United States Patent	12385000
Kind Code	B2
Date of Patent	August 12, 2025
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Cell culture analyzer and cell culture analysis method using same, additive supply unit and cell culture analyzer provided therewith, and sensor unit and cell culture analyzer provided therewith

Abstract

A cell culture analyzer comprises a stirring member and an air discharge and intake unit. The stirring member is used in a state of being immersed in a medium, and has a liquid discharge and intake port for discharging or drawing in the medium, and an air discharge and intake port for discharging or drawing in air in order to discharge or draw in the medium from the liquid discharge and intake port. The air discharge and intake unit is linked to the air discharge and intake port of the stirring member, and discharges or draws in the air discharged or drawn in from the air discharge and intake port.

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Family ID:	**1000008752020**
Appl. No.:	**17/791675**
Filed (or PCT Filed):	**March 09, 2021**
PCT No.:	**PCT/JP2021/009340**
PCT Pub. No.:	**WO2021/193029**
PCT Pub. Date:	September 30, 2021

Prior Publication Data

Document Identifier **Publication Date**
US 20230029803 A1 Feb. 02, 2023

Foreign Application Priority Data

JP	2020-054308	Mar. 25, 2020
JP	2020-054315	Mar. 25, 2020
JP	2020-054356	Mar. 25, 2020
JP	2020-054415	Mar. 25, 2020
JP	2020-054464	Mar. 25, 2020
JP	2021-035928	Mar. 08, 2021

Publication Classification

Int. Cl.: **C12M1/34** (20060101); **C12M1/00** (20060101); **C12M1/06** (20060101)

U.S. Cl.:

CPC **C12M41/30** (20130101); **C12M23/34** (20130101); **C12M27/02** (20130101); **C12M29/06** (20130101); **C12M29/24** (20130101);

Field of Classification Search

CPC: C12M (41/30); C12M (41/14); C12M (41/00); C12M (23/34); C12M (23/12); C12M (27/02); C12M (29/06); C12M (29/24); C12M (29/14)

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Background/Summary

TECHNICAL FIELD

(1) The present invention relates to a cell culture analyzer used to analyze cell culture, and to a cell culture analysis method in which this analyzer is used. The present invention also relates to an additive supply member used to analyze cell culture, and to a cell culture analyzer comprising this additive supply member. Furthermore, the present invention relates to a sensor unit used to analyze cell culture, and to a cell culture analyzer comprising this sensor unit.

BACKGROUND ART

(2) The configuration of a conventional cell culture analyzer comprises a sensor immersed in a medium in a culture vessel, a stirring member having a plunger immersed in the medium, and a drive means linked to the stirring member (for example, see Patent Literature 1).

(3) Also, a conventional cell culture analyzer comprised a sensor immersed in a medium in a culture vessel, and an additive supply means for supplying the additive to the medium, wherein the additive supply means had an additive container with an opening for supplying the medium into the culture vessel, and an air pressure supply means for applying air pressure into the additive container (see, for example, Patent Literature 2).

(4) Furthermore, the configuration of a conventional cell culture analyzer comprised a board provided with a plurality of openings, sensors disposed in these openings, and an additive container (see, for example, Patent Literature 2).

(5) That is, with a conventional cell culture analyzer, in performing cell culture analysis, the electrode portion of the sensor was immersed from above in a state in which the medium and the cells had been put in a plurality of culture vessels disposed side by side under the board, and the additive was added dropwise from the additive container into the culture vessel at a predetermined timing in the progress of the cell culture.

(6) Furthermore, a conventional cell culture analyzer comprised a sensor immersed in a medium in a culture vessel, and an additive addition member for supplying the additive to the medium. The additive addition member had an additive container with an opening for supplying the additive into the culture vessel, and an air pressure supply member for applying air pressure into the additive container. The additive container had a cylindrical shape having the opening underneath, and the lower portion of the cylindrical additive container was formed so that its outer peripheral diameter decreased toward the lower end (see, for example, Patent Literature 3).

(7) With the configuration of a conventional cell culture analyzer, the sensor was fixed to a through-hole portion provided to the board, and a lead wire for extracting a signal was connected to

this sensor.
(8) More specifically, with a conventional cell culture analyzer, a sensor for monitoring the state of the medium was inserted into the cell culture vessel, this sensor was provided with an electrical connection terminal, and a lead connected to this connection terminal was connected to an external control unit (see, for example, Patent Literature 4).
(9) A cell culture analyzer having a cartridge that mates with a plate provided with a plurality of cell culture vessels has also been disclosed.
(10) This cell culture analyzer has a sensor that takes measurements inside each culture vessel, a plurality of openings into which these sensors are inserted are provided to a cartridge, and the sensor and a fiber cable are connected in each opening. These fiber cables are connected to an external control unit (see, for example, Patent Literature 5).
CITATION LIST
Patent Literature
(11) Patent Literature 1: U.S. Pat. No. 6,326,058 Patent Literature 2: U.S. Pat. No. 10,359,418 Patent Literature 3: U.S. Pat. No. 9,494,577 Patent Literature 4: JP-A 2004-112092 Patent Literature 5: U.S. Pat. No. 9,170,255
SUMMARY
Technical Problem
(12) In the above-mentioned conventional example, the stirring member is constituted by a stirring rod whose lower end side is immersed in the medium. The medium in the culture vessel is stirred by moving the stirring rod up and down with a plunger. With a cell culture analyzer such as this, since a plurality of culture vessels are disposed adjacent to each other, a plurality of plungers corresponding to the plurality of culture vessels are also needed. As a result, a problem is that the cell culture analyzer ends up being larger.
(13) In view of this, it is an object of the present invention to provide a cell culture analyzer that can be made more compact.
(14) Also, in the above-mentioned conventional configuration, air pressure is applied to the additive container by the air drawn in by the air pressure supply member, and the additive in the additive container is supplied into the medium.
(15) However, with a conventional configuration, the air drawn in by the air pressure supply member is not controlled at all outside of the cell culture analyzer, and if this air should flow into the culture vessel through the additive container, it could contaminate cell culture within the culture vessel.
(16) In view of this, it is an object of the present invention to provide an additive supply member with which contamination of cell culture within a culture vessel can be prevented, as well as a cell culture analyzer comprising this additive supply member.
(17) Furthermore, in the conventional example discussed above, a plurality of culture vessels are disposed side by side under a board, but when cell culture analysis is performed, the medium and cells may not go into all of the culture vessels. If this should happen, the additive container will also be disposed above the unused cell culture vessels.
(18) Here, air pressure is applied to the upper opening of the additive container in order for the additive in the additive container to be added dropwise into the culture vessel, but if the unnecessary additive containers are not filled with additives, the air pressure applied to the plurality of additive containers will end up flowing into the empty additive container.
(19) As a result, the appropriate level of air pressure will not be applied to the required additive containers, and it may be impossible for the additives to be added properly.
(20) In view of this, conventionally, additive containers that were not to be used would be filled with some liquid other than an additive, such as water, which prevented the above-mentioned air leakage.
(21) Nevertheless, filling the unused additive containers with liquid entailed a tremendous amount

of work, so efficiency suffered.
(22) Therefore, it is an object of the present invention to provide a sensor unit with which efficiency can be improved regarding cell culture analysis, as well as a cell culture analyzer comprising this sensor unit.
(23) Furthermore, with the conventional configuration discussed above, air pressure is applied to the additive containers by the air drawn in by the air pressure supply member, so that the additives in the additive containers were supplied into the medium through the lower openings.
(24) However, with a conventional additive container, the outer peripheral diameter of the cylindrical lower portion decreases toward the lower end, and the opening in the lower end portion is provided to the pointed portion.
(25) Accordingly, when air pressure is applied to the additive container, the additive flows out continuously from the opening at the lower end and is continuously supplied to the culture vessel. However, this continuous inflow of additive causes a sudden environmental change for the cells contained in the culture vessel, and this can cause unfavorable stress on the cells. That is, with a conventional configuration, there is a possibility that the cells in the medium will be subjected to stress and that cell culture analysis cannot be carried out properly.
(26) In view of this, it is an object of the present invention to provide an additive supply member with which cell culture analysis can be carried out properly, as well as a cell culture analyzer comprising this additive supply member.
(27) With the conventional configuration discussed above, the sensor is immersed in a medium in a culture medium, for example, and senses the environment in this medium.
(28) With a cell culture device, a plurality of culture vessels are usually used when analyzing cell culture, and therefore the number of sensors corresponding to these containers also increases. Accordingly, there is a need to make a cell culture analyzer more compact.
(29) In view of this, it is an object of the present invention to provide a sensor unit with which a cell culture analyzer can be made more compact, as well as a cell culture analyzer comprising this sensor unit.

Solution to Problem

(30) To achieve the stated object, the cell culture analyzer of the present invention is a cell culture analyzer that performs cell culture analysis by detecting a specific component contained in a medium that has been put in a culture vessel, and comprises a stirring member and an air discharge and intake unit. The stirring member has a liquid discharge and intake port that is used in a state of being immersed in the medium and discharges or draws in the medium, and an air discharge and intake port that discharges or draws in air in order to discharge or draw in the medium from the liquid discharge and intake port. The air discharge and intake unit is linked to the air discharge and intake port of the stirring member and discharges or draws in air that is discharged or drawn in through the air discharge and intake port.
(31) Also, to achieve the stated object, the additive supply member of the present invention is an additive supply member that is used in a cell culture analyzer and that supplies an additive to a medium in a culture vessel, the member comprising an additive container and an air pressure supply unit. The additive container has an opening at the lower end portion and supplies an additive into the culture vessel. The air pressure supply unit has an air inlet for drawing in the air inside a culture incubator that houses the culture vessel, and applies air pressure into the additive container.
(32) Furthermore, to achieve the stated object, the sensor unit of the present invention comprises a board, additive containers, and a seal. The board is provided with a plurality of openings that match up with the positions of a plurality of culture vessels to which additives have been supplied. The additive containers are disposed in each of the plurality of openings of the board, have an upper surface opening that opens upward, and supply the additives to the culture vessel. The seal is affixed so as to cover the upper surface of the board corresponding to the upper surface openings in the additive container in a removable state.

(33) Furthermore, to achieve the stated object, the additive supply member of the present invention is an additive supply member for adding an additive to a medium in a culture vessel, the member comprising an additive container and an air pressure supply member. The additive container has an opening for adding the additive into the culture vessel. The air pressure supply member applies air pressure into the additive container. The additive container has a cylindrical shape with the opening at the bottom, and the outside diameter thereof decreases toward the lower end, and has a dropping adjustment surface that is provided on the outer peripheral edge of the opening and adjusts the amount in which the additive is discharged from the opening.
(34) To achieve the stated object, the sensor unit of the present invention is a sensor unit having a sensor for measuring the components of a culture medium in a culture vessel, comprising a sensor and a board. The sensor has a main body portion, a measurement unit that is disposed on the main body portion and measures the components of the medium, and a connection terminal portion that is electrically connected to the measurement unit. The board has a connecting portion that is connected to the connection terminal portion of the sensor, and a wiring pattern that is connected to the connecting portion. The sensor has a bent part in which a part of the main body portion is bent so that the measurement portion of the sensor projects downward in a state in which the connection terminal portion and the connecting portion of the board are connected.
Technical Effects
(35) The cell culture analyzer of the present invention does not require a stirring rod or plunger to be provided for each culture vessel, and therefore can be more compact.
(36) Also, with the additive supply member according to the present invention, the air in the culture incubator that houses the culture vessel, that is, the controlled air, is utilized as the air pressure going to the additive vessel, which prevents contamination of the cell culture in the culture vessel.
(37) Furthermore, with the cell culture analyzer of the present invention, since the opening in the board portion corresponding to the upper surface of the unused culture vessel is covered with the seal, no air will leak out through the additive containers located in this opening, and the proper air pressure can be applied to the required additive containers, allowing the proper additives to be supplied.
(38) Also, since cell culture analysis can be executed by the simple operation of removing the seal from the board portion corresponding to the upper surface of the culture vessels to be used, work efficiency can be improved.
(39) Furthermore, with the additive supply member of the present invention, when air pressure is applied to an additive container, the additive held in the additive container moves to the lower surface opening side, resulting in a clumped state in which the additive is held by the surface tension of the dropping adjustment surface provided on the outer peripheral edge of the opening. After this, the additive is dropped into the lower culture vessel as droplets that have overcome the holding force produced by surface tension.
(40) Also, when this dropping occurs, the next additive also ends up in the above-mentioned clumped state, and before long is dropped as droplets into the culture vessel below.
(41) That is, in the present invention, since the additive is supplied intermittently into the culture vessel, the cells are less likely to be subjected to sudden stress by the additive, and as a result, the cell culture analysis is carried out properly.
(42) With the sensor unit of the present invention, there is no need for a configuration in which a lead wire is directly connected to the sensor, which affords a more compact size.

