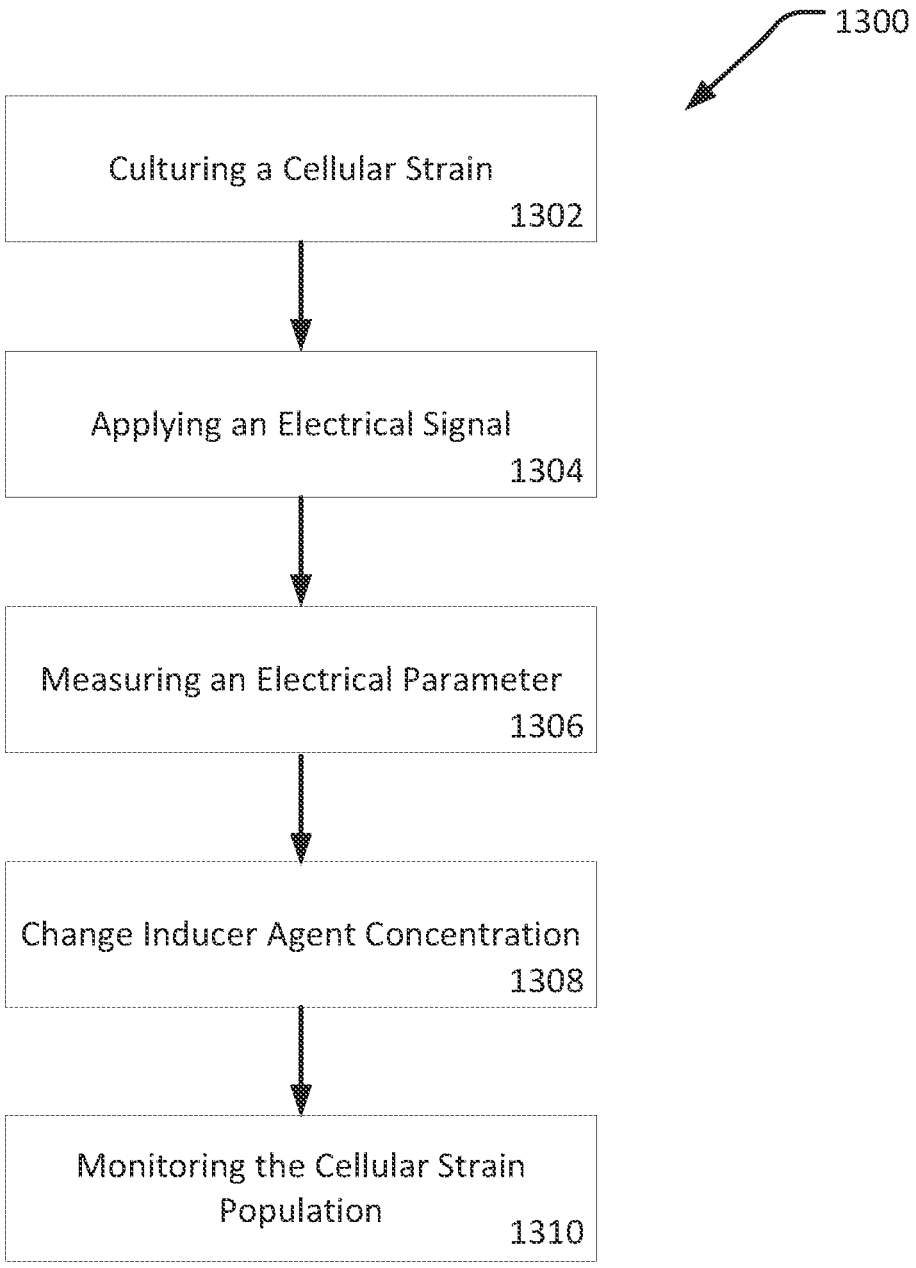

1300
Culturing a Cellular Strain
1302
Applying an Electrical Signal
1304
Measuring an Electrical Parameter
1306
Change Inducer Agent Concentration
1308
Monitoring the Cellular Strain Population
1310

FIG. 13

ELECTROCHEMICAL CELLULAR CIRCUITS

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority to U.S. Provisional Pat. Application No. 63/019,664, filed on May 4, 2020, the entire contents of which are incorporated herein by reference.

FIELD OF THE DISCLOSURE

[0002] The disclosure relates to cellular circuitry and to the interface of cellular populations with electrical circuits and circuit elements.

BACKGROUND

[0003] Conventional sensing techniques for organic and inorganic substances include a variety of molecular recognition strategies in which biomolecules bind to, and function as reporters for, the substances. To ensure that substances are detecting using these techniques, binding events are accompanied by changes in detectable properties of the biomolecules. Examples of such properties include optical absorption and emission, solution conductivity, and mobility. Applying these techniques to monitor cellular population dynamics in genetic circuits presents a number of challenges that can make the techniques impractical.

SUMMARY

[0004] This disclosure features methods, devices, and systems in which genetic circuits that include cellular populations are integrated with electrical circuits. Direct electrical measurements of the cellular populations can be used to detect an analyte of interest (e.g., a target analyte), the concentration or amount of which can be used to assess cellular population dynamics.

[0005] Cells produce a number of metabolic products during regular cellular metabolic processes. As a population of cells grows in solution, the production and concentration of the metabolic products increases correspondingly. Conversely, as the population declines, the production and concentration of metabolic products declines. When a metabolic product is charged, the product can function as a charge carrier for electrical signals in the solution. Accordingly, conductive electrodes placed in a solution containing the cellular population and connected to an electrical source can be used to measure changes in one or more electrical parameters of the solution, such as impedance or admittance, or conductivity or resistance. Such measurements can be used to determine information about the cellular population in solution, as changes in the cellular population are correlated with changes in the concentrations of metabolic products in solution.

[0006] Cellular populations can be used as biological sensors for a variety of target analytes. Cellular strains from which the cellular population are composed can be genetically engineered to include at least one control gene that is responsive to a promoter sequence. The promoter sequence can bind to an inducer agent, which can be the target analyte or can bind the target analyte, and can regulate transcription of the control gene based upon the presence of the target analyte. When the target agent is introduced into a solution containing the cellular population, the activated control gene can modify the metabolic output of the cellular population, increasing or decreasing the concentration of metabolic products in solution. For example, the control gene can cause the cellular population to lyse, thereby ceasing production of metabolic products. The change in metabolic product concentration modulates the electrical properties of the solution, and by measuring the electrical properties, the induced change in the cellular population — and therefore the presence of the target analyte — can be determined. Cellular strains that are used in the cellular populations can include more than one control gene, and more than one promoter. In some embodiments, multiple cellular strains are included in a cellular population that is used in a sensor.

[0007] Embodiments of the methods, devices, and systems described herein may provide one or more of the following advantages. First, the methods, devices, and systems can be used for long term molecular sensing. In a solution containing a target analyte, a cellular population that is also present in the solution can receive an ongoing supply of a growth medium. The cellular population can therefore survive and reproduce for long time periods, enabling the monitoring of the target analyte over long time periods. This can be advantageous in situations where the observation schedule for the target analyte is infrequent.

[0008] Second, some embodiments disclosed herein can utilize low volume sampling to produce a measureable signal change. Cellular populations can be contained in reservoirs (e.g., microfluidic wells) on a fluidic chip, and the wells can have relatively low volumes (e.g., less than 10 μL). This permits analyte detection using very low sample volumes.

[0009] Third, some embodiments disclosed herein can detect multiple analytes in multiplexed fashion. Multiple microwells on a fluidic chip can be in fluid connection with a supply channel, and each microwell can contain a cellular population that includes a different cellular strain responsive to a different target analyte. Detection of multiple target analytes in a single solution allows for multiplexed assays to be performed on a single fluidic chip.

[0010] Fourth, some embodiments disclosed herein operate as feedback circuits capable of determining the presence and quantity of a target analyte in solution. Cellular strains including two or more promoters coupled to corresponding control genes provide an oscillatory circuit capable of responding to the presence and level of an analyte. This aids in monitoring population mechanics over time in response to changing levels of target analyte and delivering near-real time measurements in situ.

[0011] Fifth, some embodiments disclosed herein implement small form-factor sensors for the detection of target analytes. Systems that include microwell arrays formed on fluidic chips can enable field detection of target analytes in a variety of environments and applications, including food analysis, agricultural production, and medical applications.

[0012] In general, in a first aspect, the disclosure includes a method of measuring a cell population, the method including culturing a cell population in a growth medium, wherein the cell population includes a cell strain including a control gene and a promoter sequence; positioning first and second electrodes in contact with the growth medium, wherein the first and second electrodes are connected to different terminals of an electrical source; applying an electrical signal

between the two electrodes; measuring an electrical parameter of the growth medium as a function of time; and determining information about the cell population based upon the measured electrical parameter.

[0013] In some embodiments, the electrical parameter can include an electrical admittance of the growth medium. The electrical parameter can include an electrical impedance of the growth medium. The electrical parameter can include a conductance of the growth medium. The electrical parameter can include a resistance of the growth medium. Applying the electrical signal can include applying an electrical potential difference between the two electrodes. The cell population can include one or more different bacterial strains. The cell population can include one or more different yeast strains. The cell population can include one or more different mammalian cell strains. The cell population can include one or more different insect cell strains. The growth medium can be selected from the group consisting of a liquid medium and a solid medium. The solid medium can include a hydrogel. The solid medium can include at least one gel-based material.

[0014] In some embodiments, the method can further include maintaining the cell population in a chamber, and flowing the growth medium through the chamber. The method can further include measuring the electrical parameter in different portions of the flowing growth medium as a function of time. The control gene can be activated by at least one mechanism selected from the group consisting of: an external inducer; a self-activated inducer; and a combination of an external inducer and a self-activated inducer. The external inducer can be selected from the group consisting of: exposure to optical radiation, a change in temperature, a chemical reagent, a viral agent, and combinations thereof. Measuring the electrical parameter can include measuring at least member selected from the group consisting of: a cyclic voltammetry signal for the growth medium; a potentiometry signal for the growth medium; an electrical conductivity of the growth medium; a phase difference between electrical signals measured for the growth medium; and combinations thereof. The electrical signal can be an alternating current (AC) electrical signal. The electrical signal can be a direct current (DC) electrical signal. The control gene can be a single-gene lysis gene, a multi-gene lysis gene, one or more TA module genes, one or more toxin genes, or one or more peptide toxin gene. The control gene can be activated by a target analyte. The target analyte can include a chemical species. The target analyte can include a biochemical molecule.

[0015] In some embodiments, the method can further include determining a relative concentration of the target analyte in the growth medium as a function of time based on the information about the cell population. The information about the cell population can include information about a relative concentration of at least one metabolite generated by the cell population. Measuring the electrical parameter of the growth medium without introducing a redox active species into the growth medium.

[0016] In a second aspect, the disclosure includes a system for monitoring a cell population, the apparatus including: a reservoir configured to contain a cell population, wherein the cell population includes a cell strain including a control gene and a promoter sequence; electrodes configured to contact a growth medium in the reservoir; an electrical source connected to the electrodes and configured to apply an electrical signal to the growth medium; a detector configured to measure an electrical parameter of the growth medium as a function of time; and an electronic processor connected to the detector and configured to receive measurements of the electrical parameter, and to determine information about the cell population based on the measured electrical parameter.

[0017] In some embodiments, the reservoir can be formed in a microfluidic device. The reservoir can include an inlet and an outlet, and wherein the apparatus can include a fluidic device configured to transport the growth medium through the reservoir from the inlet to the outlet. The reservoir can include a recess configured to support a solid growth medium. The electrical parameter can include an electrical admittance of the growth medium. The electrical parameter can include an electrical impedance of the growth medium. The electrical parameter can include a conductance of the growth medium. The electrical parameter can include a resistance of the growth medium. Applying the electrical signal can include applying an electrical potential difference between the two electrodes. The cell population can include one or more different bacterial strains. The cell population can include one or more different yeast strains. The cell population can include one or more different mammalian cell strains. The cell population can include one or more different insect cell strains. The growth medium can be selected from the group consisting of a liquid medium and a solid medium. The solid medium can include a hydrogel. The solid medium can include at least one gel-based material. The cell population in a chamber, and flowing the growth medium through the chamber. The electrical parameter in different portions of the flowing growth medium as a function of time. The control gene can be activated by at least one mechanism selected from the group consisting of: an external inducer; a self-activated inducer; and a combination of an external inducer and a self-activated inducer. The external inducer can be selected from the group consisting of: exposure to optical radiation, a change in temperature, a chemical reagent, a viral agent, and combinations thereof. Measuring the electrical parameter can include measuring at least member selected from the group consisting of: a cyclic voltammetry signal for the growth medium; a potentiometry signal for the growth medium; an electrical conductivity of the growth medium; a phase difference between electrical signals measured for the growth medium; and combinations thereof. The electrical signal can be an alternating current (AC) electrical signal. The electrical signal can be a direct current (DC) electrical signal. The control gene can be a single-gene lysis gene, a multi-gene lysis gene, one or more TA module genes, one or more toxin genes, or one or more peptide toxin gene. The control gene can be a lysis gene. The control gene can be activated by a target analyte. The target analyte can include a chemical species. The target analyte can include a biochemical molecule.

[0018] The system can further include determining a relative concentration of the target analyte in the growth medium as a function of time based on the information about the cell population. The information about the cell population can include information about a relative concentration of at least one metabolite generated by the cell population. The apparatus can further include measuring the electrical parameter of the growth medium without introducing a redox active species into the growth medium.

[0019] In a third aspect, the disclosure includes a method for determining a presence of a target analyte in a medium, the method including: culturing a cell population a medium, wherein: the cell population includes a cell strain including a control gene and a promoter sequence; the promoter sequence can be configured to bind a target analyte and, upon binding the target analyte, to activate the control gene to be expressed; and the control gene can be configured to modulate growth of the cell population; introducing the target analyte into the medium; measuring a change in an electrical parameter of the medium arising from a change in a concentration of a metabolite generated by the cell population following introduction of the target analyte; and identifying a presence of the target analyte in the medium based on the measured change in the electrical parameter.