Description

BRIEF DESCRIPTION OF TUE. DRAWINGS
(1) FIG. **1** is a diagram showing the configuration of the cell culture analyzer according to an

embodiment of the present invention;
(2) FIG. **2** is a diagram showing the state when the analysis unit of the cell culture analyzer in FIG. **1** is installed in a culture incubator;
(3) FIGS. **3**A and **3**B are diagrams showing the configuration of a drive unit included in the cell culture analyzer in FIG. **1**;
(4) FIG. **4** is a cross-sectional view showing the configuration of a multi-directional switching valve included in the drive unit in FIG. **3**A;
(5) FIG. **5** is a diagram showing the configuration of an analysis unit included in the cell culture analyzer in FIG. **1**;
(6) FIG. **6** is a diagram showing the state when the adapter unit constituting the analysis unit in FIG. **5** is attached between a top unit and a bottom unit;
(7) FIG. **7**A is a diagram showing the configuration of the adapter unit in FIG. **6**, and FIG. **7**B is a diagram showing the configuration of a board unit installed in the adapter unit in FIG. **7**A;
(8) FIG. **8** is an exploded oblique view showing the configuration of a board unit included in an adapter unit disposed on a sensor unit;
(9) FIG. **9** is an oblique view showing the connection state between the board unit and a piping tube;
(10) FIG. **10** is a diagram showing the configuration of an intake port used as an air pressure supply unit;
(11) FIGS. **11**A to **11**C are diagrams showing the routing of pipes formed in a piping board portion;
(12) FIG. **12** is an exploded oblique view showing the configuration of an adapter unit;
(13) FIGS. **13**A and **13**B are diagrams showing the configuration of a sensor disposed in a sensor unit;
(14) FIG. **14** is an exploded oblique view showing the configuration of a sensor unit;
(15) FIGS. **1**.**5**A to **15**C are diagrams illustrating a method for fixing and holding a sensor;
(16) FIG. **16**A is a diagram showing a state in which a plurality of sensors have been placed on a bottom plate, and FIG. **16**B is a detail view of the A portion in FIG. **16**A;
(17) FIG. **17**A is a diagram showing the state when a middle plate has been put over a plurality of sensors placed on the bottom plate, while FIG. **17**B is a detail view of the B portion in FIG. **17**A;
(18) FIG. **18**A is a diagram showing a state in which the middle plate is slid in a direction substantially parallel to the diagonal line of the bottom plate with respect to the bottom plate, and FIG. **18**B is a detail view of the C portion in FIG. **18**A;
(19) FIG. **19**A is a top view of the state when the middle plate has been fixed to the bottom plate, and FIG. **19**B is a cross-sectional view along the A-A′ line;
(20) FIG. **20**A is a diagram of a sensor positioned and fixed while sandwiched between the bottom plate and the middle plate in a state in which the upper portion of a vertical side has been bent further downward from a bent portion, FIG. **20**B is an oblique view of the state when the top plate is disposed on the middle plate as seen from under the bottom plate, and FIG. **20**C is a cross-sectional view of the cross-sectional structure thereof;
(21) FIG. **21**A is a top view of a gasket sheet, and FIG. **21**B is a detail view of a port input/output portion of the gasket sheet in FIG. **21**A;
(22) FIG. **22** is a cross-sectional view along the E-E′ line of the port input/output portion shown in FIG. **21**B;
(23) FIG. **23** is a cross-sectional view showing the state when a board unit is incorporated into a sensor unit on the upper surface side;
(24) FIG. **24** is a top view of a well plate;
(25) FIG. **25**A is an oblique view of a port for adding additives to the wells, as seen from below, and FIG. **25**B is an oblique view of the port as seen from above;
(26) FIG. **26**A is a top view of the port, and FIG. **26**B is a cross-sectional view along the F-F′ line in FIG. **26**A;

(27) FIG. **27** is a detail cross-sectional view showing the configuration in the vicinity of the discharge port of an additive A;
(28) FIGS. **28**A to **28**C are cross-sectional views showing the additive as it is dropped from the lower end portion of an additive A container of an additive addition portion A;
(29) FIG. **29** is a plan view of the additive A discharge port side of the additive addition portion A (additive A container);
(30) FIG. **30**A is a diagram of when an additive is loaded into the additive A container as an initial step, FIG. **30**B is a diagram showing the state after the additive has been loaded, and FIG. **30***c* is a detail view of the additive A discharge port when the additive is loaded;
(31) FIG. **31**A is a diagram showing the state when a piping board portion has been linked from above after loading the additive, and FIG. **31**B is a diagram showing the state when an additive has been added into a well;
(32) FIG. **32**A is a top view of a port including a stirring member, FIG. **32**B is a cross-sectional view along the G-G′ line, and FIG. **32**C is a detail oblique view of a stirring member discharge and intake port provided at the lower end portion of the stirring vessel of the stirring member;
(33) FIG. **33**A is a diagram showing the initial state of the stirring member, FIG. **33**B is a diagram showing the state when an air discharge and intake unit is linked to the stirring member, and FIG. **33**C is a diagram showing the state when the air discharge and intake unit acts in the direction of discharging air;
(34) FIG. **34** is a plan view showing the state when the medium discharged from the stirring vessel is stirred along the inner peripheral surface of the culture vessel (well);
(35) FIG. **35**A is a diagram showing the configuration of the stirring member, including a liquid discharge and intake port in which no undercut is required, and FIG. **35**B is a detail oblique view of a portion of the liquid discharge and intake port;
(36) FIG. **36**A is a flowchart showing an analysis method including an addition step and a measurement step, including a stirring treatment and a homogenization treatment, FIG. **36**B is a flowchart showing the flow processing in addition steps A and B included in FIG. **36**A, and FIG. **36**C is a flowchart showing a process flow of the measurement processing included in FIG. **36**A;
(37) FIG. **37** is an exploded oblique view of a seal affixed to the upper surface of the sensor unit;
(38) FIG. **38**A is a plan view of a sensor unit, FIG. **38**B is a plan view of the configuration of the top seal affixed to the upper surface of the sensor unit, FIG. **38**C is a plan view of the configuration of the bottom seal affixed to the upper surface of the sensor unit, and FIG. **38**D is a plan view showing a state in which the top seal and the bottom seal have been affixed to the upper surface of the sensor unit;
(39) FIG. **39**A is a plan view showing a state in which a seal provided to the user has been affixed, FIG. **39**B is a plan view showing a state in which the top seal has been peeled away from the state in FIG. **39**A, and FIG. **39**C is a plan view showing a state in which the bottom seal has been peeled away from the state in FIG. **398**;
(40) FIG. **40** is a diagram showing a state in which the seal is steadily peeled off as the product is used;
(41) FIG. **41**A is a top view of a port including a stirring member according to Embodiment 2 of the present invention, and FIG. **41**B is a cross-sectional view along the G-G′ line;
(42) FIG. **42**A is a detail view showing the configuration in the vicinity of a liquid discharge and intake port of the stirring member, and FIG. **42**B is a cross-sectional view showing the internal structure in FIG. **42**A;
(43) FIG. **43**A is a top view of a port including a stirring member according to Embodiment 3 of the present invention, and FIG. **43**B is a plan view of the flow of the culture medium discharged from the stirring member;
(44) FIG. **44**A is a lateral cross-sectional view showing a state in which a port including a stirring member according to Embodiment 4 of the present invention is immersed in a culture medium in a

culture vessel (well), and FIG. **44**B is a lateral cross-sectional view showing the remaining medium left behind in the stirring vessel when the port in FIG. **44**A has been pulled up out of the medium;
(45) FIG. **45**A is a lateral cross-sectional view showing a state in which a port including a stirring member according to Embodiment 5 of the present invention is immersed in a culture medium in a culture vessel (well), and FIG. **45**B is a lateral cross-sectional view showing the remaining medium left behind in the stirring vessel when the port in FIG. **45**A has been pulled up out of the medium;
(46) FIG. **46** is a detail cross-sectional view showing a rib formed in the stirring vessel of the stirring member in FIG. **45**A, etc.; and
(47) FIG. **47**A is a lateral cross-sectional view showing a state in which a port including a stirring member according to Embodiment 6 of the present invention is immersed in a culture medium in a culture vessel (well), and FIG. **47**B is a detail view showing the vicinity of the liquid discharge and intake port.

DESCRIPTION OF EMBODIMENTS

Embodiment 1

(48) The cell culture analyzer **1** according to an embodiment of the present invention will now be described with reference to the appended drawings.
(49) Summary of Cell Culture Analyzer **1**
(50) FIG. **1** shows the configuration of the cell culture analyzer **1**.
(51) The cell culture analyzer **1** is a device that electrochemically senses the concentration of a specific component contained in a medium in a state in which a part (sensing electrode) of a sensor **43** is immersed in the medium (liquid) contained in a culture vessel, and comprises an analysis unit **2**, a drive unit **3** (serving as an air pressure supply unit), and a control unit **4** that controls the analysis unit **2** and the drive unit **3**. The control unit **4**, the analysis unit **2**, and the drive unit **3** are connected by an electrical cable **5**. The drive unit **3** and the analysis unit **2** are connected by a piping tube **6**.
(52) FIG. **2** shows a usage example of the cell culture analyzer **1** disposed in a culture incubator **7**.
(53) The analysis unit **2** of the cell culture analyzer **1** is disposed in the culture incubator **7**. The control unit **4** connected to the analysis unit **2** by the electrical cable **5**, and the drive unit **3** connected to the analysis unit **2** by the piping tube **6**, are disposed outside the culture incubator **7**.
(54) Consequently, the user can analyze the culture state in the culture incubator **7** via the control unit **4** without having to open and close the door **8** of the culture incubator **7**. That is, in analyzing the culture state, air contamination inside the culture incubator **7** can be prevented.
(55) FIGS. **3**A and **3**B show the configuration of the drive unit **3**.
(56) The drive unit **3** is an air pressure supply unit for the analysis unit **2**, and as shown in FIGS. **3**A and **3**B, has a syringe **9**, a plunger **10**, a multi-directional switching valve **11**, a plunger motor **12**, and a valve motor **13**. The air pressure is adjusted by compressing or drawing in the air in the syringe **9** with the plunger **10**. The plunger **10** is linked to the multi-directional switching valve **11**.
(57) The plunger motor **12** and a motor **13** for the multi-directional switching valve **11** are disposed in the housing **3***a* of the drive unit **3**. These motors **12** and **13** are controlled by the control unit **4**, which is connected via the electrical cable **5**.
(58) FIG. **4** shows the configuration of the multi-directional switching valve **11** included in the drive unit **3**.
(59) The multi-directional switching valve **11** has a valve **14** for the additive addition portion A, a valve **15** for the additive addition portion B, and a valve **16** for the stirring member, as air supply system valves for the analysis unit **2**.
(60) The multi-directional switching valve **11** has a stirring member valve **16** and an intake valve **17**, as system valves for the analysis unit **2**.
(61) The multi-directional switching valve **11** controls the rotation of the rotating portion **18** to determine the position of a rotating flow path **19** in the circumferential direction, and is controlled so as to connect the flow path of a specific valve with the syringe **9** to supply air pressure.

(62) More specifically, in the supply of air to the analysis unit **2**, first, the rotation of the rotating portion **18** is controlled to connect the flow path of the intake valve **17** and the syringe **9**. Then, the plunger **10** is pulled in the intake direction, and air is drawn into the syringe **9** through the intake valve **17**. Next, the rotation of the rotating portion **18** is controlled so that the flow path of the syringe **9** is connected to the valves **14**, **15**, and **16** of a specific air supply system, and then the plunger **10** is depressed in the compression direction to send air to the specific valves **14**, **15**, and **16**,

(63) FIG. **5** shows the configuration of the analysis unit **2**.

(64) The analysis unit **2** is designed to be short in the horizontal direction, low in the height direction, and long in the depth direction so that a plurality of units can be installed in the culture incubator. The reason for this is that the culture space of a typical culture incubator is long in the depth direction and low in the height direction, and this shape is suitable for this.

(65) The analysis unit **2** has an adapter unit **20**, a top unit **21**, and a bottom unit **22**, and is configured to sandwich the adapter unit **20** between the top unit **21** and the bottom unit **22**.

(66) As shown in FIG. **6**, the adapter unit **20** is attached by being slid from a front opening **23** formed between the top unit **21** and the bottom unit **22**. As a result, the height of the analysis unit **2** can be kept low.

(67) Also, as shown in FIG. **6**, the adapter bottom **24**, the well plate **25**, the adapter top **26**, and the sensor unit **27** are disposed in the adapter unit **20** in that order, starting from the bottom.

(68) The top unit **21** of the adapter unit **20** shown in FIG. **7**A includes the board unit **28** shown in FIG. **7**B.

(69) FIG. **8** shows an exploded oblique view of the board unit **28** disposed on the sensor unit **27**. As shown in FIG. **8**, a piping board portion **29**, a board base **30**, and a board **31** are disposed in the board unit **28** is that order from below the side opposite the sensor unit **27**.

(70) The piping board portion **29** includes an air pipe to which an air flow path from the drive unit **3** is connected. The board base **30** is provided so that the board **31** is attached to the upper surface thereof. The board **31** is provided with a connecting portion **32** for electrical connection to an electrochemical sensor **43** (see FIG. **14**, etc.) provided to the sensor unit **27** below.

(71) A plurality of connecting portions **32** are disposed facing downward from the board **31**, go through contact through-holes **30***a* disposed in the board base **30**, pass through the piping hoard portion **29**, and are electrically connected to a plurality of sensors **43** disposed at corresponding positions in the lower sensor unit **27**.

(72) A wiring pattern electrically connected to the connecting portions **32** is provided on the board **31**. The board **31** is connected to the external control unit **4** (see FIG. **1**, etc.) via the electrical cable **5**.

(73) FIG. **9** shows the connection state: between the hoard unit **28** and the piping tubes **33**, **34**, **35** and **36**.

(74) In this embodiment, a total of four kinds of piping tubes connected to the drive unit **3** are connected to the board unit **28**.

(75) More specifically, the board unit **28** is provided with an additive addition portion A piping tube **33** and an additive addition portion B piping tube **34**, as piping tubes for the air supply system to the board unit **28**.

(76) The board unit **28** is further provided with an intake piping tube **36**, as an intake system valve for the analysis unit **2**.

(77) The stirring member piping tube **35** is provided to the board unit **28** as a bi-directional valve for air supply and intake.

(78) FIG. **10** shows the configuration of an intake port used as an air pressure supply unit.

(79) The air pressure supply unit has an air inlet (intake port) **37** for drawing in the air inside the culture incubator **7** that houses the culture vessels.

(80) More specifically, the air inlet (intake port) **37** is provided on the lower bottom surface of the

piping board portion **29**. The air inlet (intake port) **37** goes through the through-hole **38** in the piping board portion **29**, and is connected to the multi-directional switching valve **11** of the drive unit **3** via the piping tube **36** that is linked to the piping tube connecting portion **39** above.
(81) Consequently, since the air pressure supply unit has the air inlet (intake port) **37** for drawing in the air that is inside the culture incubator **7** housing the culture medium vessel, contamination of the cell culture in the culture vessel can be prevented.
(82) In other words, in this embodiment, the air inside the culture incubator **7** housing the culture medium container, that is, controlled air, is utilized as air pressure to the additive containers (additive A container **85**, additive B container **86**) and the stirring member **81**. This prevents the contamination of the cell culture in the culture vessel.
(83) Also, since the air inlet (intake port) **37** is provided on the lower bottom surface of the piping board portion **29**, water droplets and the like can be prevented from flowing into through the opening of the air inlet **37**.
(84) Also, the piping tube **36** is formed from a moisture-permeable material such as a Nafion tube. This prevents the water in the culture incubator **7** from flowing into the drive unit **3**, and prevents condensation in the drive unit **3**.
(85) FIGS. **11**A to **11**C show the routing of the piping formed on the piping board portion **29**.
(86) The piping tube **33** for the additive addition portion A is connected to the piping board portion **29**. The culture vessel (well plate **25**) in this embodiment contains 24 wells **80**. Therefore, the piping for the additive addition portion A branches off into 24 parallel pipes, and the outlet openings of the pipes are disposed above specific wells **80**.
(87) Similarly, the piping tube **34** for the additive addition portion B is connected to the piping board portion **29**. The piping for the additive addition portion B branches off into 24 parallel pipes, and the outlet openings of the pipes are disposed above specific wells **80**.
(88) Similarly, the piping tube **35** for the stirring member is connected to the piping board portion **29**. The piping for the stirring member branches off into 24 parallel pipes, and the outlet openings of the pipes are disposed above specific wells **80**.
(89) That is, the same air pressure is applied all at once to the additive addition portions A of all 24 of the wells **80** of the culture vessel. Similarly, the same air pressure is applied all at once to the additive addition portions B of all 24 of the wells **80**. Similarly, the same air pressure is applied all at once to the stirring members of all 24 of the wells **80**.
(90) FIG. **12** shows the configuration of the adapter unit **20**.
(91) As shown in FIG. **12**, the adapter unit **20** has an adapter bottom **24** (as a culture vessel installation portion), a well plate **25** (as a culture vessel), an adapter top **26**, and a sensor unit **27**, placed in that order starting from the bottom.
(92) In this embodiment, the well plate **25** has 24 wells **80** in a 4×6 matrix. The adapter top **26** is provided in order to adjust the height of the well plate **25**, and a different adapter top **26** is used according to the height of the well plate **25**. The reason for this is to adjust the height relationship between the sensor unit **27** and the well plate **25** when the sensor unit **27** is placed on the adapter top **26**.
(93) There are several types of well plate **25**, including general-purpose types, and the proper adapter top **26** is used according to this type.
(94) In the sensor unit **27** disposed on the adapter top **26**, four leg portions (supports) **40** provided on the lower surface side thereof go through through-holes **41** in the adapter top **26** below, and are inserted into positioning holes **42** provided in the adapter bottom **24** serving as a culture vessel installation portion.
(95) Consequently, the sensor unit **27** is installed on the well plate **25** spaced apart at a specific gap. That is, the sensor unit **27** is provided with leg portions **40** for ensuring a housing space for the well plate **25** (culture vessel) on the adapter bottom **24**. The sensor unit **27** is disposed on the adapter bottom **24** in a state of being supported by the leg portions **40**.

(96) As discussed above, the leg portions **40** support the sensor unit **27** with respect to the adapter bottom **24** in order to ensure a housing space for the well plate **25** (culture vessel) (a gap between the upper surface of the adapter bottom **24** and the lower surface of the sensor unit **27**).
(97) Here, the supports that support the sensor unit **27** is not limited to the leg portions **40** provided to the sensor unit **27**. For instance, the supports may be provided on the adapter bottom **24** side, so long as they support the sensor unit **27** from below with respect to the adapter bottom **24**.
(98) FIGS. **13**A and **13**B show the configuration of the sensor **43** disposed in the sensor unit **27**.
(99) As shown in FIGS. **13**A and **13**B, the sensor **43** in this embodiment has a main body portion **43***a* that is substantially L-shaped, except for the upper portion where electrode pads **52** to **55** are disposed. The sensor **43** has a bent portion **44** that is bent during use, at the upper part of a vertical edge of the substantially L-shaped main body portion **43***a*.
(100) The bent portion **44** is a portion in which the other substantially L-shaped portions (lateral edge portion **45** and vertical edge portion **46**) are bent at a substantially right angle with respect to the upper portion where the electrode pads **52** to **55** are disposed. Thus bending the main body portion **43***a* at a substantially right angle at the bent portion **44** allows the sensing electrodes **47**, **48**, **49**, and **50** of the sensor **43** to be immersed in the wells **80**.
(101) A notched portion **44***a* is located near the bent portion **44**, and is formed at the portion where the upper portion where the electrode pads **52** to **55** are disposed is linked to the substantially L-shaped portion (lateral edge portion **45** and vertical edge portion **46**). The notched portion **44***a* is formed by a cutout formed in the lengthwise direction of the vertical edge portion **46**.
(102) This allows the substantially L-shaped portion (lateral edge portion **45** and vertical edge portion **46**) to be moved in the lengthwise direction of the vertical edge portion **46** without the crease formed in bending with respect to the upper portion where the electrode pads **52** to **55** are disposed being limited to the portion where the vertical edge portion **46** is connected to the upper portion where the pads **52** to **55** are disposed.
(103) Consequently, the portion that because a crease formed in the sensor **43** can be moved according to the positional relationship, such as the size of the sensor **43** and the depth of the wells **80**.
(104) In this embodiment, the sensor **43** is substantially L-shaped, and the culture state in the culture vessel is sensed by placing the lateral edge portion **45** of the sensor in the wells **80** of the culture vessel and holding this portion in a horizontal state.
(105) Also, sensing electrodes **47** to **50** for sensing the culture state in the culture vessel are provided on the lower lateral edge portion **45** of the sensor **43**.
(106) This allows the electrode surface area of the sensing electrodes **47** to **50** to be larger, which improves the sensitivity of the sensor **43**. The horizontal width of the lower lateral edge portion **45** of the sensor **43** is greater than the horizontal width of the upper vertical edge portion **46**.
(107) The shape of the sensor **43** is not limited to substantially L-shaped, and may, for example, be substantially I-shaped, substantially inverted T-shaped, or the like. Also, in order to improve the sensitivity of the sensor **43**, the horizontal dimension (width) of the horizontal edge portion of the sensor **43** is preferably made wider.
(108) The lateral edge portion **45** of the sensor **43** is provided with a first working electrode **47**, a counter electrode **48**, a reference electrode **49**, and a second working electrode **50** as sensing electrodes.
(109) Also, a silver layer (a silver layer and/or a silver chloride layer is provided on the surface of the reference electrode **49**, A reagent layer formed from an enzyme and a mediator, etc., is provided on the surfaces of the first and second working electrodes **47** and **50**. The sensing electrode portions thereof are covered with a protective membrane **51**.
(110) The sensor **43** electrochemically senses the concentration of a specific component of the medium in a state in which the first working electrode **47**, the counter electrode **48**, the reference electrode **49**, and the second working electrode **50** are immersed in the medium in the culture

vessel.
(111) For instance, when sensing the concentration of the glucose component in the medium, the reagent layer immobilized on the surface of the first working electrode **47** contains an enzyme (such as GOx) and a redox mediator.
(112) The principle behind this glucose detection is that glucose that permeates from the medium through the protective membrane **51** is oxidized in an enzymatic reaction with an enzyme (such as GOx) in the reagent layer and turns into gluconolactone, and at the same time, the redox mediator in the reagent layer is reduced into a reductant. The glucose concentration in the medium can be measured by measuring, as a current value, the electrons generated when this reductant goes back to being an oxidant.
(113) The protective membrane **51** is provided to allow glucose in the medium to penetrate into the sensing electrode portion of the sensor **43** while limiting its permeation, and to prevent the components of the reagent layer (enzyme and mediator) immobilized on the first working electrode **47** from flowing to the outside of the protective membrane **51**.
(114) The enzyme and mediator are cross-linked and immobilized on electrodes. Therefore, the reagent layer is polymerized and its molecular weight increases. Consequently, glucose can permeate the protective membrane **51**, while the enzyme and mediator are prevented from flowing out of the protective membrane **51** (see WO 2019/146788 for details).
(115) The first working electrode **47**, the counter electrode **48**, the reference electrode **49**, and the second working electrode **50** are electrically connected to the electrode pads **52** to **55**, which are connection terminals above the sensor **43**. The electrode pads **52** to **55** have a first working electrode pad **52**, a counter electrode pad **53**, a reference electrode pad **54**, and a second working electrode pad **55**.
(116) A reagent for sensing lactic acid, for example, is immobilized on the second working electrode **50**.
(117) As shown in FIGS. **13**A and **13**B, the sensor **43** is configured such that sensing electrodes, which are measurement units (first working electrode **47**, counter electrode **48**, reference electrode **49**, second working electrode **50**), and connection terminal portions (first working electrode pad **52**, counter electrode pad **53**, reference electrode pad **54**, second working electrode pad **55**) are formed on the same board.
(118) A PET (polyethylene terephthalate) film, which is a resin material, is used as the base material, for example.
(119) The first working electrode **47**, the counter electrode **48**, the reference electrode **49**, and the second working electrode **50** are formed on the same board. Also, in the lateral edge portion **45** of the sensor **43**, the first working electrode **47**, the counter electrode **48**, the reference electrode **49**, and the second working electrode **50** are disposed substantially horizontally in their usage state. In order to increase the surface area of the working electrodes, a plurality of working electrodes **47** and **50** are disposed in left and right symmetry in the horizontal direction around the reference electrode **49**. As a result, the electrode surface area of the working electrodes **47** and **50** can be increased to raise the detection sensitivity.
(120) The method for manufacturing the sensor **43** will now be described.
(121) First, a gold electrode layer is formed by sputtering on the upper surface of a PET (polyethylene terephthalate) film, which is a resin material. Next, the electrode layer is patterned in an approximate L shape to match the sensor **43**. That is, the electrode layer is transpired with a laser, and thereby formed into a substantially L-shaped electrode layer.
(122) Further, the substantially L-shaped electrode layer is divided into a first working electrode **47**, a counter electrode **48**, a reference electrode **49**, and a second working electrode **50**. Signals are taken off from the four divided conductive paths at the connection terminal portions (first working electrode pad **52**, counter electrode pad **53**, reference electrode pad **54**, second working electrode pad **55**).