[0020] In some embodiments, the target analyte can be a protein, oligonucleotide, or chemical compound. The chemical compound can be a metal-containing chemical compound. The electrical parameter can include an electrical admittance of the growth medium. The electrical parameter can include an electrical impedance of the growth medium. The electrical parameter can include a conductance of the growth medium. The electrical parameter can include a resistance of the growth medium. Applying the electrical signal can include applying an electrical potential difference between the two electrodes. The cell population can include one or more different bacterial strains. The cell population can include one or more different yeast strains. The cell population can include one or more different mammalian cell strains. The cell population can include one or more different insect cell strains. The growth medium can be selected from the group consisting of a liquid medium and a solid medium. The solid medium can include a hydrogel. The solid medium can include at least one gel-based material.

[0021] The method can further include maintaining the cell population in a chamber, and flowing the growth medium through the chamber. The method can further include measuring the electrical parameter in different portions of the flowing growth medium as a function of time. The control gene can be activated by at least one mechanism selected from the group consisting of: an external inducer; a self-activated inducer; and a combination of an external inducer and a self-activated inducer. The external inducer can be selected from the group consisting of: exposure to optical radiation, a change in temperature, a chemical reagent, a viral agent, and combinations thereof. Measuring the electrical parameter can include measuring at least member selected from the group consisting of: a cyclic voltammetry signal for the growth medium; a potentiometry signal for the growth medium; an electrical conductivity of the growth medium; a phase difference between electrical signals measured for the growth medium; and combinations thereof. The electrical signal can be an alternating current (AC) electrical signal. The electrical signal can be a direct current (DC) electrical signal. The control gene can be a single-gene lysis gene, a multi-gene lysis gene, one or more TA module genes, one or more toxin genes, or one or more peptide toxin gene. The control gene can be a lysis gene. The control gene can be activated by a target analyte. The target analyte can include a chemical species. The target analyte can include a biochemical molecule.

[0022] The method can further include determining a relative concentration of the target analyte in the growth medium as a function of time based on the information about the cell population. The information about the cell population can include information about a relative concentration of at least one metabolite generated by the cell population. The method can further include measuring the electrical parameter of the growth medium without introducing a redox active species into the growth medium.

[0023] In a fourth aspect, the disclosure includes a device for detecting a target analyte, the device including: a substrate including a reservoir configured to contain a cell population, wherein: the cell population includes a cell strain including a control gene and a promoter sequence; the promoter sequence can be configured to bind a target analyte and, upon binding the target analyte, to activate the control gene to be expressed; and the control gene can be configured to modulate growth of the cell population; an inlet formed on or in the substrate and connected to the reservoir; a fluid delivery mechanism connected to the inlet and configured to introduce the target analyte into the reservoir; electrodes configured to contact a medium supporting the cell population in the reservoir; an electrical source connected to the electrodes and configured to apply an electrical signal to the medium; a detector configured to measure an electrical parameter of the medium arising from a change in a concentration of a metabolite generated by the cell population following introduction of the target analyte; and an electronic processor connected to the detector and configured to receive measurements of the electrical parameter, and to identify the target analyte based on the measured change in the electrical parameter.

[0024] In some embodiments, the electrical parameter can include an electrical admittance of the growth medium. The electrical parameter can include an electrical impedance of the growth medium. The electrical parameter can include a conductance of the growth medium. The electrical parameter can include a resistance of the growth medium. The electrical signal can include applying an electrical potential difference between the two electrodes. The cell population can include one or more different bacterial strains. The cell population can include one or more different yeast strains. The cell population can include one or more different mammalian cell strains. The cell population can include one or more different insect cell strains. The growth medium can be selected from the group consisting of a liquid medium and a solid medium. The solid medium can include a hydrogel. The solid medium can include at least one gel-based material.

[0025] The device can further include maintaining the cell population in a chamber, and flowing the growth medium through the chamber. The device can further include measuring the electrical parameter in different portions of the flowing growth medium as a function of time. The control gene can be activated by at least one mechanism selected from the group consisting of: an external inducer; a self-activated inducer; and a combination of an external inducer and a self-activated inducer. The external inducer can be selected from the group consisting of: exposure to optical radiation, a change in temperature, a chemical reagent, a viral agent, and combinations thereof. Measuring the electrical parameter can include measuring at least member selected from the group consisting of a cyclic voltammetry signal for the growth medium; a potentiometry signal for the

growth medium; an electrical conductivity of the growth medium; a phase difference between electrical signals measured for the growth medium; and combinations thereof. The electrical signal can be an alternating current (AC) electrical signal. The electrical signal can be a direct current (DC) electrical signal. The control gene can be a single-gene lysis gene, a multi-gene lysis gene, one or more TA module genes, one or more toxin genes, or one or more peptide toxin gene. The control gene can be a lysis gene. The control gene can be activated by a target analyte. The target analyte can include a chemical species. The target analyte can include a biochemical molecule.

[0026] The device can further include determining a relative concentration of the target analyte in the growth medium as a function of time based on the information about the cell population. The information about the cell population can include information about a relative concentration of at least one metabolite generated by the cell population. The device can further include measuring the electrical parameter of the growth medium without introducing a redox active species into the growth medium. The reservoir can be formed in a microfluidic device. The reservoir can include an inlet and an outlet, and wherein the apparatus can include a fluidic device configured to transport the growth medium through the reservoir from the inlet to the outlet. The reservoir can include a recess configured to support a solid growth medium.

[0027] Embodiments of the methods, systems, and devices described herein can include any one or more of the foregoing features, and any of the other features described, and can generally include combinations of multiple features, including features that are individually described in connection with different embodiments, in any combination except as expressly stated otherwise herein.

[0028] The term "population dynamics" as used herein refers to the temporal evolution of a cellular population as a dynamical system, and includes increases, decreases, and steady-state population numbers and the biological and environmental processes driving changes in population numbers such as, but not limited to, cellular replication and lysis rates.

[0029] The term "multiplex" as used herein refers a type of assay in which information about multiple analytes is obtained in the assay. The information about the multiple analytes can be obtained simultaneously, sequentially, or a combination of simultaneously and sequentially (i.e., certain analyte information can be obtained simultaneously, while information about other analytes is obtained sequentially). For example, a multiplex assay can include the detection or quantification of two or more (e.g., three or more, four or more, five or more, six or more, eight or more, ten or more, 15 or more, 20 or more, or even more) analytes in a single experiment.

[0030] The term "control gene" as used herein refers to a sequence of nucleotides which, when expressed through transcription and translation, modifies the metabolic process of the cellular populations. The metabolic process can be increased or decreased. For example, a control gene, when expressed, can produce a product which causes the host cell to lyse, thereby ceasing metabolic output.

[0031] The terms "promoter" and "promoter sequence" as used herein are synonymous, and refer to a sequence of nucleotides that directs the level of transcription of a given gene, e.g., the control gene. A promoter may be induced in response to changes in an inducer agent (e.g., in an inducer agent concentration) which activates transcription of the control gene.

[0032] The term "cellular integrated circuit" as used herein refers to a device which integrates electrical circuitry components measuring an electric parameter of a solution containing a cellular population capable of responding to the concentration of an inducer agent. For example, a cellular integrated circuit can include electrodes, a cellular population, and a growth medium providing nutrients to the cellular population.

[0033] The term "inducer agent" as used herein refers to a stimulus which functions to interact with the promoter sequence and activate transcription of the control gene. Inducer agents can include, by way of example, chemical inducer agents, temperature-sensitive inducer agents, and light-sensitive inducer agents, including intermediary mediation elements such as protein co-factors.

[0034] The term "redox active species" as used herein refers to a chemical species which undergoes a reduction reaction with a metabolite product in solution thereby gaining one or more electrons. The redox active species then acts as an intermediary charge carrier for coupled electronic systems and propagates electronic signals facilitating the indirect detection of the metabolite product, wherein increases or decreases in metabolite product concentrations are coupled to the increases or decreases in the concentration of redox active species, respectively.

[0035] The term "genetic circuit" as used herein refers to one or more cellular populations including genetic modification which facilitate the cellular population performing logical functions mimicking those observed in electronic circuits. For example, a combination of promoter sequences and control genes can be designed as a genetic circuit to inducing a specific cellular process, protein production, or adding a measurable element to the host cell or solution in response to a stimulus.

[0036] Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this disclosure belongs. Although methods and materials similar or equivalent to those described herein can be used in the practice or testing of the subject matter herein, suitable methods and materials are described below. All publications, patent applications, patents, and other references mentioned herein are incorporated by reference in their entirety. In case of conflict, the present specification, including definitions, will control. In addition, the materials, methods, and examples are illustrative only and not intended to be limiting.

[0037] The details of one or more embodiments are set forth in the accompanying drawings and the description below. Other features and advantages will be apparent from the description, drawings, and claims.

BRIEF DESCRIPTION OF THE DRAWINGS

[0038] FIG. **1** is a schematic illustration of an array of microwells in a cellular integrated circuit.

[0039] FIGS. **2**A through **2**C are schematic illustrations of a microwell in a cellular integrated circuit.

[0040] FIG. **3**A is a schematic electronic diagram of a cellular circuit and the corresponding electrical circuit.

[0041] FIGS. **3**B and **3**C are schematic illustrations of a microwell including a cellular strain measuring a response to an analyte.
[0042] FIG. **4**A is a line chart comparing voltammetry measurements of a cellular population versus media only.
[0043] FIG. **4**B is a three-axis line chart comparing impedance and signal phase versus frequency for a cellular population.
[0044] FIGS. **4**C and **4**D are line charts comparing impedance versus frequency for a cellular population.
[0045] FIG. **4**E is a schematic diagram and a bar chart histogram showing the impedance contributions from media, a solution of concentrated bacteria, and depleted medium.
[0046] FIGS. **5**A through **5**C are three-axis line chart comparing turbidity and admittance versus time for cellular populations.
[0047] FIG. **6**A is a schematic diagram showing the process of using engineered cellular populations to detect the presence of an inducer agent.
[0048] FIG. **6**B is a line chart comparing admittance versus time for a cellular population in the presence of IPTG.
[0049] FIG. **6**C is a schematic diagram comparing the genetic circuit to an electrical circuit.
[0050] FIG. **6**D is a schematic diagram showing the components of a micro-fluidic chip including four cellular integrated circuits.
[0051] FIG. **7**A is a schematic diagram and a picture of an experimental setup to measure the electrical parameter and turbidity response of a cellular population to an inducer agent.
[0052] FIG. **7**B is a schematic diagram showing the genetic circuit for detecting the presence of arsenic.
[0053] FIGS. **7**C and **7**D are three-axis line chart comparing turbidity and admittance versus time for arsenic- and copper-sensing cellular populations, respectively.
[0054] FIG. **7**E is a line chart comparting current to potential in the absence and presence of arsenic and copper in solutions including arsenic- and copper-sensing cellular populations.
[0055] FIG. **7**F is a heat map diagram of selectivity to arsenic versus copper in arsenic- and copper-sensing cellular populations.
[0056] FIG. **8**A is an image of a macro-chemostat system showing a pumping system under continuous mixing conditions.
[0057] FIG. **8**B is an image of certain components of the system of FIG. **8**A including a culture tube.
[0058] FIG. **8**C is a schematic diagram showing a customized turbidity sensor.
[0059] FIG. **8**D is a three-axis line chart comparing optical density and transmittance as a function of time.
[0060] FIGS. **9**A and **9**B are three-axis line chart comparing turbidity and admittance as a function of time for arsenic- and copper-sensing cellular populations, respectively.
[0061] FIG. **10**A is a schematic diagram showing an oscillatory genetic circuit.
[0062] FIG. **10**B is a three-axis line chart comparing turbidity and admittance as a function of time for the oscillatory genetic circuit of FIG. **10**A.
[0063] FIG. **11**A is a schematic diagram of a micro-fluidic chip in a cellular circuit and a single microwell including the reference and working electrodes.
[0064] FIG. **11**B is schematic diagram showing multiplexed genetic circuits.
[0065] FIG. **11**C is a set of four images of a microwell during exposure of a cellular population in the microwell to an inducer agent.
[0066] FIG. **11**D is a three-axis line chart comparing turbidity and admittance as a function of time for the four images of FIG. **11**C.
[0067] FIG. **11**E is a set of two images of the microwell and chambers before and after exposure to an inducer agent corresponding to images (i) and (ii) of FIG. **11**C.
[0068] FIG. **11**F is a three-axis line chart comparing turbidity and admittance as a function of time for the two images of FIG. **11**E.
[0069] FIG. **12**A is a schematic diagram showing electrical contacts of a cellular integrated circuit on a micro-fluidic chip.
[0070] FIG. **12**B is an image of a micro-fluidic chip including eight cellular integrated circuits.
[0071] FIG. **12**C is a line chart comparing electrical signal phase to frequency for a cellular integrated circuit.
[0072] FIG. **12**D is a line chart showing admittance as a function of time for a NaCl-sensing cellular integrated circuit.
[0073] FIG. **12**E is an image of a system for cellular integrated circuit measurements including a microscope and an impedance analyzer.
[0074] FIG. **13** is a flow-chart showing a set of example steps for detecting the presence of an inducer agent using a cellular integrated circuit.
[0075] Like reference symbols in the various drawings indicate like elements.

DETAILED DESCRIPTION

[0076] A wide variety of biological molecules have been applied to the detection and quantification of analytes ex vivo using various detection methodologies. For example, biological molecules that selectively bind certain analytes and generate signals and/or can be isolated from complex environments are routinely used for such measurements. Such biological molecules have even be interfaced with microelectronic devices to yield compact, low-power sensing technologies that can be deployed in a variety of environments. Among other factors, devices based on these biological molecules can be limited to applications where the biological molecules do not saturate (i.e., the amount of analyte does not exceed the available concentration of biological molecules that bind them). Further, making quantitative measurements may involve interfacing the biological molecules with multiple components, yielding relatively complex devices and systems.
[0077] As an alternative, in synthetic biology, detection methods based on genetic expression can be used to characterize genetic circuit behavior and detect target analytes. Detectable signals generated by cellular populations have conventionally been generated through the addition of one or more redox active species. Redox active species are reagents that are added to a solution or suspension containing a cellular population. The redox state of the redox active species is modified by a cellular metabolite generated by the cellular population, resulting in a change in the electrical properties of the growth medium containing the cellular population. The change in electrical properties due to the

perturbation of the redox state of the redox active species is then detected.

[0078] However, it has been discovered that metabolites and other species generated by cellular populations also directly affect the electrical properties of the solution, suspension, or other medium containing the cellular populations. As a result, concentrations of the metabolites and other species can be determined by measuring one or more electrical parameters of the medium directly, without adding redox active species into the medium.

[0079] To make such measurements, cellular populations are interfaced with electronic components and devices to form cellular integrated circuits. The interface between a cellular population and an electrical device can be implemented in various ways. For example, as will be described in further detail below, the interface is implemented by immersion of a pair of electrodes (e.g., containing a relatively inert, conductive material such as gold and/or platinum) in medium containing the active cellular population. By applying an electrical potential between the electrodes, one or more electrical parameters associated with the medium can be measured. Changes in the one or more electrical parameters as a function of time reflect changes in the cellular population. If the cellular population is responsive to a particular stimulus (e.g., a target analyte), then changes in the one or more electrical parameters can correlate with, and provide information about, the presence and concentration of the target analyte as a function of time. A variety of different electrical parameters of the medium can be measured, including (but not limited to), electrical admittance, impedance, conductivity, resistance, and relative phases between different parameter values.

[0080] Disclosed herein are cellular integrated circuits in which a cellular population is integrated with electrical devices and/or components to form the cellular integrated circuits. Typically, the cellular populations include one or more genetically engineered bacterial strains, although the cellular populations may also include only naturally occurring bacterial cell strains. As the population of cells reproduces, the production and concentration of charged metabolic products increases correspondingly in the medium supporting the population. Conductive electrodes are placed in contact with the solution containing the cellular population and an electrical source applies a potential between the electrodes. A detector connected to the electrodes determines a value of at least one electrical parameter of the medium containing the cellular population. As concentration of charged metabolic products increases, the value of the at least one electrical parameter varies correspondingly. Measured values of the one or more electrical parameters provide an indirect measurement of the cellular population.

[0081] In some of the methods, devices, and systems described herein, cellular populations include one or more bacterial strains that are specifically engineered to respond to a target analyte, a signal, or some other stimulus of interest. In response to the stimulus, the engineered bacteria may multiply, undergo cell death, or provide another type of response. During multiplication, production of certain cellular metabolic products increases and therefore the concentrations of those products in the medium increases as a function of time. Conversely, during population reduction via cell death, production of certain cellular metabolic products decreases, and the concentrations of those products in the medium decreases as a function of time. As noted above, it has been discovered that the perturbative effect of the changing concentrations of cellular metabolic products on the medium containing the cellular population is manifest as changes in one or more electrical parameters of the medium. These changes can be measured due to the interface between the cellular populations and electrical devices such as a detector, and the measurement information is used to determine information about the presence, absence, and magnitude of the stimulus. Such measurements can be performed to detect the perturbative effect of the metabolic products directly, without addition of a redox active species.

[0082] FIG. **1** is a schematic illustration showing a microfluidic chip **100** having four cellular integrated circuits **120***a-d* integrated on a substrate, which can be a wafer **110**. The wafer **110** can be manufactured to include cellular integrated circuits **120** in a range from one to 24 (e.g., from two to 20, from 4 to 16, or from 8 to 12). Increasing the number of cellular integrated circuits **120** on a wafer **110** increases the degree to which the cellular integrated circuits **120***a-d* can be multiplexed, e.g., run in parallel. The cellular integrated circuits **120***a-d* are shown connected to a single channel **130**, though in alternative embodiments, the cellular integrated circuits **120***a-d* can be connected to one or more channels in different arrangements. For example, in some embodiments, each cellular integrated circuit can be connected to a different channel.

[0083] Each cellular integrated circuit **120***a-d* includes a reservoir for containing a cell population, implemented as a micro-well **112***a-d*, and an electrode pair **124***a-d* in electrical communication with an electrical source **140** which provides an electrical signal **142***a-d*. Each micro-well **112** includes two chambers **114***a-h* in fluid connection with a channel **130**. A fluid source **150** provides a fluid to the channel **130** which transmits the fluid to the connecting chambers **114***a-h* of each micro-well **112***a-d*. The upstream chamber of each micro-well, e.g., the chamber nearest the fluid source **150**, provides a fluid inlet into the connected microwell **112***a-d* while the chambers furthest from the fluid source **150** provides an outlet. Fluid flowing into the upstream inlet chamber displaces fluid within the microwell, which exits through the downstream chamber, thereby exchanging fluid volumes within the micro-wells **112***a-d*. The micro-wells **112***a-d* function as growth chambers for cellular populations that are present. The cellular populations can be seeded into the micro-wells 112a-dbefore the channel **130** is connected to the fluid source **150**, or the fluid source **150** can provide a fluid containing one or more cellular populations to seed the micro-wells **112***a-d*.

[0084] The dimensions of the channel **130** are defined by the overall dimensions of the wafer **110** and orientations of the micro-wells **112***a-d*, but in general has a width in a range from 50 μm to 500 μm, for example 200 μm. The length of the channel **130** extends across portion of the wafer **110** and fluidly connects all the chambers **114***a-h* of the micro-wells **112***a-d*. In general the channel **130** length is in a range from 1000 μm to 5000 μm, for example, 2000 μm. Longer channel **130** dimensions can provide fluid to higher numbers of cellular integrated circuits **120** present on the micro-fluidic chip **100**.

[0085] The micro-fluidic chip **100** includes a substantially transparent (e.g., optical transmission > 95%) wafer **110** onto which the cellular integrated circuits **120** are constructed. The wafer **110** is a rigid unitary body which provides a stable platform for the construction of multiple cel-

lular integrated circuits **120**. The wafer **110** is composed of a chemically- and biologically inert solid medium material such as glass, quartz, or rigid polymer (e.g., polymethyl methacrylate (PMMA), or polydimethylsiloxane (PDMS)) which supports the construction of the cellular integrated circuits **120** and electrode pairs **124** onto the wafer **110**. In some embodiments, the wafer **110** is composed, partially or completely, of a gel-based substrate, e.g., a hydrogel, and other structures including micro-wells **112***a-d*, chambers **114***a-h*, and channel **130** can be formed in the gel-based material.

[0086] The cellular integrated circuits **120** are formed on or in wafer **110** through one or more manufacturing methods. As an example, the wafer **110** can be additively manufactured (e.g., 3D printed, or resin cast/molded), or subtraction manufactured (e.g., lithography) to include the micro-wells **112***a-d* and channel **130**. For example, described further herein, the wafer **110** can be resin molded from a master wafer (e.g., silicon or glass) using polydimethylsiloxane (PDMS). The master wafer can be photo-lithographically etched to create a negative master of the molded features of the wafer **110**, including the channel **130**, chambers **114***a-h*, and micro-wells **112***a-d*. A liquid resin (e.g., PDMS) is flowed over the master, cured, and removed to create the wafer **110**. The dimensions of the molded features are etched into the master wafer which sets the dimensions for wafers **110** constructed from the master. The molded feature dimensions can be the same across the micro-wells **112***a-d* of a micro-fluidic chip **100**, or the micro-wells **112***a-d* can have different dimensions between elements.

[0087] Each cellular integrated circuit **120** includes an electrode pair **124***a-d* (e.g., two electrodes). For examples, each cellular integrated **120***a*, **120***b*, **120***c*, and **120***d*, includes electrode pair **124***a*, **124***b*, **124***c*, and **124***d*, respectively. In some embodiments, to facilitate fabrication, the electrode pairs **124** are constructed onto the upper surface of the wafer **110** such that a first end of each of the electrode pairs **124** is positioned within one of the micro-wells **112***a-d*. In some embodiments, the electrode pairs **124** are deposited using masked lithography. As an example, the wafer **110** upper surface can be coated in one or more layers of a conductive material via a deposition method, such as spin coating, sputter deposition, or vapor deposition. The conductive material which composes the electrode pairs **124** can be a pure or alloyed material consisting of gold, silver, platinum, chromium, or other conductive, biosafe material. A portion of the deposited layers are removed whereby the electrode pairs **124** remain on the wafer **110** upper surface. In some embodiments, the portion of the deposited layers can be removed via lithographic surface patterning, electrochemical etching, or dry etching.

[0088] As an example, a photoresist layer is deposited on top of the deposited layers and exposed to a masked light pattern corresponding with the shape of the electrode pairs **124** (e.g., negative photoresist etching) to cure the unmasked photoresist layer. Alternatively, the masked light pattern corresponds with the portion of the deposited layer corresponding with the area surrounding the electrode pairs **124** (e.g., positive photoresist etching). The wafer **110** is then exposed to a developer (e.g., solvent) to remove the un-cured photoresist layer and expose the deposited conductive layer beneath. The exposed conductive layer is chemically etched to remove unwanted material from the layer and leave the electrode pairs **124**.

[0089] The electrode pairs **124** provide an electrical connection for each of the cellular integrated circuits **120** to the electrical source **140**. The electrical source **140** includes a power source (e.g., a battery), or alternatively, a means of connecting to a power source (e.g., a wall outlet) and circuitry to produce one or more electrical signals **142** to electrically connected cellular integrated circuits **120**.

[0090] In general, the electrical source **140** applies an electrical potential difference between the electrodes of each cellular integrated circuit. In some embodiments, the electrical source **140** is an electronic hardware device that is configured to operate as a potentiostat for each connected electrode pair **124**, such that the electrical source **140** provides a constant or variable electrical potential to one electrode of an electrode pair **124***a-d* which acts as a working electrode (WE). In certain embodiments, the electrical source **140** maintains the second electrode at a constant electrical potential such as, for example, a neutral (or grounded) electrical potential. As such, the second electrode functions as a reference/counter electrode (R/CE). In some embodiments, the electrical source **140** adjusts the current supplied to the WE to maintain the constant potential between the electrode pairs **124**.

[0091] In certain embodiments, the electrical source **140** additionally includes circuitry to monitor one or more electrical parameters of the cellular integrated circuits **120**. For example, the electrical source **140** determines a conductivity parameter of the fluid within the micro-wells **112***a-d* by monitoring the current needed to maintain the constant potential and calculating a conductivity parameter of the fluid in the micro-wells **112***a-d* providing the electrical connection between the electrode pairs **124**. In some embodiments, the electrical source **140** includes circuitry which measures an electrical potential difference (e.g., a potentiometer), electrical current (e.g., ammeter), electrical impedance, electrical admittance, electrical conductance, electrical resistance, a phase difference between two electrical signals or between an electrical signal and a reference signal, and combinations of any of the foregoing.

[0092] Without wishing to be bound by theory, the impedance of an electric circuit is related to an effective resistance of the circuit to alternating current (AC) flow, arising from the combined effects of ohmic resistance and reactance, and is the measure of the opposition that a circuit presents to a current when a voltage is applied. When a cellular population is in a homeostatic growth phase within the micro-wells **112***a-d*, the concentration of metabolic products remains constant based on the metabolic products produced by the cellular population and the fluid exchanged with (e.g., flowing in and out of) the chambers **114***a-h*. The electrical source **140** can apply constant current to the R/CE to maintain a constant potential at the WE. Changes in the cellular population change the concentration of metabolic products within the micro-wells **112***a-d* and thereby the current needed to maintain the constant WE potential. The electrical source **140** can calculate the conductivity of growth medium (e.g., the fluid flowing in and out of the chambers) and determine any one or more of the parameters described above based upon the change in the current.

[0093] For example, as shown in FIG. **1**, the electrical source **140** applies electrical signal **142***a* to electrode pair **124***a*, electrical signal **142***b* to electrode pair **124***b*, electrical signal **142***c* to electrode pair **124***c*, and electrical signal **142***d* to electrode pair **124***d*. In some embodiments, the electrical

signals **142** are common (e.g., the same) between electrode pairs **124**, and in alternative embodiments, the electrical source **140** applies more than one electrical signals **142** to the electrode pairs **124**. By way of example, the electrical source **140** applies a first electrical signal **142***a* to integrated circuit **120***a*, and a second electrical signals **142** to integrated circuit **120***b*, **120***c*, and **120***d*. As described above the electrical signals **142** include applying a moderated current applied to the R/CE to maintain a constant potential at the WE. In alternative embodiments, the electrical signals **142** can include a constant current signal, a varying current signal, a varying voltage signal, or combinations thereof.

[0094] A fluid source **150** is in fluid connection with a first end of the channel **130**. The fluid source **150** provides a fluid at a positive pressure to the channel **130** and connected micro-wells **112***a-d*. The fluid is a medium containing chemicals, and/or nutrients for sustaining the growth and activity of the cellular populations within the micro-wells **112***a-d*, such as culture media, minimal media, selective media, differential media, transport media, or indicator media. In some embodiments, the fluid can optionally include a buffer, solution, suspension, or mixture. For example, the fluid can be an LB media (Miller) and can be mixed with additional adjuncts to support the growth of the cellular populations. Alternative examples of media include general culture media, such as M9 medium, EZ Rich, or MacConkey medium. The fluid source **150** provides the fluid at a flow rate to the first end of the channel **130** by a positive pressure mechanism such as a pump, or gravity feed. For example, in some embodiments, the fluid source **150** is a peristaltic pump, such as a P625/10K.143 peristaltic pump manufactured by Instech. A removable lid provides the inlet and outlet ports and fluid connections for the wafer **110**.