(123) After this substantially L-shaped electrode layer has been divided up for use as the first working electrode **47**, the counter electrode **48**, the reference electrode **49**, and the second working electrode **50**, a resist film **56** is provided in a state in which the electrode portions are masked off. After this, a silver layer (a silver layer and/or a silver chloride layer) is provided on the surface of the reference electrode **49**, and a reagent layer is provided on the surfaces of the first working electrode **47** and the second working electrode **50**.
(124) The first working electrode **47**, the counter electrode **48**, the reference electrode **49**, and the second working electrode **50** are covered by the protective membrane **51**.
(125) FIG. **14** shows an exploded oblique view of the sensor unit **27**.
(126) In the sensor unit **27**, a bottom plate **57**, a middle plate **58**, a top plate **59**, and a gasket sheet (board) **60** are stacked in this order, starting from the bottom.
(127) Then, the sensor **43** is fixed and held by the bottom plate **57**, the middle plate **58**, and the top plate **59**, and is bent substantially vertically downward.
(128) The upper portion of a port (additive supply member) **61** is fixed to the upper surface of the top plate **59**, and goes through the top plate **59**, the middle plate **58**, and the bottom plate **57**, and the lower portion is disposed below the bottom plate **57**.
(129) FIGS. **15**A to **15**C are diagrams illustrating the method for immobilizing and holding the sensor **43**.
(130) As shown in FIG. **15**A, first, a plurality (a 4×6 matrix) of sensors **43** are placed on the bottom plate **57**. The sensors **43** are placed in parallel with the diagonal line of the rectangular bottom plate **57** so that length of the sensors **43** in the lengthwise direction will be sufficient.
(131) Next, as shown in FIG. **15**B, the middle plate **58** is placed over the sensors **43** mounted on the bottom plate **57**.
(132) Next, as shown in FIG. **15**C, the middle plate **58** slides in a direction parallel to the diagonal line of the bottom plate **57** to fix the bottom plate **57** and the middle plate **58**. At this point, the sensors **43** are fixed and held by being sandwiched between the bottom plate **57** and the middle plate **58**.
(133) The method for fixing and holding the sensors **43** will be described in more specific terms with reference to FIGS. **16**A to **18**B.
(134) FIG. **16**A shows a state in which a plurality of sensors **43** have been placed on the bottom plate **57**, and is a diagram showing FIG. **15**A more specifically. FIG. **16**B is a detail view of the A portion in FIG. **16**A.
(135) FIG. **16**A is a top view showing a state in which a plurality of sensors **43** have been mounted on the bottom plate **57**, In this state, the sensors **43** are placed in parallel with the diagonal line of the rectangular bottom plate **57**. In FIG. **16**A, each sensor **43** is such that a connection terminal portion **62** is disposed in the upper right direction on the drawing, and a sensing electrode **66** (a measurement unit) is disposed in the lower left direction.
(136) As shown in FIG. **16**B, a positioning portion **67** that surrounds and fixes the connection terminal portion **62** of the sensor **43** from all four sides is provided on the bottom plate **57**. On the bottom plate **57**, at least one fixing portion **63** that mates with the middle plate **58** and fixes the middle plate **58** and the bottom plate **57** in the up and down direction is provided.
(137) Also, a slide guide protrusion **64** is provided on the bottom plate **57**. A through-hole **65** is provided on the bottom plate **57**, through which the sensor **43** is bent downward.
(138) FIG. **17**A is a diagram showing a state in which the middle plate **58** is placed over the sensors **43** mounted on the bottom plate **57**, and is a diagram showing FIG. **15**B more specifically. FIG. **17**B is a detail view of the B portion in FIG. **17**A.
(139) FIG. **17**A is a top view of a state in which the middle plate **58** has been placed over the sensors **43** mounted on the bottom plate **57**. In this state, the sensors **43** are placed in parallel with the diagonal line of the rectangular bottom plate **57**. In FIG. **17**A, the sensors **43** each have a connection terminal portion **62** disposed in the upper right direction of the drawing, and a sensing

electrode **66** (a measurement unit) disposed in the lower left direction.
(140) As shown in FIG. **17**B, the middle plate **58** is provided with a slide hole **68** with which a slide guide protrusion **64** provided to the bottom plate **57** is slidably mated, and a fixed portion **69** that mates with a fixing portion **63** of the bottom plate **57**. The middle plate **58** is put over the bottom plate **57** in a state in which the slide guide protrusion **64** provided on the bottom plate **57** has been passed through the slide hole **68**.
(141) Consequently, the middle plate **58** slides with respect to the bottom plate **57** in the direction in which the slide guide protrusion **64** slides along the slide hole **68**.
(142) FIG. **18**A is a more specific diagram of FIG. **15**C, showing a state in which the middle plate **58** is slid with respect to the bottom plate **57** in a direction substantially parallel to the diagonal line of the bottom plate **57**, so that the middle plate **58** is fixed to the bottom plate **57**. FIG. **18**B is a detail view of the C portion in FIG. **18**A.
(143) In the state shown in FIG. **18**A, the sensors **43** are placed in parallel with the diagonal line of the rectangular bottom plate **57**. In FIG. **18**A, the sensors **43** each have a connection terminal portion **62** disposed in the upper right direction of the drawing, and a sensing electrode **66** (a measurement unit) disposed in the lower left direction.
(144) As shown in FIG. **18**B, when the middle plate **58** is slid in a direction substantially parallel to the diagonal line of the bottom plate **57**, the bottom plate **57** and the middle plate **58** are fixed. In this state, the slide guide protrusion **64** provided to the bottom plate **57** slides along the slide hole **68**, and the fixing portion **63** of the bottom plate **57** is mated with the fixed portion **69** of the middle plate **58**.
(145) More specifically, the fixing portion **63** of the bottom plate **57** is a prong, and the fixed portion **69** of the middle plate **58** is a mating portion to which the prong is latched.
(146) As a result, the connection terminal portion **62** of the sensor **43** is sandwiched between the bottom plate **57** and the middle plate **58** from above and below and fixed in this positioned state.
(147) FIG. **19**A is a top view of the middle plate **58** fixed to the bottom plate **57**. FIG. **19**B is a cross-sectional view along the D-D′ line in FIG. **19**A.
(148) As shown in FIG. **19**B, the sensors **43** placed on the bottom plate **57** are positioned and fixed while being sandwiched between the bottom plate **57** and the middle plate **58**. Each of the sensors **43** is provided with one fixing portion **63** and one fixed portion **69**. The bottom plate **57** and the middle plate **58** are fixed in the horizontal direction and the height direction by the fixing portion **63** and the fixed portion **69** at a plurality of places. As a result, the sensors **43** are positioned and fixed while being sandwiched between the bottom plate **57** and the middle plate **58**.
(149) FIG. **20**A shows a state in which a sensor **43** that has been positioned and fixed while being sandwiched between the bottom plate **57** and the middle plate **58** is further bent downward from the bent portion **44** at the vertical edge upper part. FIG. **20**B is an oblique view of a state in which the top plate **59** is disposed on the middle plate **58**, as seen from below the bottom plate **57**, and FIG. **20**C is a cross-sectional view showing the cross-sectional structure thereof.
(150) In this embodiment, when the top plate **59** is placed over the upper surface of the middle plate **58** shown in FIG. **20**A, as shown in FIGS. **2013** and **20**C, the pressing portion **71** formed so as to protrude downward from the lower surface of the top plate **59** goes through the through-hole **65** and moves downward in a state of being in contact with the upper surface of the sensor **43**.
(151) At this point, the support portion **70** that supports the lower edge side of the bent portion **44** of the sensor **43** is provided at the opening edge of the through-hole **65** of the bottom plate **57**. The pressing portion **71** that pushes the upper edge side of the bent portion **44** of the sensor **43** downward is provided at a portion of the top plate **59** that is opposite the support portion **70**.
(152) Consequently, the upper surface of the sensor **43** is pushed down by the pressing portion **71**, so that the sensor **43** is bent from near the base of the vertical edge portion **46** and is supported from below by the support portion **70** provided on the upper surface side of the middle plate **58**.
(153) As shown in FIG. **20**C, the support portion **70** has an upper surface curved shape including a

curved surface on its upper surface. Also, as shown in FIG. **20**C, the pressing portion **71** has a lower surface curved shape including a curved surface on its lower surface.
(154) Consequently, as shown in FIGS. **20**B and **20**C, when the sensor **43** is sandwiched between the top plate **59** and the bottom plate **57** from above and below, the bent portion **44** of the sensor **43** is held in a state of being sandwiched from above and below by the support portion **70** and the pressing portion **71**.
(155) Therefore, the sensor **43** is bent along the one-dot chain line in the drawing around the bent portion **44**, so that the lateral edge portion **45** of the sensor **43** (the portion where the first working electrode **47**, the counter electrode **48**, reference electrode **49**, and the second working electrode **50** are present) is disposed substantially in the horizontal direction in a stable state below.
(156) In this substantially horizontal state, the lateral edge portion **45** of the sensor **43** (the first working electrode **47**, the counter electrode **48**, the reference electrode **49**, and the second working electrode **50**) is held in a stable position in each well **80** of the culture vessel, and the culture state in each well **80** can be properly sensed.
(157) Also, since the curve of the arc portion of the bent portion **44** of the sensor **43** is defined by the bottom plate **57** and the top plate **59**, no excessive stress is exerted on the bent portion **44**, so it is possible to prevent disconnection of sensor **43** due to cracking.
(158) The bent portion **44** of the sensor **43** may be bent in a state in which the sensor **43** has been attached to either the top plate **59** or the bottom plate **57**. Also, heat may be applied to the bent portion **44** in the bending of the sensor **43**. In this case, the top plate **59** or the bottom plate **57** is unnecessary.
(159) FIG. **21**A is a top view of the gasket sheet **60**.
(160) As shown in FIG. **21**A, a plurality of port input/output portions **72**, which are disposed close together on the upper surfaces of the plurality of ports **61** (see FIG. **14**), are disposed on the upper surface of the gasket sheet **60**.
(161) FIG. **21**B is a detail view of one of the port input/output portions **72** in FIG. **21**A. The port input/output portion **72** has an additive addition portion A addition port (upper surface opening) **73**, an additive addition portion B addition port (upper surface opening) **74**, and a stirring member air discharge and intake port **75**. Also, the port input/output portion **72** has through-holes for connecting to the connection terminal portion **62** of the sensor **43**. As shown in FIG. **21**B, four through-holes are formed, namely, a through-hole **76** for the first working electrode pad, a through-hole **77** for the counter electrode pad, a through-hole **78** for the reference electrode pad, and a through-hole **79** for the second working electrode pad.
(162) FIG. **22** is a cross-sectional view along the line of the port input/output portion **72** shown in FIG. **21**B.
(163) As described with reference to FIGS. **21**A and **21**B, a plurality of port input/output portions **72** are disposed on the upper surface of the gasket sheet **60**. FIG. **22** shows the state before the board unit **28** is attached from above on the upper surface of the sensor unit **27** on the upper surface of which the gasket sheet **60** is disposed.
(164) Before the board unit **28** is attached to the sensor unit **27**, an additive is preloaded from the additive addition portion A addition port **73**. As shown in FIG. **22**, the additive addition portion A addition port **73** has a recess **73***a* formed on the upper surface, and an addition port **73***b* formed in the center of the recess **73***a*.
(165) Here, if the upper surface of the gasket sheet **60** should be accidentally soiled with an additive, when the board unit **28** is attached to the sensor unit **27**, the lower surface of the piping board portion **29** constituting the lower surface of the hoard unit **28** may end up being soiled by the additive.
(166) With the configuration in this embodiment, since the recess **73***a* is provided, even if an additive should adhere to the bottom surface of the recess **73***a* when the additive is loaded, the lower surface of the board unit **28** will not come into contact with the bottom surface of the recess