[0095] The channel **130** and connected micro-wells **112***a-d* have low respective volumes and the flow rate through the channel **130** is correspondingly low. The channel **130** receives fluid from the fluid source **150** at a rate in a range from 1 μL/min to 75 μL/min (e.g., 5 μL/min to 60 μL/min, 10 μL/min to 40 μL/min, or 20 μL/min to 30 μL/min). In some embodiments, the channel **130** receives fluid from the fluid source **150** at a flow rate of 25 μL/min.

[0096] The fluid flows from the first end through the length of the channel **130** and exchanges fluid with the connected micro-wells **112***a-d*. The fluid flowing through the chambers **114***a-h* and being exchanged with the fluid within the micro-wells **112***a-d* provides growth materials for the cellular populations therein. Cellular waste products are removed from the micro-wells **112***a-d* during the exchange which flows through the chambers **114***a-h* and into the channel **130**. The pressure and fluid flow provided by the fluid source **150** drive the excess material to the waste vessel **152**.

[0097] Referring now to FIGS. **2**A and **2**B, top-down and transverse cross-section views are shown, respectively, of a portion of the wafer **110** including micro-well **112***a*, chamber **114***a*, chamber **114***b*, and the portion of channel **130** in fluid connection with chambers **114***a* and **114***b*. The micro-wells **112***a-d* are approximately cylindrical with a diameter, d, of 600 μm and a height, h, of 30 μm. The diameter, d, can be in a range from 100 μm to 1000 μm (e.g., 200 μm, 250 μm, 300 μm, 500 μm, 700 μm, or 900 μm). Smaller diameters facilitate lower pitch distances and higher micro-well **112***a-d* density on a wafer **110**. The micro-well **112***a* height, h, can be in a range from 5 μm to 80 μm (e.g., 10 μm to 60 μm, 20 μm to 40 μm, or 15 μm to 75 μm). Larger height values enable higher micro-well **112***a* volumes and thereby higher cellular population counts and higher metabolic product concentrations, both of which can increase the steady state average electrical parameter value, and changes therein, as measured by the electrode pairs **124**.

[0098] The chambers **114***a* and **114***b* have a height of 4 μm, a maximum width of 100 μm and are connected to the micro-well **112***a* and channel **130** by connection conduits having a width of 10 μm. The chambers **114***a-h* extend vertically into the wafer **110** by a smaller distance than the micro-wells **112***a-d* (e.g., 4 μm > 30 μm). This allows the majority of the cellular populations to remain within the micro-wells **112***a-d* when fluid is flowing from the micro-wells **112***a-d* into the chambers **114***a-h* and channel **130**.

[0099] FIG. **2**C shows the top-down view of FIG. **2**A and includes the first ends **125***a* of the electrode pair **124***a*. The first ends **125***a* are oriented above and extend across the micro-well **112** by a portion of the first ends **125***a* length. The first ends **125***a* can be of any orientation, length, or width, such that a separation length, L, is maintained. As non-limiting examples, the first ends **125***a* can extend radially inward as depicted, in parallel, perpendicular, or colinear into the micro-wells **112***a-d*. The separation length, L, can be in a range from 5 μm to 1000 μm, and the distance, L, is generally less than the diameter, d. In some embodiments, the separation length, L, can be between 100 μm to 800 μm, 200 μm to 600 μm, or 400 μm to 500 μm.

[0100] In some embodiments, the cellular population sustained within the micro-fluidic chip **100** is a microbial population. In some embodiments, the cellular population is a prokaryotic, eukaryotic, or archaic cellular population. In embodiments in which the cellular population is prokaryotic, the cellular population can include one or more of a *Salmonella enterica*, or *Escherichia coli* cellular population. As specific non-limiting examples, the cellular population can be a *Salmonella enterica* subsp. *enterica serovar typhimurium*, SL1344 strain ELH1301 or a *Escherichia coli* MG1655. In some examples, the cellular population can be a strain of *E. coli*, *Salmonella Typhimurium*, *Salmonella typhi*, *Yersinia enterocolitica*, *Serratia* (e.g., *Serratia marcescens*, *Serratia aquatilis*, *Serratia entomophila*, or *Serratia symbiotica*), *Bacillus Subtilis*, *Pseudomonas Aeruginosa*, *Staphylococcus*, *Bacteroides*, *Vibrio* (e.g., *Vibrio parahaemolyticus*, *Vibrio harveyi*, *Vibrio natriegens*, *Vibrio alginolyticus*), or *Actinobacteria*.

[0101] In certain embodiments, the cellular population includes one or more modifications to the cellular genome. The modifications can be made through any genomic modification method known in the art and can include electroporation and transformation. Briefly, electroporation, or electropermeabilization, is a microbiology technique in which an electrical field is applied to cells in order to increase the permeability of the cell membrane, allowing chemicals, reagents, or DNA to be introduced into the cell. A plasmid carrying one or more modification genes is introduced to the cellular population which is then exposed to electroporation. The increased permeability of the cellular population cell membranes facilitates the introduction of the plasmid into the cellular population lumen where the cellular population transforms the plasmid DNA and introduces the modification genes into the cellular genome. Examples of methods for performing electroporation/electropermeabilization are described, for example, in Electropermeabilization, a physical method for the delivery of ther-

apeutic molecules into cells, by Marie-Pierre Rols, *BBA-Biomembranes*, Volume 1758, Issue 3, March 2006, the entire contents of which are incorporated by reference herein.

[0102] In certain embodiments, the cellular population includes at least two genetic modifications: a control gene and a promoter sequence. In some embodiments, the cellular population can include more than one control gene and/or more than one promoter sequence. The promoter sequence is a non-constitutive, e.g., inducible, promoter sequence which is active under certain conditions, such as active in the presence of an inducer agent. The inducer agent can be a target analyte or the inducer agent can interact with the target analyte present in the fluid supplied from the fluid source **150**. In some embodiments, the promoter sequence is a positive inducible promoter sequence which promotes transcription of the control gene in the presence of the inducer agent. In alternative embodiments, the promoter sequence is a negative inducible promoter sequence which promotes transcription of the control gene in to the absence of the inducer agent. As an example, the cellular population genome includes a lac promoter which promotes transcription of the control gene when the isopropyl β-D-1-thiogalactopyranoside (IPTG) inducer agent is present. Alternative examples of the promoter sequences include pArs, pMer, pCop, pLux, pRpa, pLas, pRhl, pLac, pBAD, or pTet.

[0103] Examples of inducer agent include a chemical inducer agent, a temperature-sensitive inducer agent, or a light-sensitive inducer agent, such as IPTG, tetracycline, Doxycycline, Arabinose, or AHL. Alternative examples of inducer agents can include heavy metals or heavy-metal-containing molecules (e.g., arsenic, copper, zinc, mercury, etc.), bacterial signaling or quorum-sensing molecules (e.g., autoinducing peptides, or N-acyl homoserine lactones), bacteriophages (e.g., T4, Mu, P1, P2, λ, T5, HK97, N15, T7, T3, Φ29, P22, MS2, Qβ, Acidianus filamentous virus 1, Sulfolobus islandicus rod-shaped virus 1, Φ6, ΦX174, M13, PM2, or FLiP), sugars, metabolites, a light signal, and a temperature (e.g., an elevated, or a decreased temperature).

[0104] In general, a control gene can be any genetic sequence that controls the growth and/or metabolic activity of the cellular population. In some embodiments, the control gene may be multiple genes, gene fragments, genetic sequences, and associated regulatory species. Examples of control genes include, but are not limited to, single-gene lysis genes such as phage phiX174 lysis gene, E, which causes lysis of the bacterium *Escherichia coli*. Alternative lysis genes include multi-gene lysis genes such as λ S and the S107 genes, or the ϕ21 S^{21} and the $S^{21}71$ genes. Cellular population genomes including a lac promoter that transcribes gene E after IPTG induction, causes lysis of the host. Therefore, presence of the IPTG inducer agent causes lysis of the host which ceases the production of charged metabolic products.

[0105] Alternative examples of control genes can include one or more genes from bacterial toxin-antitoxin (TA) modules. A TA module is a set of two or more closely linked genes that together encode both a "toxin" protein and a corresponding "antitoxin", which can perform cell functions such as responding to stresses, causing cell cycle arrest and bringing about programmed cell death. For example, mazEF, a TA locus found in *E. coli* induces programmed cell death in response to starvation. Other TA modules (e.g., Type I TA modules) are small, hydrophobic proteins which can confer toxicity by damaging cell membranes. Other non-limiting examples of TA modules include Type II, or Type III TA modules; symR/symE, tisB/istR-1, phd-doc, or ibs/sib module of *E. coli*; fstI/rnaII, higB/higA, hok/sok, vapB/vapC, ω-ε-ζ, paaR-paaA-parE, pasA/pasB/pasC, toxI/toxN, or ccdA/ccdB. Expression of TA module control genes can regulate the transcription in general, reducing cellular metabolism by reducing the overall expression of the host cell genome.

[0106] Examples of control genes can also include genes that express compounds which are generally toxic to bacteria, such as peptide toxins. For example, melittin is a toxin which is a component of bee venom. Increased expression of peptide toxins can produce a quorum sensing response in the cellular population triggering programmed cell death or apoptosis. Other examples of peptide toxins include mastoparan-L, APETx2, BDS-I and II, APETx1, ZorO, epidermal growth factor (EGF)-like toxin (gigantoxin I), or D(KLAKLAK)2, α-KTx peptides, cysteine-stabilized α/β (CSα/β) peptide toxins, or synthetic peptide toxins. Yet more non-limiting examples can include one or more bacterial genes producing proteinaceous toxins, e.g., bisin, halocin, lactococcin, mutacin, pediocin, pyocin, sakacin, vibriocin, nisin, microcins, colicins, tailocins, Class I, Class IIa/b/c, and/or Class III bacteriocins.

[0107] FIG. **3**A depicts a genetic circuit **300** having an inducible promoter sequence **302** in sequence with a control gene **304**. In the genetic circuit **300**, the promoter sequence **302** is a positive inducible promoter which when the inducer agent is present in the cellular population environment, the promoter sequence **302** allows the control gene **304** to be expressed. When the control gene **304** is expressed, the host cell undergoes a change in its growth cycle, metabolic activity, or another disruption to its homeostatic state. For example, in some embodiments, the host cell undergoes lysis, the production of charged metabolic products ceases, and a conductivity parameter of the fluid in the micro-well **112** changes, reflecting the reduction in the production of charged metabolic products. As such, the change in the measured conductivity parameter is related to the presence of, and in certain embodiments the concentration of, the inducer agent. The genetic circuit **300** of FIG. **3**A can be considered a 'resistive' genetic circuit (left) with the electrical conductivity parameter varying based upon the presence or absence of the inducer agent.

[0108] FIGS. **3**B and **3**C schematically illustrate a micro-well **312** including a cellular population **310** in a micro-well **312**. The cellular population **310** includes the genetic circuit **300** of FIG. **3**A. When the fluid flowing through the channel **330** does not include the inducer agent corresponding to the promoter sequence **302**, the population does not undergo an induced change (e.g., communal lysis) and continues to produce charged metabolic products and maintains the measured electrical conductivity parameter value, as shown on the right. The right side of FIG. **3**B shows a line chart comparing an electrical conductivity parameter on the y-axis, e.g., an admittance, as measured by the electrode pair **324** to time on the x-axis. The line chart includes a conductivity signal **340** showing the admittance profile over time. The cellular population **310** continues in a homeostatic growth cycle producing a constant amount of charged metabolic products and the conductivity signal **340** remains approxi-

mately constant, e.g., within a noise level of an average value.

[0109] As shown in FIG. **3**C, the fluid in the channel **330** includes a quantity of inducer agent **332** such as inducer agents described herein. The inducer agent **332** flows into the micro-well **312** and the cellular population **310** within is exposed to the inducer agent **332**. The inducer agent **332** interacts with the promoter sequence **302** which allows the control gene **304** to be expressed and cause the host cellular population **310** to undergo lysis responsive to the inducer agent **332**. As the cellular population **310** lyses and ceases to produce charged metabolic products, the concentration of the metabolic products drops as the fluid within the micro-well **312** is exchanged with the fluid supplied by the channel **330**, e.g., fluid supplied from the fluid source **150**. As the concentration of metabolic products reduces, the measured electrical conductivity parameter value, e.g., conductivity signal **340**, reduces correspondingly.

[0110] The genetic circuit **300** of FIGS. **3**A through **3**C is a resistive circuit that provides an output change in the conductivity parameter and produces conductivity signal **340**. In general, the micro-well **312**, which can correspond to the micro-wells **112***a-d*, of FIG. **1**, can include multiple cellular populations **310** each including a different genetic circuit **300** based on upon different combinations of one or more promoter sequence **302**, and one or more control gene **304**. Combinations of promoter sequences **302** and control genes **304** can respond to one or more inducer agents **332** present in the channel **330** fluid and determine the output conductivity signal **340**. Combinations of promoter sequences **302** and control genes **304** can produce conductivity signals **340** as constant, linear, polynomial, or oscillatory signals.

[0111] For example, if the concentration of the inducer agent **332** in the fluid supplied by the fluid source **150** changes with respect to time, the cellular population can respond accordingly either promoting or inhibiting the control gene based upon the inducer agent **332** concentration. The output conductivity signal **340** reflects the change in the inducer agent **332** concentration by increasing or decreasing correspondingly.

[0112] FIG. **13** schematically illustrates an example method for using a micro-fluidic chip **100** including cellular integrated circuits **120** to detect the presence of a target analyte, and/or an inducer agent. A cellular strain is engineered to include one or more promoter sequences and one or more control genes, the promoter sequence(s) moderating the expression of one or more control genes. The cellular strain constitutes a cellular population and is cultured in a medium, such as a growth medium, in a micro-well **112** of a cellular integrated circuit **120***a-d* constructed on a micro-fluidic chip **100** (step **1302**). The cellular population grows to a homeostatic growth cycle. A fluid source **150** supplies a fluid to the micro-fluidic chip **100** which can include a growth medium to maintain the cellular population. The fluid flows through the channel **130** and into connected chambers **114***a-h* and micro-wells **112***a-d* to continually exchange the fluid in the micro-wells **112***a-d* with new fluid from the fluid source **150**.

[0113] The cellular integrated circuit **120***a-d* includes an electrode pair **124***a-d* having a WE and an R/CE in contact with the fluid within the micro-well **112**. An electrical source **140** applies an electrical signal **142***a-d* to the electrode pair **124***a-d* to maintain a constant electrical potential between the electrode pair **124***a-d* (step **1304**). The cellular population in homeostatic growth provides a concentration of charged metabolic products to the micro-well **112** fluid. The metabolic products provide an electrical connection between the electrode pair **124***a-d* and allows the electrical signals **142** to propagate through the solution.

[0114] The electrical source **140** determines an electrical parameter, such as a conductivity parameter, of the fluid by monitoring the change in the current applied to the WE with respect to time (step **1306**). The electrical parameter is calculated from the applied current. As the concentration of the metabolic products change in the micro-wells **112***a-d* solution, the electrical parameter value changes correspondingly. Cellular populations in homeostatic growth cycles maintain a constant concentration of metabolic products and a corresponding constant electrical parameter value.

[0115] The promoter sequence of the cellular population responds to the concentration of, e.g., presence or absence of, an inducer agent and correspondingly promotes or inhibits expression of the control gene. The fluid supplied by the fluid source **150** can include the inducer agent, or can be caused to include the inducer agent, thereby changing the concentration of the inducer agent present in the fluid in the micro-wells **112***a-d* (step **1308**), which modifies the expression of the control gene by interacting, e.g., binding, with the promotor sequence.

[0116] When expressed, the control gene modifies the metabolic process of the cellular populations. As described herein, in some embodiments, the expression of the control gene causes the cellular population to lyse thereby ceasing the production of metabolic products and decreasing the concentration of metabolic product in solution. The electrical source **140** monitors the measured electrical parameter for changes which is an indirect measurement of the cellular population (step **1310**) and corresponds with the concentration of the inducer agent.

[0117] As discussed above, in some embodiments, wafer **110** contains a number of micro-wells **112***a-d* in electrical connection with electrode pairs **124***a-d*. The wafer **110** can be composed of a solid material, or a gel-based substrate such as a hydrogel optionally including one or more growth medium (e.g., culture media, minimal media, selective media, differential media, transport media, or indicator media), buffer, solution, suspension, or combination thereof. In some embodiments in which the wafer **110** is composed of a gel-based substrate, each microwell **112***a-d* can include a single channel **130** supplying the fluid to only the connected micro-well **112***a-d*, or one or more micro-wells **112***a-d* on the wafer **110**. In some embodiments, the fluid source is configured to perfuse the fluid through the gel-based substrate to supply the micro-wells **112***a-d*.

[0118] In some embodiments, the cellular integrated circuits **120** are integrated with a biological sample, such as a tissue sample. The electrode pairs **124***a-d* of the cellular integrated circuits **120** can be affixed onto or into the biological sample. In some embodiments, the biological sample can be a tissue slice or tissue sample, such as a biopsy, a core biopsy, or needle aspirate, a fluid sample, such as a blood sample, urine sample, or saliva sample, a skin sample, a colon sample, a cheek swab, a histology sample, a histopathology sample, a plasma or serum sample, a tumor sample, living cells, cultured cells, a clinical sample such as, for example, whole blood or blood-derived products, blood cells, or cultured tissues or cells, including cell suspensions.

In some embodiments, the biological sample can be a preserved biological sample, including non-limiting examples of a formalin-fixed, or paraffin-embedded biological sample.

[0119] In some embodiments, the cellular population is a eukaryotic cellular population including at least two genetic modifications, such as a protist (e.g., Guillardia theta, Plasmodium falciparum, Plasmodium yoelii yoelii, Cryptosporidium hominis, Cryptosporidium parvum, or Thalassiosira pseudonana), plant (e.g., Arabidopsis thaliana, Cyanidioschyzon merolae, Oryza sativa, Ostreococcus tauri, Populus trichocarpa), fungi (e.g., Saccharomyces cerevisiae, Encephalitozoon cuniculi, Schizosaccharomyces pombe, Neurospora crassa, Phanerochaete chrysosporium), insect (e.g., Drosophila melanogaster, Anopheles gambiae), or animal (e.g., Caenorhabditis elegans, or Takifugu rubripes) cellular population. In some embodiments, the cellular population is a mammalian cellular population (e.g., Homo sapiens).

[0120] As discussed above, in some embodiments, electrical source **140** measures a conductivity parameter of the medium, which is optionally used to determine a value of at least one of an electrical admittance of the medium, an electrical impedance of the medium, an electrical conductance of the medium, an electrical resistance of the medium, and/or a value of another quantity that is related to any of these (e.g., a value of a quantity from which admittance, impedance, conductance, and/or resistance can be determined for the medium). As such, an electrical source **140** can function as both a source of an electrical potential difference, and a detector. In certain embodiments, the systems and devices described herein can include a separate detector from electrical source **140**. The detector can be implemented as any one of a variety of conventional electrical detectors that measure any of the quantities described above. Such detectors are well known in the art and widely available commercially, and can be readily connected to and/or integrated with the other components of the systems and devices described herein.

[0121] In certain embodiments, the systems and methods described herein include one or more electronic processors. The electronic processor(s) can be connected to any of the other components described herein, and can transmit and/or receive information to any of the other components. In particular, the electronic processor(s) can transmit control instructions to any of the other components to perform any of the actions and steps described in connection with any of the methods herein including, but not limited to, regulating the flow of growth media, introducing an inducer agent, detecting values of any of the quantities described above, and determining whether an inducer agent (e.g., a target analyte) is present in the medium based on changes in detected values of any of the above quantities.

[0122] The systems and devices can also optionally include components such as a storage medium (e.g., a non-transitory storage medium such as a ROM, magnetic storage device, optical storage device, and/or solid state storage device) for storing instructions for the electronic processor(s) and measured values obtained by the electronic processor(s), an input interface (e.g., a touchscreen and/or a conventional computing interface such as keypad, keyboard, and/or pointing device), and an output device (e.g., a display device such as a screen, an interface to a remote computing device such as a server, a computer, and/or a mobile phone). Each of these components can optionally be connected to the electronic processor(s).

[0123] Software instructions executed by the electronic processor(s) to perform any of the control and other functions and steps described herein can be stored in a memory unit connected to the electronic processor, and accessible to retrieve the instructions for execution. The instructions can also be implemented as a computer storage product with a nontransitory computer-readable medium (also can be referred to as a non-transitory processor-readable medium) having instructions or computer code thereon for performing various computer-implemented operations. The computer-readable medium (or processor-readable medium) is nontransitory in the sense that it does not include transitory propagating signals per se (e.g., a propagating electromagnetic wave carrying information on a transmission medium such as space or a cable). Examples of non-transitory computer-readable media include, but are not limited to, magnetic storage media such as hard disks, floppy disks, and magnetic tape; optical storage media such as Compact Disc/Digital Video Discs (CD/DVDs), Compact Disc-Read Only Memories (CD-ROMs), and holographic devices; magneto-optical storage media such as optical disks; carrier wave signal processing modules; and hardware devices that are specially configured to store and execute program code, such as Application-Specific Integrated Circuits (ASICs), Programmable Logic Devices (PLDs), Read-Only Memory (ROM) and Random-Access Memory (RAM) devices.

[0124] Certain methods and steps described herein can be performed by software (executed on hardware), hardware, or a combination thereof. Hardware modules may include, for example, a general-purpose processor, a field programmable gate array (FPGA), and/or an application specific integrated circuit (ASIC). Software modules (executed on hardware) can be expressed in a variety of software languages (e.g., computer code), including C, C++, Java™, Ruby, Visual Basic™, and/or other object-oriented, procedural, or other programming language and development tools. Examples of computer code include, but are not limited to, micro-code or micro-instructions, machine instructions, such as produced by a compiler, code used to produce a web service, and files containing higher-level instructions that are executed by a computer using an interpreter. For example, embodiments may be implemented using imperative programming languages (e.g., C, Fortran, etc.), functional programming languages (Haskell, Erlang, etc.), logical programming languages (e.g., Prolog), object-oriented programming languages (e.g., Java, C++, etc.) or other suitable programming languages and/or development tools. Additional examples of computer code include, but are not limited to, control signals, encrypted code, and compressed code.

EXAMPLES

[0125] To transduce gene circuit output to an electronic signal, bacterial circuits were engineered to be capable of controlling the release of ionic species via cell growth and death. Inert gold electrodes were used to measure the AC impedance when applying a sinusoidal voltage signal to the bacterial population without externally added redox mediators (FIG. **4**A).

[0126] FIG. **4**A is a line chart comparing current (μA) on the y-axis to potential (V). The chart depicts cyclic voltam-

metry measurements showing the inert redox response of MOD46A strain in growing media (grey line **400**) and media in absence of cells (black line **402**). A potential signal was applied to the media initially at -0.5 V and increased to 1 V. The potential signal was then decreased to -0.5 V. Lines **400** and **402** increase and decrease cyclically corresponding with the applied voltage, initiating and returning to less than -2.5 μA. The MOD46A+media line is below the media-only line at between approximately -0.5 V to -0.25 V and approximately 0.75 V to 1 V.

[0127] This measurement includes resistive and capacitive effects which can be decoupled by using different input potential frequencies between 1 Hz to 100 kHz. FIG. **4**B is a three-axis line chart comparing impedance (Ω) on the left axis and phase (°) on the right axis versus frequency (10^3 Hz) on the x-axis (e.g., a Bode plot) for the MOD46A strain. The impedance (black line) decreases and the phase (grey line) increases as frequency increases and correspondent equivalent circuits in the different range of frequencies.

[0128] The correspondent Bode plots (representing impedance over frequency) indicate that at high frequencies (e.g., the grey box from 30×10^3 Hz to 100×10^3 Hz), the AC current and voltage signals were in phase with each other and the corresponding genetic circuit was resistive (shown inset). At these frequencies, the phase is approximately 0, and therefore, the circuit is simplified to an active bacterial media resistance. At low frequencies (e.g., the grey box from 0×10^3 Hz to 5×10^3 Hz) the corresponding biological circuit was capacitive (shown inset) and at mid-range frequencies (e.g., the white box from 5×10^3 Hz to 30×10^3 Hz) the corresponding genetic circuit was capacitive and resistive.

[0129] FIG. **4**C is a Bode plot for fresh LB media (second line from top), only bacteria (top line), supernatant (bottom line), saturated culture (second from bottom line), and heat-killed culture (middle line) between **1000** to 50,000 Hz, where impedance measures resistive effects. Error bars show standard deviation for n=3 samples.

[0130] FIG. **4**D is a Bode plot for the same samples as FIG. **4**C between 1.5 and 4 Hz, where impedance measures resistive effects (e.g., where the equivalent circuit corresponds to a capacitor according to FIG. **4**B). The lines correspond to fresh LB media (second from bottom line), only bacteria (top line), supernatant (middle line), saturated culture (second from top line), and heat-killed culture (bottom line).

[0131] Since the impedance resulting from these resistive effects (FIG. **4**C) was generally more robust to changes in the input frequency than the one at lower frequencies (FIG. **4**D), the culture resistance was tracked. In addition, this optimized measurement was found to correlate with the population density (monitored via turbidity) as opposed to measurements at lower frequencies (e.g., see FIGS. **5**A through **5**C, which are discussed below).

[0132] FIGS. **5**A through **5**C are line charts showing signal optimization results using wild-type bacteria. FIGS. **5**A through **5**C are three-axis line charts comparing turbidity (a.u.) on the left axis and Admittance (μS) on the right axis to time (hours) on the x-axis. FIGS. **5**A through **5**C show profiles of turbidity (**500**) and admittance (**502**). FIG. **5**A corresponds to the resistance frequency range, FIG. **5**B corresponds to the combined frequency range, and FIG. **5**C corresponds to the capacitive frequency range of FIG. **4**B, respectively, e.g., for strain MG1655 *E. coli* at 100 kHz (FIG. **5**A), intermediate 1.5 kHz frequencies (FIG. **5**B), and low 1 Hz (FIG. **5**C).

[0133] To demonstrate the electrochemical interface with a minimal expression system in bacteria, a previously described construct pE35GFP containing a bacterial lysis gene was tested. The engineered bacteria grew to a stable density in the continuous culture before triggering the lysis gene upon chemical induction with isopropyl β-d-1-thiogalactopyranoside or IPTG. A decrease of more than half in culture density upon lysis induction, corresponding to an admittance (inverse of the impedance) change of approximately 2 mS (see FIG. **6**B) was observed.

[0134] FIG. **6**A is a schematic illustration of the approach using a culture of bacteria with a killing gene as the circuit output in contact with inert gold electrodes. The impedance of the culture reduces during growth (left) and increases upon the induction of bacterial death due to the clearance of charged metabolites (right).

[0135] FIG. **6**B is a line chart comparing admittance (μS) on the y axis to time (hours) on the x-axis. The line of FIG. **6**B is the admittance, which is the inverse of impedance, using an IPTG-inducible lysis construct (pE35GFP) in an electrochemostat device. The shaded region represents induction of lysis with 1 mM IPTG in the medium.

[0136] FIG. **6**B demonstrates that the induction of bacterial lysis was sufficient to cause a marked change in the conductive properties of the culture. In addition, the impedance was lower in a heat-killed culture than in an intact one (FIG. **4**C).

[0137] The contribution of the bacterial cells and their metabolic by-products to the culture resistance (FIGS. **4**C to **4**E) was also investigated. Resistive effects were dependent on the metabolic by-products from the bacterial growth and not the bacterial cells (FIG. **4**E). FIG. **4**E is a schematic illustration and histogram showing the different impedance contributions without additional redox mediators from fresh LB media, a solution of fresh LB with concentrated bacteria, and depleted LB medium resulting from bacterial growth. Error bars represent standard deviation for n=3 samples.