73*a* in a state in which the board unit **28** has been attached.
(167) Consequently, preventing an additive from adhering to the lower surface of the board unit **28** makes it possible to prevent the upper surface of the gasket sheet **60** from being soiled by the additive when the additive is loaded.
(168) The configuration for preventing soiling by an additive is similarly applied to the additive addition portion B addition port **74**.
(169) The connecting portion **32** provided so as to extend downward from the board **31** protrudes from the lower surface of the board unit **28**, and electrically connects the second working electrode pad **55** and the reference electrode pad **54** of the connection terminal portion **62** of the sensor **43** via the through-hole **78** for the reference electrode pad and the through-hole **79** for the second working electrode pad.
(170) This electrical connection structure is the same on the side of the through-hole **76** for the first working electrode pad and the through-hole **77** for the counter electrode pad.
(171) FIG. **23** shows a state in which the board unit **28** has been attached from above on the upper surface side of the sensor unit **27**. In this state, a specific pipe of the piping board portion **29** is connected to the additive addition part A addition port **73**. The same applies to the additive addition portion B addition port **74** and the stirring member air discharge and intake port **75**.
(172) Then, the connecting portion **32** extending downward from the board **31** goes through the reference electrode pad through-hole **78** and the second working electrode pad through-hole **79** and is electrically connected to the reference electrode pad **54** and the second working electrode pad **55** of the connection terminal portion **62** of the sensor **43**.
(173) This electrical connection structure is the same for the through-hole **76** for the first working electrode pad and the through-hole **77** for the counter electrode pad.
(174) The gasket sheet **60** of the sensor unit **27** is disposed so as to cover the periphery of the through-hole **76** for the first working electrode pad, the through-hole **77** for the counter electrode pad, the through-hole **78** for the reference electrode pad, and the through-hole **79** for the second working electrode pad, which are through-holes for connecting to the additive addition portion A addition port **73**, the additive addition portion B addition port **74**, the stirring member air discharge and intake port **75**, and the connection terminal portion **62** of the sensor **43**. Consequently, the gasket sheet **60** is used for the purposes of waterproofing and preventing dew condensation.
(175) FIG. **24** is a top view of the well plate **25**.
(176) As shown in FIG. **24**, the well plate **25** has, for example, 24 wells (containers) **80** (4 vertical×6 horizontal). Each well **80** contains a liquid medium (liquid sample) for culturing cells.
(177) The wells **80** are, for example, substantially cylindrical vessels having a diameter of 15.1 mm, into which a sensor **43** having a width of about 7.0 mm is inserted. The amount of medium (liquid sample) that is put into each well **80** is 0.5 to 1.0 mL, for example.
(178) FIG. **25**A is an oblique view of the port **61** for adding an additive to the wells **80**, as viewed from below. FIG. **25**B is an oblique view of the port **61** as viewed from above.
(179) In this embodiment, the port **61** has a stirring member **81**, an additive addition portion A**82**, and an additive addition portion B**83**.
(180) The additive addition portion A**82** and the additive addition portion B**83** are used to add a specific additive to the medium, estimate the subsequent degree of culturing while measuring with the sensor **43**, and determine the optimal cell culture method.
(181) The additive addition portion A**82** and the additive addition portion B**83** have additive containers (additive A container **85**, additive B container **86**) having additive discharge ports (additive A discharge port **85***a*, additive B discharge port **86***a*) as openings for adding the additive into the well **80** (culture vessel), as well as an air pressure supply unit (drive unit **3**, piping tube **6**, piping board portion **29**) for applying air pressure to this additive container.
(182) The additive A container **85** and the additive B container **86** each have a cylindrical shape having an opening (additive A discharge port **85***a*, additive B discharge port **86***a*) that is underneath

in the usage state.
(183) The stirring member **81** is used to stir the medium in the well **80** after the additive has been added, and to stir the additive uniformly into the medium.
(184) As shown in FIG. **25**B, the additive addition portion A addition port **73**, the additive addition portion B addition port **74**, and the stirring member air discharge and intake port **75** are provided on the upper surface of the port **61**.
(185) FIG. **26**A is a top view of the port **61**. FIG. **26**B is a cross-sectional view along the F-F′ line in FIG. **26**A.
(186) As shown in FIG. **26**B, the lower portion of the cylindrical additive A container **85** is formed so that its inside diameter decreases toward the lower end, and the agent A discharge port **85***a* is provided as an opening at the lower end thereof.
(187) A substantially annular dropping adjustment surface **88** (see FIG. **27**) is formed at the outer peripheral edge of the additive A discharge port **85***a*.
(188) FIG. **27** is a detail cross-sectional view of the vicinity of the additive A discharge port **85***a* provided at the lower end of the additive A container **85** of the port **61**.
(189) The lower end of the additive A container **85** is formed so that its outside diameter decreases toward the lower end. Therefore, as shown in FIG. **27**, a substantially conical inclined surface **87** that narrows downward is formed at the lower end of the additive A container **85**.
(190) The substantially annular dropping adjustment surface **88** is provided at the outer peripheral edge of the additive A discharge port **85***a* so as to be disposed substantially horizontally in the usage state.
(191) FIGS. **28**A to **28**C show how the additive is dropped from the lower end portion of the additive A container **85** of the additive addition portion A**82**.
(192) When air pressure is gradually applied from the upper part of the additive A container **85**, the additive is pushed out near the opening of the additive A discharge port **85***a* as shown in FIG. **28**A. As shown in FIG. **28**B, the additive pushed out of the additive A discharge port **85***a* gradually becomes a large water droplet due to surface tension. Then, as shown in FIG. **28**C, the water droplet that has grown in size along the dropping adjustment surface **88** swells out to the outer periphery along the substantially horizontal dropping adjustment surface **88**, and once the weight of the water droplet exceeds its surface tension, the drop falls from the A discharge port **85***a*.
(193) As described above, because the dropping adjustment surface **88** is provided to the additive A discharge port **85***a*, the additive falls into the medium as a water droplet of the desired size. Therefore, the concentration of the additive contained in the medium can be gradually increased, which means that the additive can be added to the cells being cultured without leading to any sudden change in the concentration in which the additive is contained in the medium.
(194) That is, in this embodiment, when air pressure is applied into the additive A container **85**, the additive held in the additive A container **85** moves over to the additive A discharge port **85***a* side. Then, at the dropping adjustment surface **88** provided on the outer peripheral edge of the additive A discharge port **85***a*, the additive is held by the surface tension to form a large droplet, which then falls into the culture vessel (well **80**) below once the weight of the additive becomes greater than the holding force produced by surface tension.
(195) Also, when this drop falls, another drop of additive begins to form in the additive A discharge port **85***a*, and then falls as a droplet into the lower culture vessel (well **80**).
(196) That is, in this embodiment, since the additive is supplied intermittently into the culture vessel (well **80**), the additive is less likely to subject the cells to sudden stress, which allows cell culture analysis to be performed properly.
(197) FIG. **29** is a plan view of the additive A**82** (additive A container **85**) on the additive A discharge port **85***a* side.
(198) The above-mentioned dropping adjustment surface **88** is provided at the outer peripheral edge of the opening portion of the additive A discharge port **85***a*. The inclined surface **87** is

provided at the outer periphery of the dropping adjusting surface **88**.
(199) The dropping adjustment surface **88** is subjected to a hydrophilic treatment in an annular shape, while the inclined surface **87** is rendered hydrophobic.
(200) Consequently, the dropping adjustment surface **88** holds the droplet using the surface tension of the additive, but when the droplet of the additive grows all the way to the inclined surface **87**, the force holding the droplet by surface tension of the additive decreases, so the additive drops into the lower culture vessel (well **80**).
(201) Also, as shown in FIG. **29**, the inner peripheral side (first surface) **88***a* of the annular dropping adjustment surface **88** may be subjected to a hydrophilic treatment, and the outer peripheral side (second surface) **88***b* may be subjected to a hydrophobic treatment.
(202) In this case, the inner peripheral side **88***a* of the dropping adjustment surface has a force for holding a droplet, and the outer peripheral side **88***h* of the dropping adjusting surface does not have a force for holding a droplet. Therefore, once the droplet grows all the way to the outer peripheral side **88***b*, the force for holding the droplet suddenly decreases, and a drop of the additive falls into the lower culture vessel (well **80**).
(203) Next, the operation of the additive addition portion will be described with reference to FIGS. **30**A to **31**B.
(204) FIG. **30**A is a diagram of the first step, when the additive **90** is put into the additive A container **85**.
(205) As shown in FIG. **30**A, the additive **90** is preloaded into the additive A container **85** from the additive addition portion A addition port **73** using a pipette tip **89**.
(206) FIG. **30**B shows the state after the loading of the additive **90**.
(207) Since the amount of the additive **90** loaded into the additive A container **85** is less than the volume of the additive A container **85**, the additive **90** does not overflow from the additive A container **85**.
(208) FIG. **30**C is a detail view of the additive A discharge port **85***a* when the additive is loaded.
(209) During the loading of the additive, the surface tension of the additive at the opening of the additive A discharge port **85***a* is greater than the gravity exerted on the additive **90**, as shown in FIG. **30**C, so the additive **90** is held within the agent A container **85**.
(210) FIG. **31**A shows a state in which the piping board portion **29** has been connected from above after the loading of the additive.
(211) In this state, the additive addition portion A piping line **91** included in the piping board portion **29** is linked to the additive addition portion A addition port **73** of the additive A container **85**. Air pressure is then applied from the additive addition portion A piping line **91** in the piping board portion **29** linked to the additive addition portion A addition port **73** of the additive A container **85**. Consequently, as shown in FIG. **31**B, the additive **90** is added to the well **80**.
(212) Even after the additive **90** has been added, a small amount of the additive **90** remains in the additive A container **85**. This prevents air or bubbles from being discharged from the additive A discharge port **85***a*.
(213) Next, the configuration and operation of the stirring member **81** will be described with reference to FIGS. **32**A to **32**C.
(214) FIG. **32**A is a top view of the port **61** including the stirring member **81**. FIG. **32**B is a cross-sectional view along the G-G′ line in FIG. **32**A. FIG. **32**C is a detail oblique view of a liquid discharge and intake port **93** provided at the lower end of the stirring vessel **92** of the stirring member **81**.
(215) As shown in FIGS. **32**A to **32**C, the stirring member **81** has the liquid discharge and intake port **93** that is provided under the stirring vessel **92** and is immersed in the medium, and an air discharge and intake port **94** that is formed on the upper surface of the stirring vessel **92**.
(216) FIG. **33**A shows the initial state of the stirring member **81**. FIG. **33**B shows a state in which an air discharge and intake unit **95** is linked to the stirring member **81**. FIG. **33**C shows a state in