[0138] Since the engineered bacteria are essentially modulating the flow of electrical current in this circuit, they can be viewed as a bacterial "conductor" (or "resistor"), where the conductance or admittance can be modulated by genetic modifications that control the population dynamics. FIG. **6**C is a schematic illustration of the equivalent electrical circuit for the strategy using an alternating input voltage. The bacterial population is visualized as a simplified resistor, the output of which is controlled by the genetic circuit.

[0139] A platform interfacing this property with genetic circuits can be envisioned, where bacterial colonies with unique circuit behaviors (sensors, logic gates, and/or oscillators) are directly connected to electrodes. FIG. **6**D is a schematic diagram showing a microelectronic platform to interface between engineered bacteria and electronics. Several chambers may contain unique genetic circuits, connected via electrodes to an impedance detector.

[0140] The dynamics of gene circuits have primarily been investigated using well-defined submillimeter-scale bacterial colonies. Interfacing synthetic biology with electrodes utilizes robust population behavior at larger scales, where dynamical behaviors are less characterized. To study the dynamic behavior in a macroscale continuous culturing system, a customized milliliter scale chemostat with disposable

gold electrodes in contact with the culture was fabricated, shown in FIG. **7**A and FIG. **8**A).

[0141] FIG. **7**A is a schematic illustration (left) and image of electrochemostat components included in a 3D-printed holder containing a culture tube, in which the disposable electrode consisting of gold counter/reference (C/RE) and working (WE) electrodes are immersed, and an external turbidimeter detector.

[0142] FIG. **8**A is an image showing the complete macrochemostat set-up including the pumping system under continuous mixing conditions (left) and an image of 'electrochemostat' components included in a 3D printed holder containing a culture tube, in which the disposable electrode consisting of gold counter/reference (C/RE) and working (WE) electrodes are immersed, and a printed turbidimeter circuit board (right). The image of FIG. **7**A was taken within the dashed box on the left image of FIG. **8**A.

[0143] A turbidity sensor provided real-time data and direct correlation between impedance and culture density, resulting in an "electrochemostat" system (FIGS. **8**C and **8**D). FIG. **8**C is a schematic illustration of the complete macro-chemostat system showing the pumping system under continuous mixing conditions. FIG. **8**D is a three-axis line chart comparing the logarithmic optical density (OD) at 600 nm on the left axis (corresponding to the upper points) and the logarithmic transmittance on the right axis (corresponding to the lower points) to time (min) on the x-axis using a culture containing MG1655 *E. coli*.

[0144] A cell population with a control gene capable of inducing lysis in the presence of a heavy metal toxin (e.g., arsenic) was investigated (As-lysis, as shown in FIG. **7**B). FIG. **7**B is a schematic illustration of an equivalent electrical circuit for a bacterial population engineered with an inducible promoter driving the expression of the lysis gene, E, and schematic of engineered bacterial (As-lysis) death upon arsenic induction.

[0145] After reaching a steady population state, a fast triggering of lysis resulted from the induction with arsenic, in both admittance and turbidity was observed in FIG. **7**C. FIG. **7**C is a three-axis line chart comparing the profiles of the admittance (**700**) and turbidity (**702**) using an arsenic-sensitive strain in an electrochemostat device. The shaded region represents the duration of 250-ppb arsenic induction.

[0146] Good reproducibility was found for the drop in signal with relative standard deviations (RSDs) of 7.2 and 2.1 % for turbidity readouts, respectively, in n = 3 experiments. On the other hand, this bacterial cell population did not lyse in response to a related toxin such as copper. FIG. **7**D is a three-axis line chart comparing the profiles of the admittance (**704**) and turbidity (**706**) using an arsenic-sensitive strain with 250 ppb copper in an electrochemostat device.

[0147] Furthermore, the absence of sensitive bacteria showed a negligible increase in conductivity and no change in turbidity when induced with arsenic. FIG. **9**A is a three-axis line chart comparing the profiles of the admittance (**900**) and turbidity (**902**) for LB media in absence of bacteria with shaded region representing induction with 250 ppb arsenic.

[0148] This arsenic biosensor was compared with a chemical sensing methodology, stripping voltammetry. This approach relies on the reduction of the arsenic ions on the gold electrode and next stripping and study of the arsenic oxidation at different potentials. Arsenic and copper were individually detected at +0.20 V versus Ag/AgCl. FIG. **7**E is a line chart comparing measured current (μA) on the y-axis to potential (V) on the x-axis and showing square-wave voltammetry profiles for 250-ppb arsenic (black) and 250 ppb copper (grey). The dashed line shows the signal of the HNO_3 buffer (background). The oxidation potential at the maximum current intensity versus background signal is indicative of the presence of the metal.

[0149] However, the similarity in oxidation potential causes copper to interfere in the chemical approach for arsenic analysis but not for the bacterial approach. FIG. **7**F is a heat map of selectivity to arsenic versus copper using both bacterial lysis and chemical approaches. A further control using a copper-sensitive strain (Cu-lysis) demonstrated the lysis triggering in the presence of copper. FIG. **9**B is a a is a three-axis line chart comparing the profiles of the admittance (**904**) and turbidity (**906**) including for copper detection using copper-sensitive strain with shaded region representing induction with 250 ppb copper (shaded area). Thus, using the bacterial lysis approach described above, it was possible to discriminate between two related heavy metal toxins compared to the standard electrochemical assay for this application, in which discrimination is considerably more difficult.

[0150] The detection methods described herein can also be applied to oscillatory synthetic circuits such as the synchronized lysis circuit (SLC). FIG. **10**A is a schematic illustration of the equivalent electrical circuit when using a SLC connected to the potential source. The circuit is composed of an AHL-based quorum sensing system driving a phage lysis gene. The protein LuxI regulates the AHL-based quorum sensing system. This bacterial circuit was previously shown to generate oscillatory population dynamics in microfluidic devices and in animal models, which occurs via cycles of growth and lysis. The genetic circuit contains a common promoter (pLuxI) that drives the expression of LuxI, which produces the quorum sensing molecule acyl homoserine lactone (AHL) that further activates the promoter by binding the activator LuxR.

[0151] The promoter also drives the expression of the lysis gene from the phage phiX174, E. AHL provides an intercellular synchronization mechanism: after reaching a threshold level, synchronized lysis at the population level ensues. A remaining population of bacteria yields new AHL where the cycle repeats, resulting in oscillatory growth and lysis behavior. When measuring the SLC strain with the electrochemostat, oscillatory culture density was observed, with corresponding behavior in the admittance signal over time. FIG. **10**B is a three-axis line chart comparing the profiles of the admittance (**1000**) and turbidity (**1002**) using this strain in an electrochemostat device. The SLC is harbored by a strain Salmonella enterica subsp. enterica serovar Typhimurium.

[0152] Correlation of the periodical lysis events was found between the admittance and turbidity signals, with a period of 3.2 ± 0.6 hours. The long-term bacterial population oscillations resulted in accumulation of bacterial debris over time, leading to increasing baseline in the turbidity signal. Compared to common variable resistors, which usually require manual manipulation or computer coding to modify the resistance, the SLC allows a bacterial population to exhibit autonomous oscillatory resistance variations over time.

[0153] A challenge in synthetic biology is a minimally tracking gene expression without the need for fluorescent

proteins and associated complex imaging equipment. Given recent efforts in the development of electrochemical platforms and the minimal nature of impedance measurement for circuit dynamics by directly connecting bacteria with electrodes, a miniature device to demonstrate the utility of the methods described herein for this purpose was developed. A fluidic chip with multiple milli-scale growth chambers, where each chamber contained electrodes, was fabricated. FIG. **11**A is an image of the bacterial integrated circuit (bIC) device (left) and schematic diagram (right) showing the design of the growth chamber and adjoining gold electrodes. Each bIC integrates lithography-fabricated electrodes, including a reference/counter electrode (R/CE) and a WE electrode, on 0.6-mm-diameter traps where bacteria form 3D continuous cultures.

[0154] The electrodes were composed of conductive interlayers of chrome and gold, which made up the reference/counter and working electrodes, see FIGS. **12**A, **12**B, **12**E. FIG. **12**A is a schematic diagram showing the fabrication of electrodes on glass via sequential deposition of conductive chrome (Cr) and gold (Au) layers and a final spin of a polyimide insulator (PI) layer. FIG. **12**B is an image of the lithography fabricated electrodes on glass.

[0155] Each of the electrode-containing growth chambers was seeded with engineered bacteria, which were referred to as bICs, since each one connects genetic circuit output to electrochemical measurement. Multiple bICs were connected in parallel to a single potentiostat and a multiplexing module, allowing for the measurement of multiple strains. FIG. **11**B is a schematic illustration of the equivalent electrical circuit illustrating multiple parallel interconnecting bICs measured with a single potentiostat. The dynamics of each bIC were measured sequentially in time intervals (Δt). A device containing n unique bICs may output an oscillatory signal at ti, a steady signal at t_{ii}, and other distinct signals up to t_n.

[0156] Resistive effects were measured in the growth chambers of this device over a wide range of input frequencies, see FIG. **12**C. FIG. **12**C is a line chart comparing the signal phase (°) on the y-axis to frequency (kHz) on the x-axis in a bIC for LB media and corresponding equivalent circuits (inset) at different frequencies.

[0157] The electrodes detected changes in environmental ion concentration via admittance measurements, see FIG. **12**D. FIG. **12**D is a line chart comparing the admittance (μS) on the y-axis to time (h) on the x-axis showing the response of microfluidic electrodes using LB, induced with 100 μM NaCl (shadowed) using same procedure as with multiple bICs.

[0158] The bIC chip displayed the potential to detect gene circuit behavior via impedance detection of ions in solution. The capabilities of the electrochemical platform using the miniaturized device were characterized. The SLC strain was investigated first, using both impedance and transmitted light (TL) measurements to confirm that the impedimetric output corresponded to population dynamics. Bacterial growth oscillations were observed in both TL and admittance, showing that oscillatory impedance output could be achieved with this circuit as a bIC. FIG. **11**C is an image of a bIC, containing the Salmonella SLC strain, taken using transmitted light (TL). The bacteria began at a low cell density (i) from which they reached the quorum threshold and lysed (ii and iii), and then repeated the process (iv). FIG. **11**D is a three axis line chart comparing admittance (mS) on the left axis and inverse transmittance (a.u.) on the right axis to time (hours) on the x-axis. FIG. **11**D shows profiles of admittance (**1100**) and inverse of the TL (**1102**) for the strain in FIG. **11**C. FIG. **11**D includes numerals i, ii, iii, and iv corresponding to the time points of the images of FIG. **11**C.

[0159] Periodical lysis events were found every 7.6 ± 1.2 hours for both admittance and turbidity signals. In comparison with the electrochemostat, the admittance values in this device were lower, likely due to the lower electrode surface area. The period discrepancy was related to the differences between approaches and culture sizes. The sensing capabilities of the device were also investigated using a bacterial cell population with a control gene capable of inducing lysis in the presence of arsenic. The bacterial population reached a steady state in both TL and admittance before induction with arsenic. Subsequent lysis resulted in the admittance sharply decreasing to another steady state within 1 hour.

[0160] FIG. **11**E includes images showing the response of the bIC with the arsenic-inducible cell population in *E. coli* MG1655 showing the steady growth state (i) and after 250-ppb arsenic induction (ii). FIG. **11**F is a three axis line chart comparing admittance (mS) on the left axis and inverse transmittance (a.u.) on the right axis to time (hours) on the x-axis. FIG. **11**F shows profiles of admittance (**1104**) and inverse of the TL (**1106**) for the strain in FIG. **11**E.

[0161] The TL exhibited a slight drift after the lysis event, likely due to a small portion of the bacterial population exhibiting both growth and lysis in the presence of arsenic, but where the population was small enough not to be detected by electrochemical measurement. Reproducibility between traps indicated RSDs of 10.8 and 13.0% (n = 4) for TL and admittance, respectively. These results demonstrate the functionality of the bIC device as a miniature platform for electrochemically measuring genetic circuit output. More generally, the electrochemical monitoring of engineered bacteria provides a label-free method for the real-time collection of expression data, in contrast to standard optical modalities.

[0162] Conventionally, assessment of gene circuit functionalities has been predominantly reliant on detection of specific fluorescent or colorimetric proteins. In contrast, the methods, systems, and devices described herein implement the direct interfacing of engineered microorganisms with microelectronics to provide a framework for a new class of hybrid biological electronic devices and biosensors where cellular "logic" informs electronic output. Engineered bacterial circuits can be interfaced with microelectronics with simple electrical measurement values detected. The increase of conductivity of the solution is generally due to energy metabolism or transportation of ions through the cell membrane, where uncharged or weakly charged substrates are transformed into highly charged smaller ions. Direct interfacing of electrodes with the cell culture represents a simplified format with which cell populations can be connected to an electrical output. As noted previously, redox active species were not introduced into the medium prior to measuring electrical signals, and the measured values of electrical parameters such as admittance, impedance, resistance, and conductance reflect changes in concentrations of metabolic products generated by the cell populations, rather than changes in concentrations of reduced or oxidized redox active species.

[0163] Consequently, the bacterial population dynamics were not affected by external inputs other than the target analytes of interest. In this sense, bacterial lysis, engineered or otherwise induced, functions as a general interface for electronics. As an alternative to conventional optics, electrical parameter measurement allows for exploring more complex nonoptically transparent materials and samples. Compared to other electrochemical approaches such as stripping voltammetry, genetic circuits can provide the selectivity expected from a biosensor by connecting genetic sensing elements naturally present in cells to impedimetric output via lysis. By connecting the bacterial sensing output directly to electrodes, isolation and preparation steps are not required to build a biochemical sensor. In addition, compared to common molecular-based biosensors, which use enzymes or antibodies, the proposed lysis triggering of bacteria enables dynamic sensing because cells can be grown in continuous environmental conditions. For example, the sensor responds in 40 ± 10 min after triggering (FIG. **7**C), an interval which is likely dependent on the translation of proteins.