which the air discharge and intake unit **95** acts in the direction in which air is discharged.
(217) As shown in FIG. **33**B, the air discharge and intake unit **95** is linked to the air discharge and intake port **94**. The air discharge and intake unit **95** is constituted by the drive unit **3**, the piping tube **6**, and the piping board portion **29**.
(218) As shown in FIG. **33**A, in the state before the stirring member **81** is linked to the air discharge and intake unit **95**, the liquid discharge and intake port **93** is immersed in the medium in the well **80**. In this state, the medium flows into the stirring vessel **92** from the liquid discharge and intake port **93**, and the medium flows into the stirring vessel **92** up to almost the same height as the liquid level L**1** of the medium in the well **80**.
(219) When the stirring member **81** is linked to the air discharge and intake unit **95**, the air discharge and intake unit **95** first acts in the direction in which air is drawn in, as shown in FIG. **33**B. Consequently, the inside of the stirring vessel **92** is under negative pressure, so the medium in the well **80** is drawn up through the liquid discharge and intake port **93**, and the liquid level L**2** of the medium in the stirring vessel **92** becomes higher than the liquid level L**1** of the medium in the well **80**.
(220) After this, the air discharge and intake unit **95** acts in the direction in which air is discharged, as shown in FIG. **33**C. Consequently, the inside of the stirring vessel **92** is under positive pressure, so the medium is discharged from the liquid discharge and intake port **93** into the well **80**.
(221) At this point, as shown in FIG. **33**B, the discharged amount is the same as the amount of medium drawn in into the stirring vessel **92**, so in the initial state shown in FIG. **33**A, all of the medium that has flowed into the stirring vessel **92** still remains in the stirring vessel **92**. Consequently, no air or bubbles are discharged from the liquid discharge and intake port **93** into the well **80**.
(222) As discussed above, the cell culture analyzer **1** of this embodiment comprises the stirring member **81** that has the liquid discharge and intake port **93** immersed in the medium, and the air discharge and intake port **94** connected to the air discharge and intake unit **95**.
(223) Consequently, there is no need for a stirring rod or plunger to be provided for each culture vessel, which allows the configuration of the device to be simplified.
(224) That is, with the cell culture analyzer **1** in this embodiment, in the air discharge and intake unit **95**, air is discharged from and drawn into to the stirring member **81**, so that after the medium in the culture vessel (well **80**) is drawn into the stirring member **81**, the medium in the culture vessel (well **80**) is stirred by being discharged.
(225) This eliminates the need for a stirring rod or a plunger to be provided for each culture vessel, and allows the device configuration to be simplified.
(226) Also, as shown in FIG. **32**B, the liquid discharge and intake port **93** of the stirring member **81** is provided on the lower side surface of the stirring vessel **92**.
(227) Consequently, the medium discharged from the lower side surface of the stirring vessel **92** is extruded in the horizontal direction and stirred along the inner peripheral surface of the culture vessel (well **80**), as shown in FIG. **34**. As a result, convection can be generated in the medium in the culture vessel (well **80**), and stirring can be performed up to the gap between the sensor **43** and the inner peripheral surface of the well **80**, so the medium can be stirred more effectively.
(228) Furthermore, as shown in FIG. **34**, the liquid discharge and intake port **93** of the stirring member **81** is located at a position that is away from the center O in the culture vessel (well **80**), that is, a position that is close to the inner peripheral surface of the culture vessel (well **80**), and the opening thereof is disposed in an orientation that is opposite the inner peripheral surface of the culture vessel (well **80**).
(229) Consequently, the medium discharged from the liquid discharge and intake port **93** collides with the inner peripheral surface of the culture vessel (well **80**) and circulates within the culture vessel (well **80**) along the inner peripheral surface, and this stirs the entire medium in the container (well **80**). As a result, the medium can be properly stirred.

(230) Also, the distance from the liquid discharge and intake port **93** of the stirring member **81** to the additive A discharge port **85***a* is equal to the distance to the additive B discharge port **85***b*.
(231) Thus disposing the components such that the distances from the openings (additive A discharge port **85***a*, additive B discharge port **86***a*) of the two additive addition parts (additive addition part A**82**, additive addition part B**83**) to the liquid discharge and intake port **93** of the stirring member **81** is are equal to each other means that the additive A discharge port **85***a* and the additive B discharge port **86***a* are disposed in left and right symmetry with respect to the inner peripheral surface of the culture vessel (well **80**).
(232) Consequently, the additives from the respective additive addition portions (additive addition portion A**82**, additive addition portion B**83**) are uniformly stirred in the culture vessel by the stirring member **81**.
(233) As shown in FIGS. **35**A and **35**B, the shape of the opening of the liquid discharge and intake port **93** may be set by taking into account ease of molding when using a material such as resin.
(234) More specifically, as shown in FIGS. **35**A and **35**B, the liquid discharge and intake port **93** of the stirring member **81** is provided with an opening portion on the lower side surface of the stirring vessel **92** so that no undercut will be necessary. That is, the liquid discharge and intake port **93** shown in FIG. **35**B is formed at the corner portion of the lower end portion of the stirring member **81**.
(235) This eliminates the need for an undercut during molding, which simplifies the manufacturing process and allows the port **61** to be manufactured at a lower cost.
(236) FIG. **36**A is a flowchart of an analysis method including a measurement step and an addition step including stirring and homogenization. FIG. **36**B is a flowchart of the addition steps A and B included in FIG. **36**A. FIG. **36**C is a flowchart of the flow in the measurement step included in FIG. **36**A.
(237) The cell culture analysis method used in the cell culture analyzer **1** of this embodiment comprises two kinds of stirring steps (stirring and homogenization).
(238) First, as shown in FIG. **36**A, in the measurement step S**11**, the components of the medium are measured in a state in which the sensor **43** is immersed in the culture vessel (well **80**), after which stirring is performed by the stirring member **81**.
(239) The stirring step included in this measurement step S**11** shall be referred to as the second stirring step.
(240) Next, the addition step A of S**12** is carried out.
(241) In the addition step A of S**12**, the additive is added to the well **80**, which is a culture vessel, by the additive addition portion A**82** or the additive addition portion B**83**, after which the medium is stirred by the stirring member **81**.
(242) The stirring step included in the addition step A shall be referred to as the first stirring step.
(243) After this are carried out the measurement step S**13**, in which the procedure is the same as in the measurement step S**11**; the addition step B S**14**, in which the procedure is the same as in the addition step A of S**12**; and the measurement step S**15**, in which the procedure is the same as in S**11** and S**13**. The process is then ended.
(244) In the addition steps A and B carried out in S**12** and S**14**, as shown in FIG. **36**B, the additive is first dropped in S**21**, intake (stirring) is performed in S**22**, and discharge (stirring) is performed in S**23**. The intake and discharge of S**22** and S**23** are repeated N number of times.
(245) In the measurement steps carried out in S**11**, S**13**, and S**15**, as shown in FIG. **36**C, first, measurement is performed in S**31**, intake (uniform) is performed in S**32**, and discharge (uniform) is performed in S**33**. The intake and discharge in S**32** and S**33** are repeated N number of times, depending on the type of medium and additive, the amount in which the additive is added, and so forth (N=1, 2, 3, . . .). At this point, the amounts of medium that are drawn in and discharged in the intake and discharge are substantially the same.
(246) Thus performing first and second stirring steps during measurement and each time an

additive is added allows the medium to be stirred in such a way that the additive concentration is not higher in one location than another, so measurement accuracy is improved.
(247) Also, the absolute value of the air pressure generated by the air discharge and intake unit **95** during stirring in the first stirring step is greater than the absolute value of the air pressure generated by the air discharge and intake unit **95** during stirring in the second stirring step.
(248) Consequently, when the additive is added, a more powerful stirring operation can be performed, and during measurement, a gentler stirring operation can be performed as compared with that at the time of addition.
(249) Thus stirring forcefully when the additive is added and stirring more gently during measurement as compared with that at the time of adding the additive allows the stirring to be performed more reliably, and improves the measurement accuracy.
(250) In the cell culture analyzer **1** of this embodiment, as discussed above, after the additive has been added to the medium using the additive addition portion A**82** or the additive addition portion B**83**, the stirring member **81** performs the first stirring operation. Then, when the sensor **43** measures the components of the medium, the stirring member **81** performs the second stirring operation.
(251) At this point, the absolute values of the air pressures associated with discharge and intake from and to the air discharge and intake unit **95** during the first stirring operation are greater than the absolute values of the air pressures associated with discharge and intake from and to the air discharge and intake unit **95** during the second stirring operation.
(252) Consequently, the stirring can be stronger during the addition of the additive and gentler during measurement as compared with that at the time of adding the additive, which means that the proper stirring can be performed for each step, and the accuracy of measurement using the sensor **43** will be improved.
(253) Also, when the sensor **43** measures the components of the medium in the culture vessel (well **80**), the second stirring operation is halted.
(254) Consequently, the concentration distribution of the medium will be more uniform, and measurement accuracy is further improved.
(255) FIG. **37** is an exploded oblique view of a seal **96** that is affixed to the upper surface of the sensor unit **27**.
(256) As shown in FIG. **37**, a plurality of port input/output portions **72** connected to the upper surface of the port **61** are disposed in a plurality of openings formed in the gasket sheet **60** disposed on the upper surface side of the sensor unit **27**.
(257) As described above, each port input/output portion **72** has an additive addition part A addition port (upper surface opening) **73**, an additive addition part B addition port (upper surface opening) **74**, a stirring member air discharge and intake port **75**, and four through-holes for connecting to the connection terminal portion **62** of the sensor **43** (through-hole **76** for the first working electrode pad, through-hole **77** for the counter electrode pad, through-hole **78** for the reference electrode pad, and through-hole **79** for the second working electrode pad).
(258) Thus, a plurality of openings are disposed in the plurality of port input/output portions **72** disposed in the plurality of openings formed in the gasket sheet **60**.
(259) In this embodiment, the seal **96** is provided, which can be affixed to a plurality of openings in a removable state. More specifically, the seal **96** is used to seal off the opening of the port input/output portion **72** located above an unused well **80**.
(260) Since an opening in the board (gasket sheet **60**) corresponding to an unused culture vessel (well **80**) is covered with the seal **96**, this prevents air from leaking out through the additive container (additive A container **85**, additive B container **86**) disposed at the position corresponding to that opening. Meanwhile, an appropriate air pressure is applied to an additive container (additive A container **85**, additive B container **86**) disposed at a position corresponding to the opening corresponding to a culture vessel (well **80**) that is being used, and the additive can be supplied

appropriately.
(261) Also, cell culture analysis can be performed by simply removing a part of the seal **96** affixed to the position corresponding to a culture vessel that is being used (additive A container **85**, additive B container **86**) from the gasket sheet (base) **60**, and this makes the job easier.
(262) That is, as shown in FIG. **37**, the seal **96** is configured by affixing a bottom seal **96***a* and a top seal **96***b* one over the other.
(263) More specifically, as shown in FIG. **38**A, the seal **96** (bottom seal **96***a* and top seal **96***h*) is affixed by means of the adhesive force of a plurality of adhesive portions **97** disposed on the upper surface of the gasket sheet **60** on the upper surface of the sensor unit **27**, in the left-right direction in the drawing.
(264) As shown in FIG. **38**B, the bottom seal **96***a* has a single overall peeling tab **96***aa*, plurality of individual peeling tabs **96***ab*, cut portions **96***ac*, and perforated portions **96***ad*.
(265) The overall peeling tab **96***aa* is a portion that is grasped in the user's fingers when the entire bottom seal **96***a* is to be peeled off, and is provided along the lengthwise direction from the lower end portion of the short side of the substantially rectangular bottom seal **96***a*.
(266) The individual peeling tabs **96***ab* are portions that are grasped in the user's fingers when a bottom seal **96***a* is to be partially peeled off, and are provided in a direction intersecting the long side of the substantially rectangular bottom seal **96***a* in the lengthwise direction.
(267) The cut portions **96***ac* are cuts formed between adjacent individual peeling tabs **96***ab*, and are formed substantially parallel to the short side of the bottom seal **96***a*. Also, the cut portions **96***ac* are formed from the end of the bottom seal **96***a* to a position about two-thirds of the way along the short side. The cut portions **96***ac* are formed by making cuts from the long side of the bottom seal **96***a* on the side where the individual peeling tabs **96***ab* are provided.
(268) The perforated portions **96***ad* are formed at positions contiguous with the cut portions **96***ac*, and are formed from the end of the bottom seal **96***a* to a position about one-third of the way along the short side. The perforated portions **96***ad* are formed from the end on the side where the overall peeling tab **96***aa* is formed.
(269) Consequently, when the user grasps the overall peeling tab **96***aa* and peels off the bottom seal **96***a*, the entire bottom seal **96***a* can be peeled off at once because it is connected at the perforated portions **96***ad*.
(270) Meanwhile, when user grasps an individual peeling tab **96***ab* and peels off a part of the bottom seal **96***a*, a part of the bottom seal **96***a* can be easily peeled off at the desired position by simply peeling off from the cut side of the cut portions **96***ac* and thereby tearing along the perforated portion **96***ad*.
(271) As shown in FIG. **38**C, the top seal **96***h* has an overall peeling tab **96***ba*, individual peeling tabs **96***bb*, cut portions **96***bc*, and perforated portions **96***bd*.
(272) The overall peeling tab **96***ba* is a portion that is grasped in the user's fingers when the entire top seal **96***b* is to be peeled off, and is provided along the lengthwise direction from the lower end portion of the short side of the substantially rectangular top seal **96***h*.
(273) The individual peeling tabs **96***bb* are portions that are grasped in the user's fingers when the top seal **96***b* is to be partially peeled off, and are provided in a direction intersecting the long side of the substantially rectangular bottom seal **96***a* in the lengthwise direction.
(274) The cut portions **96***bc* are cuts formed between adjacent individual peeling tabs **96***bb*, and are formed substantially parallel to the short side of the top seal **96***b*. Also, the cut portions **96***bc* are formed from the end of the top seal **96***b* to a position about two-thirds of the way along the short side. The cut portions **96***bc* are formed by making cuts from the long side of the bottom seal **96***a* on the side where the individual peeling tabs **96***bb* are provided.
(275) The perforated portions **96***bd* are formed at positions contiguous with the cut portions **96***bc*, and are formed from the end of the top seal **96***b* to a position about one-third of the way along the short side. The perforated portions **96***bd* are formed from the end on the side where the overall