[0164] Biofouling processes may increase the divergence between turbidity and admittance (FIG. **10**B) over time. The population control and the interfacing of genetic lysis circuits with electronics provides an inherent anti-biofouling mechanism because of the continuous clearing of the chamber after certain periods of time or after certain triggering conditions.

[0165] Isolating the electrodes from the bacterial culture using permeable membranes or using microfluidic chambers that exclusively allow the supernatant to be measured can improve the performance of individual, and arrays of, bICs. Three-dimensional (3D) layering is possible with the use of electrodes compared to conventional imaging methods, as well as in vivo depth tissue measurements using implantable electronics for applications where bacteria reside in vivo.

[0166] The compartmentalization of bacterial colonies in the bIC facilitates high-throughput monitoring of large libraries. The methods, systems, and devices described herein are applicable to readily connect microelectronics with a variety of genetic circuits such as logic gates, switches, oscillators, sensors, and more sophisticated microbial communities, which permit construction of a variety of hybrid computational devices, where an array of bacterial colonies process external information through genetic circuitry, which is then transferred via impedance measurements (or measurements of another electrical parameter), to an electronic device for analysis.

Procedures

[0167] Engineered bacterial strains used in the methods, systems, and devices described herein can be generated using plasmids constructed by the circular polymerase extension, standard restriction digest/ligation, or Gibson assembly cloning methods. Attenuated Salmonella enterica subsp. enterica serovar typhimurium, SL1344 strain ELH1301, and *Escherichia coli* MG1655 were used as the bacterial hosts for transformation and electroporation of the experimental plasmids. For the IPTG-induced lysis experiments, we used the plasmid pE35GFP, which was provided by R. Young. This plasmid contains a lac promoter that transcribes gene E after IPTG induction, which causes lysis of the host. The arsenic-induced lysis experiments were performed with the As-lysis plasmid. This plasmid contains an arsenic-sensitive promoter driving the expression of its repressor (ArsR, from E. coli plasmid R773) and the lysis gene, E. The control with copper-sensitive (Cu-lysis) strain contains a copper-sensitive promoter driving the expression of its repressor (CueR, from E. coli plasmid) and the lysis gene, E. For the SLC, the strains designated MOD46A and MOD101 were used, and are described in Din et al., Nature 536(7614): 81-85 (2016), the entire contents of which are incorporated by reference herein.

[0168] Impedance characterization was performed using an electrode pairs fabricated from an inert gold material. All electrochemical measurements were performed with a commercial potentiostat (PalmSens4 with an impedance module, Enschede, Holland) at a 10-mV voltage amplitude and zero DC bias to avoid possible electrochemical reactions. Measurements were performed in the range of frequencies starting from 1 MHz to 1 Hz. Corresponding impedance versus frequency plots were analyzed using the Randles model as the electrical equivalent circuit in a non-Faradaic system. Under these conditions, the total impedance could be expressed by the medium resistance (Rs) and the double-layer capacitance (Cdl) in series. By treating the impedance as a vector, the total impedance is divided into real and imaginary parts.

[0169] Thus, when the phase is zero, the real impedance is the main component of the total impedance and can be represented as the medium resistance. Stripping voltammetry measurements (FIG. **7**E) were performed using commercial screen-printed gold electrodes using Ag/AgCl as the reference electrode (C220AT, DropSens-Metrohm, Riverview, FL, USA) and a commercial potentiostat (PalmSens4, Enschede, Holland).

[0170] The arsenic and copper solutions were prepared in 0.2 M HNO_3 from standard solutions at 10 ppm (ERA Waters, Golden, CO, USA). Deposition of metals on the working electrode was performed at -0.6 V for 300 s. Square-wave voltammetry profiles were obtained between 0 to +0.3 V versus Ag/AgCl, at 25 Hz, 5 mVs-1, and 40 mV, applying a cleaning step at +0.3 V for 60 s between measurements.

[0171] Macroscale bacterial characterization was performed using an electrochemostat composed of a custom-built 3D printed housing where the electrochemical and turbidity detectors were enclosed (FIG. **8**). This assembly was placed in a 37° C. incubator for experiments. The 3-ml bacterial culture, confined in an autoclavable beaker, is stirred at 700 rpm. Fresh LB media (containing 0.075% Tween-20; BD Difco, LB broth, Miller) was continuously supplied with a P625/10K.143 peristaltic pump (Instech, Plymouth Meeting, PA, USA) at a typical flow rate of 25 µl/min. A removable lid held in place inlet and outlet tubings and the disposable screen-printed electrochemical sensor consisting of a 3-mm diameter gold working electrode and a 4-mm gold counter/reference electrode (C220AT, DropSens-Metrohm, Riverview, FL, USA). The lid also had an extra opening for aerobic conditions. A new electrode was used for each experiment and connected to a type B screen-printed electrode adapter (IO Rodeo, Pasadena, CA, USA) interfaced to the potentiostat. The optimal potential frequency for impedance measurement in these experiments was 100 kHz. Turbidity and impedance values were taken every 30 s and transmitted via USB to a PC.

[0172] Photolithography was used to pattern the multi-bICs on a 75 mm by 50 mm glass slide. Interconnected layers of Cr (20 nm)/Au (200 nm) were deposited (Denton Discovery 18 Sputter System) and chemically etched to form the eight pairs of electrodes. A second layer of polyimide (PI-2545, HD Microsystems, USA) as electrical insulation was coated to yield a 1.2-μm film. This top layer was etched by reactive ion etching (Plasmalab Oxford P80), exposing only the electrodes and connection pads. All the contact pads were aligned to a 3D-printed holder, which facilitated alignment with the microscope and connection using conductive screws interconnected to a flat ribbon cable attached to the MUX8-R2 multiplexer (FIG. **12**E).

[0173] The module was controlled by the potentiostat that measured the eight electrodes with a 30-s delay between measurements. Optical and impedance measurements were taken every 4 and 5 min, respectively, for every bIC. A potential frequency of 1 MHz was used for the impedance measurements. Millifluidic devices were constructed from polydimethylsiloxane (Dow Corning, Sylgard 184), which was molded and baked on a silicon master with customized features.

[0174] A sinusoidal AC potential was applied to the system. The calculated impedance was determined from the ratio of the applied voltage and the measured current that flows through the system. The impedance spectra were obtained by sequential measurements for each single frequency. The impedance in aqueous solution was represented by an equivalent circuit consisting of two components, a double layer capacitor (Cdl) and bulk medium resistor (Rs) connected in series.

[0175] Cdl accounts for the effect of ionic species on the capacitance near the surface of an electrode. Rs represents the bulk resistance of the solution, accounting for the change in conductivity and charge transport across the bulk solution. There were three regions in the impedance spectrum shown in FIG. **4**B. These were represented by two components in the equivalent circuit individually, and their combination. The double layer region dominated by Cdl was in the frequency range of 1 Hz to 5 Hz (left shaded area). The resistive region dominated by Rs was in the frequency range between 35 kHz to 100 kHz (right shaded area). The region dominated by both Cdl and Rs was in the frequency range of 5 Hz-35 kHz (not shaded).

[0176] The effect of the double layer capacitance was noticeable at phase angle values close to -90° as shown. In the high frequency range, resistive region, the phase angle values were close to 0° and the magnitude of the impedance was constant, as resistive impedance was independent of frequency. At the high frequency interval (> 35 kHz) there was no substantial contribution to the double layer capacitance. Thus, the most important contribution to the total impedance of the system at high frequencies was related to the resistance of the medium, which was independent from the frequency. This region is defined as the resistive region, in which ion conduction in the medium was dominant (FIG. **4**B, left shaded region). The changes in the double layer of the electrode, and the changes in the medium during bacterial growth were detected by measuring impedance at different frequencies, as described herein, to explore microbiological detection at these frequencies.

[0177] Bacterial cultures and media were studied using 50 μL of sample solution as shown in FIG. **4**A through FIG. **4**E. The different solutions characterized included fresh LB media ((BD Difco, LB broth, Miller) containing 0.075 % Tween-20), concentrated bacteria mixed with fresh LB media, supernatant from a MG1655 *E. coli* culture grown to an optical density at 600 nm (OD600) of 0.61, and a heat-killed MG1655 culture (10 minutes at 90° C.), at an OD600 of 0.18. Redox inactivity of the bacteria and LB on the screen printed gold electrodes was characterized using cyclic voltammetry (CV) between -0.5 to +1.0 V with 10 mV potential step at a scan rate of 50 mV/s. CV only showed the oxidation of water to oxygen at high potentials and the reduction to hydrogen at low potentials (FIG. **4**A).

[0178] The synchronized lysis circuit (SLC) cell populations were engineered using two plasmids, an activator (KAN, ColE1), and a lysis plasmid built by taking the lysis gene, E, from the ePop plasmid via PCR and cloning it into a vector (CM, p15A) under the control of the luxI promoter. The hlyE gene was obtained via PCR from the genomic DNA of MG1655. The SLC strains were cultured in LB media with 50 μg mL-1 and 34 μg mL-1 of kanamycin (KAN) and chloramphenicol (CM) respectively, along with 0.2 % glucose, in a 37° C. incubator. The stimuli-dependent strains were cultured in LB media with 50 μg mL-1 KAN along with 0.2 % glucose, in a 37° C. incubator.

[0179] The customized turbidity printed circuit board included an OPT101 monolithic photodiode with on-chip transimpedance amplifier BPW34 infrared/visible silicon detector and a 950 nm infrared LED emitter (RadioShack, Forth Worth, TX, US). A **555** timer along with a modulator/demodulator (AD630) enable filtered signal. Turbidity values were transmitted via USB to a PC in which sampling time and gain were controlled. Detection was performed at 90° versus the LED every 30 s. (FIG. **8**B). Calibration of the turbidity sensor was performed for a growth curve of a MG1655 *E. coli* culture in LB media simultaneously measuring with the customized turbidity board and a spectrometer at 600 nm every 15 min (FIG. **8**C).

[0180] Milli-fluidic devices were constructed from polydimethylsiloxane (PDMS) (Dow Corning, Sylgard 184) which was molded and baked on a silicon master with customized features. The Si wafer formed by cross-linked photoresist consisted of three layers of 4, 30 and 100 μm height, respectively. Individual devices consisted of 10 × 9 array bacterial traps of 0.6 mm diameter and 30 μm height connected via two 4 μm height channels to a main channel, which provided fresh media to the individual bacterial cultures. Devices were cut out of the baked PDMS and holes were punched in the devices to allow the connection of fluid lines.

[0181] When electrodes were used for analysis, a conventional mask aligner was used for alignment. The devices were then kept for 2 h in a 37° C. incubator for complete bonding. The devices were placed on a microscope 3D printed stage for imaging and connected to the impedance module. Fluid lines were connected to the device from two syringes, one supplying the medium, and another acting as waste reservoir. Hydrostatic pressure driven flow was controlled by maintaining a 15-inch relative height difference between the inlet and outlet syringes. In these microfluidic experiments, 0.075 % Tween-20 was added to the media and cell suspension fluid to prevent cells from adhering to PDMS channels, electrodes, tubing or ports of the device.

[0182] For the electrochemostat experiments, 60 μL of overnight culture at an OD600 of 0.6 was added to 3 mL of LB supplemented with 0.075 % Tween-20 and the appropriate antibiotics. The surfactant addition prevented bacteria

from adhering to glass, electrodes and tubing. Bacterial loading on the bIC device was performed by an automatic echo acoustic liquid handler (Labcyte Inc., San Jose, CA, US). The robotic transducer transferred 2.5 nL of overnight culture at and OD600 of 0.8 to each trap. Cells were transferred 10 minutes after exposing glass and PDMS to oxygen plasma for optimal cell loading.

[0183] A Nikon Eclipse TI epifluorescent microscope was used for phase-contrast based imaging. For the acquisition of images, a CoolSNAP HQ2 CCD camera from Photometrics was used. The microscope and acquisition were controlled by Nikon Elements software. A plexiglass incubation chamber connected to a heating unit, which encompassed a wide area around the stage, was used to maintain the temperature of the microfluidic device at 37° C. Phase-contrast images were taken at 10x magnification at 10-50 ms exposure times. Fluorescent imaging at 10x was performed at 200 ms for GFP, 30 % setting on the Lumencor

[0184] SOLA light source. Images were taken every 4 minutes for the course of a typical experiment.

[0185] For the electrochemostat, data from PSTrace software was used to plot the reciprocal of the total impedance value overtime. Data for turbidity and impedance was smoothed with 5 points of window using a polynomial order 2 and a Savitzky-Golay method, to mitigate temperature fluctuations in the incubator. For the microfluidic device, microscope images were treated with Image J. The reciprocal of transmitted light intensity profiles were obtained by analyzing frames in Image J from the phase/contrast channel and plotting the mean pixel intensity over time. The reciprocal of that plot is presented in FIG. **11**D and FIG. **11**F. Electrochemical plots were obtained from PSTrace software and admittance was calculated from the total value of impedance.

OTHER EMBODIMENTS

[0186] A number of embodiments have been described by way of example above. However, none of the embodiments described herein should be construed to limit the scope of the following claims.

What is claimed is:

1. A method of measuring a cell population, the method comprising:

culturing a cell population in a growth medium, wherein the cell population comprises a cell strain comprising a control gene and a promoter sequence;

positioning first and second electrodes in contact with the growth medium, wherein the first and second electrodes are connected to different terminals of an electrical source;

applying an electrical signal between the two electrodes;

measuring an electrical parameter of the growth medium as a function of time; and

determining information about the cell population based upon the measured electrical parameter.

2. The method of claim **1**, wherein the electrical parameter comprises an electrical admittance of the growth medium.

3. The method of claim **1**, wherein the electrical parameter comprises an electrical impedance of the growth medium.

4. The method of claim **1**, wherein the electrical parameter comprises a conductance of the growth medium.

5. The method of claim **1**, wherein the electrical parameter comprises a resistance of the growth medium.

6. The method of claim **1**, wherein applying the electrical signal comprises applying an electrical potential difference between the two electrodes.