peeling tab **96***ba* is formed.
(276) Consequently, in a state in which the bottom seal **96***a* and the top seal **96***b* shown in FIG. **38**D have been affixed, when the user grasps the overall peeling tab **96***ba* and peels off the top seal **96***b*, the entire top seal **96***b* can be peeled off at once because it is connected at the perforated portions **96***bd*.
(277) Meanwhile, when user grasps an individual peeling tab **96***ab* and peels off a part of the top seal **96***b*, a part of the top seal **96***b* can be easily peeled off at the desired position by simply peeling off from the cut side of the cut portions **96***bc* and thereby tearing along the perforated portion **96***bd*.
(278) In a state in which the bottom seal **96***a* and the top seal **96***b* have been attached to the upper surface of the sensor unit **27**, as shown in FIG. **38**D, the overall peeling tabs **96***aa* and **96***ba* of the bottom seal **96***a* and the top seal **96***b* are disposed in positions that do not overlap with each other.
(279) Consequently, when the overall peeling tab **96***ba* is grasped to peel off only the top seal **96***b*, this prevents the user from accidentally grasping all the way to the overall peeling tab **96***aa* of the bottom seal **96***a*.
(280) Here, as discussed above, the sensor unit **27** in this embodiment is provided to the user in a state in which the bottom seal **96***a* and the top seal **96***b* have been attached to the upper surface. The user then peels off all or part of the bottom seal **96***a* and/or the top seal **96***b* of the row to be used, corresponding to the position of the well **80** to be used, and this prevents unnecessary additive from being added to a well **80** that is not to be used.
(281) Also, the bottom seal **96***a* and the top seal **96***b* are affixed to the adhesive portions **97** on the upper surface of the gasket sheet **60**.
(282) Consequently, the seal **96** is affixed to the upper surface of the gasket sheet **60** via the adhesive portions **97** provided at positions that are offset from the additive addition portions A**82** and B**83**, so the seal **96** does not need to be directly affixed to the upper surface of the additive addition portions A**82** and B**83**. This means that it is not necessary to provide an adhesive agent on the upper surfaces of the additive addition portions A**82** and B**83**, so the adhesive is prevented from becoming admixed.
(283) Here, in this embodiment, as shown in FIG. **39**A, the sensor unit **27** is provided to the user in a state in which the bottom seal **96***a* and the top seal **96***b* have been affixed to the upper surface of the gasket sheet **60**.
(284) At this point, in the state provided to the user, the various opening portions are as follows. Electrode pad portion **98**: OPEN, additive addition portion A**82**: CLOSED, additive addition portion B**83**: CLOSED, stirring member **81**: CLOSED.
(285) When in the additive addition portion A**82** is filled with an additive, as shown in FIG. **39**B, the overall peeling tab **96***ba* is grasped and just the top seal **96***b* is peeled off.
(286) At this point, the various opening portions are as follows after the top seal **96***b* has been peeled off. Electrode pad portion **98**: OPEN, additive addition portion A**82**: OPEN, additive addition portion B**83**: CLOSED, stirring member **81**: OPEN.
(287) Furthermore, when additive addition portion A**82** is filled with an additive, as shown in FIG. **39**B, the overall peeling tab **96***aa* is grasped and the bottom seal **96***a* is also peeled off.
(288) At this point, the various opening portions are as follows after the bottom seal **96***a* has been peeled off. Electrode pad portion **98**: OPEN, additive addition portion A**82**: OPEN, additive addition portion B**83**: OPEN, stirring member **81**: OPEN.
(289) Consequently, the bottom seal **96***a* and the top seal **96***b* can be peeled off in just a few steps.
(290) In this embodiment, all or part of these two seals **96***a* and **96***b* is selectively peeled off according to the position of the culture vessel to be used, which allows the openings in the board portion corresponding to the upper surface of the culture vessels (wells **80**) that will not be used to be sealed off by the seal **96**. This makes the product more convenient to use for the user.
(291) For example, in the usage scenarios 1 to 5 shown in FIG. **40**, all or part of the seal **96** is peeled off depending on the openings of the port **61** to be used.

(292) In usage scenario 1, as shown in FIG. **40**, when neither port A nor port B is to be used, that is, when the medium is to be measured without adding any additive, the product is used just as it is provided to the user, without the seal **96** being peeled off.
(293) In usage scenario 2, as shown in FIG. **40**, when only the first and second rows of the port A are used, and the port B is not used, the individual peeling tab **96***bb* of the second row of the top seal **96***b* is peeled off, and the product is used in a state in which the openings in the first and second rows of the port A are OPEN.
(294) In usage scenario 3, as shown in FIG. **40**, when all the rows of the port A are used and the port B is not used, the overall peeling tab **96***ba* for the top seal **96***b* is peeled off, and the product is used in a state in which the openings in all the rows of the port A are OPEN.
(295) In usage scenario 4, as shown in FIG. **40**, when only the first and second rows of the port A and only the first and second rows of the port B are used, the individual peeling tab **96***bb* of the second row of the top seal **96***b* is peeled off, the individual peeling tab **96***ab* in the second row of the bottom seal **96***a* is peeled off, and the product is used in a state in which the openings in the first and second rows of the ports A and B are OPEN.
(296) In usage scenario 5, as shown in FIG. **40**, when all the rows of ports A and B are used, the overall peeling tab **96***aa* of the bottom seal **96***a* is peeled off, so that top seal **96***b* affixed to the upper surface of the bottom seal **96***a* is also peeling off, and the product is used in a state in which the openings in all the rows of the ports A and B are OPEN.
(297) This configuration in which the seal **96** is used is similarly applied to the stirring member air discharge and intake port **75**.
(298) That is, the stirring member air discharge and intake port **75** is sealed off above any unused wells **80** by using the seal **96**, which allows the appropriate air pressure to be applied just to the stirring member **81** being used.
(299) In this case, cell culture analysis can be performed by removing the seal **96** corresponding to the upper surface of the stirring member **81** to be used, which makes the work easier.

Embodiment 2

(300) A cell culture analyzer according to another embodiment of the present invention will now be described with reference to the appended drawings.
(301) The cell culture analyzer in this embodiment differs from Embodiment 1 above in that a port (additive supply member) **161** including the stirring member **181** shown in FIGS. **41**A and **41**B is used instead of the stirring member **81** shown in FIGS. **32**A to **32**C, etc.
(302) As to the rest of the configuration, members having the same configuration and function will be numbered the same and will not be described again.
(303) FIG. **41**A is a top view of the port **161** including the stirring member **181**. FIG. **41**B is a cross-sectional view along the line in FIG. **41**A.
(304) As shown in FIGS. **41**A and **41**B, the stirring member **181** has a liquid discharge and intake port **193** that is provided below the stirring vessel **192** and is immersed in the medium, and an air discharge and intake port **194** that is formed on the upper surface of the stirring vessel **192**.
(305) In the stirring member **181**, when the stirring step is commenced, the liquid discharge and intake port **193** is immersed in the medium in the well **80** as shown in FIG. **41**B. In this state, the medium flows from the liquid discharge and intake port **193** into the stirring vessel **192**, and flows into the stirring vessel **192** to roughly the same height as the liquid level L**1** of the medium in the well **80**.
(306) Then, when the stirring member **181** is connected to the above-mentioned air discharge and intake unit **95** (see FIG. **33**B), the air discharge and intake unit **95** acts in the direction of drawing in air, which puts the inside of the stirring vessel **192** under negative pressure, and draws the medium in the well **80** up through the liquid discharge and intake port **193**, so that the liquid level of the medium in the stirring vessel **192** becomes higher than the liquid level L**1** of the medium in the well **80**.

(307) After this, the air discharge and intake unit **95** acts in the direction of discharging air to put the inside of the stirring vessel **192** under positive pressure and discharge the medium from the liquid discharge and intake port **193** into the well **80**.
(308) Furthermore, in this embodiment, the liquid discharge and intake port **193** provided on the lower side surface of the stirring vessel **192** shown in FIG. **42**A discharges or draws in the medium toward the side wall of the well **80**.
(309) Also, as shown in FIG. **42**B, the stirring vessel **192** has on its inner surface an inclined surface **192***a* that is provided to the portion opposite the liquid discharge and intake port **193** and is formed so as to angle downward toward the liquid discharge and intake port **193**, and a horizontal surface **192***b* that is provided between the inclined surface **192***a* and the liquid discharge and intake port **193**.
(310) The inclined surface **192***a* is a part of the inner wall surface near the bottom surface of the stirring vessel **192**, is disposed at a position opposite the liquid discharge and intake port **193**, and is formed so as to angle downward toward the liquid discharge and intake port **193**.
(311) Consequently, providing the inclined surface **192***a* to the portion facing the liquid discharge and intake port **193** makes it possible to suppress the generation of air bubbles in the corner portion opposite the liquid discharge and intake port **193**. This prevents bubbles from being discharged from the liquid discharge and intake port **193**, which suppresses the generation of spray that could be a source of contamination, so bubbles will not directly collide with the cells adhering to on the bottom surface of the well **80**, and it is less likely that there will be an adverse effect on the cells.
(312) The horizontal surface **192***b* is a bottom surface formed substantially horizontally in an orientation in which the stirring vessel **192** is immersed in the medium, and is formed as a surface linking the lower end portion of the inclined surface **192***a* and the liquid discharge and intake port **193**.
(313) Consequently, since the inclined surface **192***a* is provided in a state in which the horizontal surface **192***b* remains on the bottom surface of the stirring vessel **192**, it is possible to generate a flow of medium toward the inner side wall of the well **80**.
(314) Furthermore, as shown in FIG. **42**A, since the shape of the lower end portion of the stirring vessel **192** eliminates the need for undercut during molding, the manufacturing process is simpler, and the port **161** can be manufactured at a lower cost.
Embodiment 3
(315) The cell culture analyzer according to yet another embodiment of the present invention will now be described with reference to the appended drawings.
(316) As shown in FIG. **43**A, the cell culture analyzer in this embodiment differs from Embodiment 1 above in that the port (additive supply member) **261** including the stirring member **281** shown in FIGS. **43**A and **43**B is used instead of the stirring member **81** shown in FIGS. **32**A to **32**C, etc.
(317) As to the rest of the configuration, members having the same configuration and function will be numbered the same and will not be described again.
(318) FIG. **43**A is a top view of the port **261** including the stirring member **281**. FIG. **43**B is a plan view of the flow of the medium discharged from the stirring member **281**.
(319) In this embodiment, as shown in FIGS. **43**A and **43**B, the stirring member **281** comprises three openings (liquid discharge and intake ports **293***a*, **293***b*, **293***c*) for discharging the medium in different directions, as liquid discharge and intake ports.
(320) As shown in FIG. **43**B, the discharge and intake port **293***a* is formed so as to open toward the inner wall surface of the well **80**, and discharges the drawn-in medium to the inner wall surface of the well **80**.
(321) Consequently, the medium discharged from the liquid discharge and intake port **293***a* collides with the inner wall surface of the well **80** and circulates around the well **80** along the inner peripheral surface, which stirs the entire medium in the well **80**, so the medium can be sufficiently