7. The method of claim **1**, wherein the cell population comprises one or more different bacterial strains.

8. The method of claim **1**, wherein the cell population comprises one or more different yeast strains.

9. The method of claim **1**, wherein the cell population comprises one or more different mammalian cell strains.

10. The method of claim **1**, wherein the cell population comprises one or more different insect cell strains.

11. The method of claim **1**, wherein the growth medium is selected from the group consisting of a liquid medium and a solid medium.

12. The method of claim **11**, wherein the solid medium comprises a hydrogel.

13. The method of claim **11**, wherein the solid medium comprises at least one gel-based material.

14. The method of claim **1**, further comprising maintaining the cell population in a chamber, and flowing the growth medium through the chamber.

15. The method of claim **14**, further comprising measuring the electrical parameter in different portions of the flowing growth medium as a function of time.

16. The method of claim **1**, wherein the control gene is activated by at least one mechanism selected from the group consisting of: an external inducer; a self-activated inducer; and a combination of an external inducer and a self-activated inducer.

17. The method of claim **16**, wherein the external inducer is selected from the group consisting of: exposure to optical radiation, a change in temperature, a chemical reagent, a viral agent, and combinations thereof.

18. The method of claim **1**, wherein measuring the electrical parameter comprises measuring at least member selected from the group consisting of: a cyclic voltammetry signal for the growth medium; a potentiometry signal for the growth medium; an electrical conductivity of the growth medium; a phase difference between electrical signals measured for the growth medium; and combinations thereof.

19. The method of claim **1**, wherein the electrical signal is an alternating current (AC) electrical signal.

20. The method of claim **1**, wherein the electrical signal is a direct current (DC) electrical signal.

21. The method of claim **1**, wherein the control gene is a single-gene lysis gene, a multi-gene lysis gene, one or more TA module genes, one or more toxin genes, or one or more peptide toxin gene.

22. The method of claim **1**, wherein the control gene is activated by a target analyte.

23. The method of claim **22**, wherein the target analyte comprises a chemical species.

24. The method of claim **22**, wherein the target analyte comprises a biochemical molecule.

25. The method of claim **23** or claim **24**, further comprising determining a relative concentration of the target analyte in the growth medium as a function of time based on the information about the cell population.

26. The method of claim **1**, wherein the information about the cell population comprises information about a relative concentration of at least one metabolite generated by the cell population.

27. The method of claim **1**, comprising measuring the electrical parameter of the growth medium without introducing a redox active species into the growth medium.

28. A system for monitoring a cell population, the apparatus comprising:

a reservoir configured to contain a cell population, wherein the cell population comprises a cell strain comprising a control gene and a promoter sequence;

electrodes configured to contact a growth medium in the reservoir;

an electrical source connected to the electrodes and configured to apply an electrical signal to the growth medium;

a detector configured to measure an electrical parameter of the growth medium as a function of time; and

an electronic processor connected to the detector and configured to receive measurements of the electrical parameter, and to determine information about the cell population based on the measured electrical parameter.

29. The system of claim **28**, wherein the reservoir is formed in a microfluidic device.

30. The system of claim **29**, wherein the reservoir comprises an inlet and an outlet, and wherein the apparatus comprises a fluidic device configured to transport the growth medium through the reservoir from the inlet to the outlet.

31. The system of claim **28**, wherein the reservoir comprises a recess configured to support a solid growth medium.

32. The system of claim **28**, wherein the electrical parameter comprises an electrical admittance of the growth medium.

33. The system of claim **28**, wherein the electrical parameter comprises an electrical impedance of the growth medium.

34. The system of claim **28**, wherein the electrical parameter comprises a conductance of the growth medium.

35. The system of claim **28**, wherein the electrical parameter comprises a resistance of the growth medium.

36. The system of claim **28**, wherein applying the electrical signal comprises applying an electrical potential difference between the two electrodes.

37. The system of claim **28**, wherein the cell population comprises one or more different bacterial strains.

38. The system of claim **28**, wherein the cell population comprises one or more different yeast strains.

39. The system of claim **28**, wherein the cell population comprises one or more different mammalian cell strains.

40. The system of claim **28**, wherein the cell population comprises one or more different insect cell strains.

41. The system of claim **28**, wherein the growth medium is selected from the group consisting of a liquid medium and a solid medium.

42. The system of claim **41**, wherein the solid medium comprises a hydrogel.

43. The system of claim **41**, wherein the solid medium comprises at least one gel-based material.

44. The system of claim **28**, further comprising maintaining the cell population in a chamber, and flowing the growth medium through the chamber.

45. The system of claim **44**, further comprising measuring the electrical parameter in different portions of the flowing growth medium as a function of time.

46. The system of claim **28**, wherein the control gene is activated by at least one mechanism selected from the group consisting of: an external inducer; a self-activated inducer; and a combination of an external inducer and a self-activated inducer.

47. The system of claim **46**, wherein the external inducer is selected from the group consisting of: exposure to optical radiation, a change in temperature, a chemical reagent, a viral agent, and combinations thereof.

48. The system of claim **28**, wherein measuring the electrical parameter comprises measuring at least member selected from the group consisting of: a cyclic voltammetry signal for the growth medium; a potentiometry signal for the growth medium; an electrical conductivity of the growth medium; a phase difference between electrical signals measured for the growth medium; and combinations thereof.

49. The system of claim **28**, wherein the electrical signal is an alternating current (AC) electrical signal.

50. The system of claim **28**, wherein the electrical signal is a direct current (DC) electrical signal.

51. The system of claim **28**, wherein the control gene is a single-gene lysis gene, a multi-gene lysis gene, one or more TA module genes, one or more toxin genes, or one or more peptide toxin gene.

52. The system of claim **28**, wherein the control gene is activated by a target analyte.

53. The system of claim **52**, wherein the target analyte comprises a chemical species.

54. The system of claim **52**, wherein the target analyte comprises a biochemical molecule.

55. The system of claim **53** or claim **54**, further comprising determining a relative concentration of the target analyte in the growth medium as a function of time based on the information about the cell population.

56. The system of claim **28**, wherein the information about the cell population comprises information about a relative concentration of at least one metabolite generated by the cell population.

57. The system of claim **28**, comprising measuring the electrical parameter of the growth medium without introducing a redox active species into the growth medium.

58. A method for determining a presence of a target analyte in a medium, the method comprising:

culturing a cell population a medium, wherein:

the cell population comprises a cell strain comprising a control gene and a promoter sequence;

the promoter sequence is configured to bind a target analyte and, upon binding the target analyte, to activate the control gene to be expressed; and

the control gene is configured to modulate growth of the cell population;

introducing the target analyte into the medium;

measuring a change in an electrical parameter of the medium arising from a change in a concentration of a metabolite generated by the cell population following introduction of the target analyte; and

identifying a presence of the target analyte in the medium based on the measured change in the electrical parameter.

59. The method of claim **58**, wherein the target analyte is a protein, oligonucleotide, or chemical compound.

60. The method of claim **59**, wherein the chemical compound is a metal-containing chemical compound.

61. The method of claim **58**, wherein the electrical parameter comprises an electrical admittance of the growth medium.

62. The method of claim **58**, wherein the electrical parameter comprises an electrical impedance of the growth medium.

63. The method of claim **58**, wherein the electrical parameter comprises a conductance of the growth medium.

64. The method of claim 58, wherein the electrical parameter comprises a resistance of the growth medium.

65. The method of claim 58, wherein applying the electrical signal comprises applying an electrical potential difference between the two electrodes.

66. The method of claim 58, wherein the cell population comprises one or more different bacterial strains.

67. The method of claim 58, wherein the cell population comprises one or more different yeast strains.

68. The method of claim 58, wherein the cell population comprises one or more different mammalian cell strains.

69. The method of claim 58, wherein the cell population comprises one or more different insect cell strains.

70. The method of claim 58, wherein the growth medium is selected from the group consisting of a liquid medium and a solid medium.

71. The method of claim 70, wherein the solid medium comprises a hydrogel.

72. The method of claim 70, wherein the solid medium comprises at least one gel-based material.

73. The method of claim 58, further comprising maintaining the cell population in a chamber, and flowing the growth medium through the chamber.

74. The method of claim 73, further comprising measuring the electrical parameter in different portions of the flowing growth medium as a function of time.

75. The method of claim 58, wherein the control gene is activated by at least one mechanism selected from the group consisting of: an external inducer; a self-activated inducer; and a combination of an external inducer and a self-activated inducer.

76. The method of claim 75, wherein the external inducer is selected from the group consisting of: exposure to optical radiation, a change in temperature, a chemical reagent, a viral agent, and combinations thereof.

77. The method of claim 58, wherein measuring the electrical parameter comprises measuring at least member selected from the group consisting of: a cyclic voltammetry signal for the growth medium; a potentiometry signal for the growth medium; an electrical conductivity of the growth medium; a phase difference between electrical signals measured for the growth medium; and combinations thereof.

78. The method of claim 58, wherein the electrical signal is an alternating current (AC) electrical signal.

79. The method of claim 58, wherein the electrical signal is a direct current (DC) electrical signal.

80. The method of claim 58, wherein the control gene is a single-gene lysis gene, a multi-gene lysis gene, one or more TA module genes, one or more toxin genes, or one or more peptide toxin gene.

81. The method of claim 58, wherein the control gene is activated by a target analyte.

82. The method of claim 81, wherein the target analyte comprises a chemical species.

83. The method of claim 81, wherein the target analyte comprises a biochemical molecule.

84. The method of claim 82 or claim 83, further comprising determining a relative concentration of the target analyte in the growth medium as a function of time based on the information about the cell population.

85. The method of claim 58, wherein the information about the cell population comprises information about a relative concentration of at least one metabolite generated by the cell population.

86. The method of claim 58, comprising measuring the electrical parameter of the growth medium without introducing a redox active species into the growth medium.

87. A device for detecting a target analyte, the device comprising:

a substrate comprising a reservoir configured to contain a cell population, wherein:

the cell population comprises a cell strain comprising a control gene and a promoter sequence;

the promoter sequence is configured to bind a target analyte and, upon binding the target analyte, to activate the control gene to be expressed; and

the control gene is configured to modulate growth of the cell population;

an inlet formed on or in the substrate and connected to the reservoir;

a fluid delivery mechanism connected to the inlet and configured to introduce the target analyte into the reservoir;

electrodes configured to contact a medium supporting the cell population in the reservoir;

an electrical source connected to the electrodes and configured to apply an electrical signal to the medium;

a detector configured to measure an electrical parameter of the medium arising from a change in a concentration of a metabolite generated by the cell population following introduction of the target analyte; and

an electronic processor connected to the detector and configured to receive measurements of the electrical parameter, and to identify the target analyte based on the measured change in the electrical parameter.

88. The device of claim 87, wherein the electrical parameter comprises an electrical admittance of the growth medium.

89. The device of claim 87, wherein the electrical parameter comprises an electrical impedance of the growth medium.

90. The device of claim 87, wherein the electrical parameter comprises a conductance of the growth medium.

91. The device of claim 87, wherein the electrical parameter comprises a resistance of the growth medium.

92. The device of claim 87, wherein applying the electrical signal comprises applying an electrical potential difference between the two electrodes.

93. The device of claim 87, wherein the cell population comprises one or more different bacterial strains.

94. The device of claim 87, wherein the cell population comprises one or more different yeast strains.

95. The device of claim 87, wherein the cell population comprises one or more different mammalian cell strains.

96. The device of claim 87, wherein the cell population comprises one or more different insect cell strains.

97. The device of claim 87, wherein the growth medium is selected from the group consisting of a liquid medium and a solid medium.

98. The device of claim 97, wherein the solid medium comprises a hydrogel.

99. The device of claim 97, wherein the solid medium comprises at least one gel-based material.

100. The device of claim 87, further comprising maintaining the cell population in a chamber, and flowing the growth medium through the chamber.

101. The device of claim 100, further comprising measuring the electrical parameter in different portions of the flowing growth medium as a function of time.

102. The device of claim 87, wherein the control gene is activated by at least one mechanism selected from the group consisting of: an external inducer; a self-activated inducer;

and a combination of an external inducer and a self-activated inducer.

103. The device of claim **102**, wherein the external inducer is selected from the group consisting of: exposure to optical radiation, a change in temperature, a chemical reagent, a viral agent, and combinations thereof.

104. The device of claim **87**, wherein measuring the electrical parameter comprises measuring at least member selected from the group consisting of: a cyclic voltammetry signal for the growth medium; a potentiometry signal for the growth medium; an electrical conductivity of the growth medium; a phase difference between electrical signals measured for the growth medium; and combinations thereof.

105. The device of claim **87**, wherein the electrical signal is an alternating current (AC) electrical signal.

106. The device of claim **87**, wherein the electrical signal is a direct current (DC) electrical signal.

107. The device of claim **87**, wherein the control gene is a single-gene lysis gene, a multi-gene lysis gene, one or more TA module genes, one or more toxin genes, or one or more peptide toxin gene.

108. The device of claim **87**, wherein the control gene is activated by a target analyte.

109. The device of claim **108**, wherein the target analyte comprises a chemical species.

110. The device of claim **108**, wherein the target analyte comprises a biochemical molecule.

111. The device of claim **109** or claim **110**, further comprising determining a relative concentration of the target analyte in the growth medium as a function of time based on the information about the cell population.

112. The device of claim **87**, wherein the information about the cell population comprises information about a relative concentration of at least one metabolite generated by the cell population.

113. The device of claim **87**, comprising measuring the electrical parameter of the growth medium without introducing a redox active species into the growth medium.

114. The device of claim **87**, wherein the reservoir is formed in a microfluidic device.

115. The device of claim **114**, wherein the reservoir comprises an inlet and an outlet, and wherein the device comprises a fluidic device configured to transport the growth medium through the reservoir from the inlet to the outlet.

116. The device of claim **87**, wherein the reservoir comprises a recess configured to support a solid growth medium.

* * * * *