stirred.
(322) As shown in FIG. **43**B, the discharge and intake port **293***b* is formed so as to open toward the additive A discharge port **85***a*, and discharges the drawn-in medium to the position where the additive A is discharged.
(323) As shown in FIG. **43**B, the discharge and intake port **293***c* is formed so as to open toward the additive B discharge port **86***a*, and discharges the drawn-in medium to the position where the additive B is discharged.
(324) Consequently, once the medium has been drawn in, it can be split up and discharged from the plurality of liquid discharge and intake ports **293***a* to **293***c*, which allows the flow rate of the discharged medium to be kept lower. Also, since the liquid discharge and intake ports **293***h* and **293***c* can send the medium toward the positions where the additive A and the additive B are discharged, respectively, the stirring effect of the additives A and B can be improved.
(325) As a result, the stirring step can be performed more efficiently, without generating a strong flow rate in the discharged medium, which allows gentle stirring to be performed, without subjecting the cells in the well **80** to a load.
(326) Furthermore, just as in Embodiments 1 and 2 above, the shape of the lower end portion of the stirring vessel **292** eliminates the need for undercut during molding, so the manufacturing process is simpler, and the port **261** can be manufactured at a lower cost.
Embodiment 4
(327) The cell culture analyzer according to yet another embodiment of the present invention will now be described with reference to the appended drawings.
(328) As shown in FIG. **44**A, the cell culture analyzer in this embodiment differs from Embodiment 1 above in that the port (additive supply member) **361** including the stirring member **381** shown in FIGS. **44**A and **44**B is used instead of the stirring member **81** shown in FIGS. **32**A to **32**C, etc.
(329) As to the rest of the configuration, members having the same configuration and function will be numbered the same and will not be described again.
(330) FIG. **44**A is a lateral cross-sectional view showing a state in which the port **361** including the stirring member **381** is immersed in the medium contained in the well **80**. FIG. **44**B is a lateral cross-sectional view showing a state in which the port **361** has been pulled up from the state shown in FIG. **44**A.
(331) In this embodiment, as shown in FIGS. **44**A and **44**B, a small-diameter portion **394** having an inside diameter smaller than that of the upper portion is formed in the lower part of the stirring vessel **392** of the stirring member **381** of the port **361**.
(332) As shown in FIG. **44**A, etc., because the small-diameter portion **394** has a smaller inside diameter than the portion above the small-diameter portion **394** of the stirring vessel **392**, the volume is smaller for a given length. The small-diameter portion **394** is provided from the position of the liquid discharge and intake port **393** to a position higher than the liquid level L**1** of the medium, in an orientation of being immersed in the medium in the well **80**.
(333) Consequently, as shown in FIG. **44**B, the residual medium **394***a* remaining inside the stirring member **381** after being pulled up from the medium can be reduced as much as possible. Therefore, only a tiny amount of medium will be wasted, and it is possible to avoid adversely affecting the subsequent cell culture step.
Embodiment 5
(334) The cell culture analyzer according to yet another embodiment of the present invention will now be described with reference to the appended drawings.
(335) As shown in FIG. **45**A, etc., the cell culture analyzer of this embodiment differs from Embodiment 4 above in that a substantially annular rib **395** is additionally provided to the upper end portion of the small-diameter portion of the stirring member **381** described in Embodiment 4 above.

(336) As to the rest of the configuration, members having the same configuration and function will be numbered the same and will not be described again.
(337) As shown in FIGS. **45**A and **45**B, the rib **395** is provided to the upper end portion of the small-diameter portion **394**, that is, to a portion of an inclined surface that links the small-diameter portion **394** in the stirring vessel **392** with the other large-diameter portion. As shown in FIG. **46**, the rib **395** is formed so as to protrude inward in the radial direction from the inner peripheral surface of the stirring vessel **392**.
(338) Consequently, for example, any medium that rises up in the small-diameter portion **394** due to capillary action or the like will be held back by the rib **395**, and as shown in FIG. **45**B, the amount of the residual medium **394***a* after the stirring step is completed can be kept at or below a certain level.
Embodiment 6
(339) The cell culture analyzer according to yet another embodiment of the present invention will now be described with reference to the appended drawings.
(340) The cell culture analyzer of this embodiment is constituted by a combination of the small-diameter portion **394** described in the above Embodiments 4 and 5, the rib **395** described in the above Embodiment 5, and the liquid discharge and intake port **193** disposed opposite the inclined surface **192***a* described in the above Embodiment 2.
(341) As to the rest of the configuration, members having the same configuration and function will be numbered the same and will not be described again.
(342) The liquid discharge and intake port **193** is formed at the distal end of the small-diameter portion **394**, and as described in Embodiment 2 above, draws in or discharges the medium toward the side wall of the well **80**.
(343) Also, the stirring vessel **392** has an inclined surface **192***a* that is provided on the inner surface thereof at the portion opposite the liquid discharge and intake port **193** and is formed so as to angle downward toward the liquid discharge and intake port **193**, and a horizontal surface **192***b* that is provided between the inclined surface **192***a* and the liquid discharge and intake port **193**.
(344) The inclined surface **192***a* is a part of the inner wall surface near the bottom surface of the stirring vessel **392**, is disposed at a position opposite the liquid discharge and intake port **193**, and is formed so as to angle downward toward the liquid discharge and intake port **193**.
(345) Consequently, providing the inclined surface **192***a* to the portion opposite the liquid discharge and intake port **193** makes it less likely that air bubbles will be generated in the corner portion opposite the liquid discharge and intake port **193**. This prevents bubbles from being discharged from the liquid discharge and intake port **193**, which suppresses the generation of spray that could be a source of contamination, so bubbles will not directly collide with the cells adhering to on the bottom surface of the well **80**, and it is less likely that there will be an adverse effect on the cells.
(346) Also, as shown in FIG. **47**A, a small-diameter portion **394** having an inside diameter smaller than that of the upper portion is formed in the lower portion of the stirring vessel **392** of the stirring member **381**.
(347) As shown in FIG. **47**A, the small-diameter portion **394** has a smaller inside diameter than the portion above the small-diameter portion **394** of the stirring vessel **392**, so the volume is smaller for a given length. The small-diameter portion **394** is provided from the position of the liquid discharge and intake port **393** to a position higher than the liquid level L**1** of the medium, in an orientation of being immersed in the medium in the well **80**.
(348) Consequently, the amount of residual medium **394***a* left inside the stirring member **381** after being pulled up from the medium can be kept as small as possible. Therefore, only a tiny amount of medium will be wasted, and it is possible to avoid adversely affecting the subsequent cell culture step.
(349) Furthermore, as shown in FIG. **47**A, the rib **395** is provided to the upper end portion of the

small-diameter portion **394**, that is, to a portion of an inclined surface that links the small-diameter portion **394** in the stirring vessel **392** with the other large-diameter portion. The rib **395** is formed so as to protrude inward in the radial direction from the inner peripheral surface of the stirring vessel **392**.

(350) Consequently, for example, any medium that rises up in the small-diameter portion **394** due to capillary action or the like will be held back by the rib **395**, and the amount of the residual medium **394***a* after the stirring step is completed can be kept at or below a certain level.

INDUSTRIAL APPLICABILITY

(351) The cell culture analyzer of the present invention eliminates the need for a stirring rod or plunger to be readied for each culture vessel, and has the effect of affording a more compact size, and as such can be broadly applied to various devices used for cell culture analysis.

REFERENCE SIGNS LIST

(352) **1** cell culture analyzer **2** analysis unit **3** drive unit **3***a* housing **4** control unit **5** electrical cable **6** piping tube **7** culture incubator **8** door **9** syringe **10** plunger **11** multi-directional switching valve **12** motor **13** motor N, **15**, **16**, **17** valves **18** rotating portion **19** rotating flow path **20** adapter unit **21** top unit **22** bottom unit **23** front opening **24** adapter bottom **25** well plate **26** adapter top **27** sensor unit **28** board unit **29** piping board **30** board base **30***a* contact through-hole **31** board **32** connecting portion **33**, **34**, **35**, **36** piping tubes **37** air inlet (intake port) **38** through-hole **39** piping tube connecting portion **40** leg portion (support) **41** through-hole **42** positioning hole **43** sensor **43***a* main body portion **44** bent portion **45** lateral edge portion **46** vertical edge portion **47** first working electrode **48** counter electrode **49** reference electrode **50** second working electrode **51** protective membrane **52** first working electrode pad **53** counter electrode pad **54** reference electrode pad **55** second working electrode pad **56** resist film **57** bottom plate **58** middle plate **59** top plate **60** gasket sheet (board) **61** port (additive supply member) **62** connection terminal portion **63** fixing portion **64** slide guide protrusion **65** through-hole **66** sensing electrode **67** positioning unit **68** slide hole **69** fixed portion **70** support portion **71** pressing portion **72** port input/output portion **73** additive addition portion A addition port (upper surface opening) **73***a* recess **73***b* addition port **74** additive addition portion B addition port (upper surface opening) **75** stirring member air discharge and intake port **76** through-hole for first working electrode pad **77** through-hole for counter electrode pad **78** through-hole for reference electrode pad **79** through-hole for second working electrode pad **80** well (culture vessel) **81** stirring member **82** additive addition portion A **83** additive addition portion B **85** additive A container (additive container) **85***a* additive A discharge port (opening) **86** additive B container (additive container) **86***a* additive B discharge port (opening) **87** inclined surface **88** dropping adjustment surface **88***a* inner peripheral side (first surface) **88***b* outer peripheral side (second surface) **89** pipette tip **90** additive **91** additive addition portion A piping line **92** stirring vessel **93** liquid discharge and intake port **94** air discharge and intake port **95** air discharge and intake part **96** seal **96***a* bottom seal **96***aa* entire peeling tab **96***ab* individual peeling tab **96***ac* cut portion **96***ad* perforated portion **96***b* top seal **96***ba* entire peeling tab **96***bh* individual peeling tab **96***bc* cut portion **96***b* perforated portion **97** adhesive portion **98** electrode pad portion **161** port (additive supply member) **175** stirring member air discharge and intake port **181** stirring member **192** stirring vessel **192***a* inclined surface **192***b* horizontal surface **193** liquid discharge and intake port **194** air discharge and intake port **261** port (additive supply member) **281** stirring member **292** stirring vessel **293***a*, **293***h*, **293***c* liquid discharge and intake ports **361** port (additive supply member) **381** stirring member **392** stirring vessel **393** liquid discharge and intake port **394** small-diameter portion **394***a* residual medium **395** rib L**1** liquid level

Claims

1. A sensor unit having a sensor for measuring components of a medium in a culture vessel, comprising: a sensor having a main body portion, a measurement unit that is disposed on the main

body portion and configured to measure the components of the medium, and a connection terminal portion that is electrically connected to the measurement unit; and a board having a connecting portion that is connected to the connection terminal portion of the sensor, and a wiring pattern that is connected to the connecting portion, wherein the sensor has a bent portion in which a connecting part of the main body portion and the board is bent at a substantially right angle so that a measurement portion of the sensor projects downward and is immersed in the medium in the culture vessel in a state in which the connection terminal portion and the connecting portion of the board are connected.

2. The sensor unit according to claim 1, wherein a plurality of the sensors are connected to the board.

3. The sensor unit according to claim 1, further comprising: a bottom plate that is provided below the connection terminal portion of the sensor; a middle plate that is provided above the connection terminal portion of the sensor; and a top plate that is provided above the middle plate.

4. The sensor unit according to claim 3, wherein the connection terminal portion of the sensor is positioned by being sandwiched between the middle plate and the bottom plate from above and below.

5. The sensor unit according to claim 3, wherein the bottom plate has a plurality of through-holes through which passes the sensor that has been bent downwards.

6. The sensor unit according to claim 5, wherein the bottom plate further has a support portion that is provided to an opening edge of the through-hole and configured to support a bottom edge side of the bent portion of the sensor, and the top plate has a pressing portion that is provided to a portion opposite the support portion and configured to push down on an top edge side of the bent portion of the sensor.

7. The sensor unit according to claim 6, wherein the support portion has a curved upper surface shape, and the pressing portion has a curved lower surface shape.

8. The sensor unit according to claim 3, wherein the middle plate and the bottom plate have a sensor positioning portion configured to position the connection terminal portion of the sensor, and a fixing portion configured to fix the middle plate and the bottom plate in a vertical direction with respect to the sensor positioning portion.

9. The sensor unit according to claim 8, wherein the fixing portion includes a prong and a prong mating portion configured to fit together by sliding in a substantially horizontal direction.

10. The sensor unit according to claim 9, wherein the fixing portion further includes a slide guide configured to guide a direction of sliding in the substantially horizontal direction.

11. The sensor unit according to claim 10, wherein a sliding direction is a direction substantially parallel to a diagonal line across the bottom plate.

12. The sensor unit according to claim 1, wherein the sensor is substantially L-shaped, and the bent portion is provided on an upper portion of a lengthwise edge of the sensor.

13. The sensor unit according to claim 1, wherein the sensor is substantially I-shaped, and the bent portion is provided on an upper portion of a lengthwise edge of the sensor.

14. A cell culture analyzer, comprising: the sensor unit according to claim 1; and a culture vessel installation unit on which the sensor unit is placed.

15. The cell culture analyzer according to claim 14, further comprising a support configured to form a housing space in which to install the culture vessel, provided between the sensor unit and the culture vessel installation unit.

16. The cell culture analyzer according to claim 14, further comprising a control unit that is disposed on the sensor unit and controls the sensor unit.