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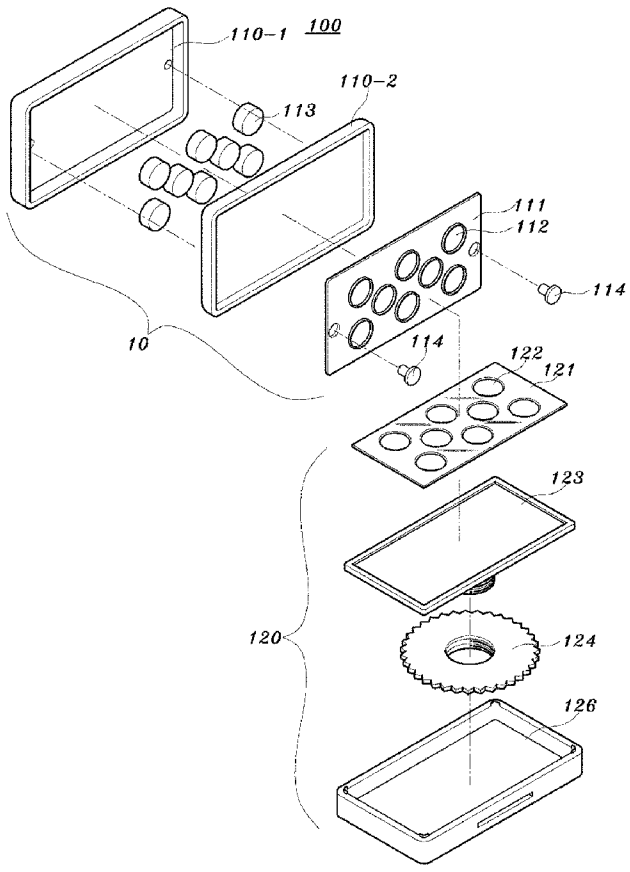
(54) **Title:** APPARATUS AND METHOD FOR MAGNETICALLY SEPARATING BIOLGICAL MATERIALS FROM MIXTURE



(57) **Abstract:** In a prior art, a cell mixture layer is formed and a thickness of the formed layer has been adjusted in a state where a user cannot observe the cell mixture layer. Also, in a process for ho¬ mogenizing the separated specific biological materials, a magnetic field is continuously applied to an upper surface of an upper plate. According to the present invention, there is provided an apparatus for separating biological materials, in which an upper surface of an upper plate is exposed to the outside and a magnetic field applying means is detached from the upper plate so that a magnetic field is not applied, whereby it is possible to conveniently and accurately verify the formation of a biological material mixture layer and whether a gap is set to make the formed layer become a optimum biological material separating state through an exposed upper surface of the upper plate made of a transparent or semitransparent material without forming an additional window, and a homog- enization process is performed in a state where no magnetic field is applied to the upper plate, whereby the separation efficiency of the specific biological materials can be enhanced.

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Description

APPARATUS AND METHOD FOR MAGNETICALLY SEPARATING BIOLOGICAL MATERIALS FROM MIXTURE

Technical Field

[1] The present invention relates to an apparatus and method for separating biological materials, and more particularly, to an apparatus and method for separating biological materials, by applying a magnetic field, from an upper portion of a biological material mixture layer in which specific biological materials such as specific cells, protein, DNA, RNA and the like tagged with magnetic carriers such as magnetic beads are mixed.

Background Art

[2] In PCT/KR2006/001077(WO2006/091060 entitled Apparatus and method for magnetically separating cells from mixture), a cell mixture layer in which specific cells tagged with magnetic beads are mixed is formed between an upper plate and a lower plate, a magnetic field is applied to the upper plate to locate the specific cells on the upper plate and the remainder cells other than the specific cells are simultaneously located on the lower plate by the gravity. Then, the cell mixture layer is separated to separate conveniently and stably the necessary cells.

[3] Here, the cell mixture layer adjusts a gap between the upper and lower plates, so that the cell mixture received in a cell mixture holding portion and having an upward convex-shape is in contact with a lower surface of the upper plate.

[4] Further, in order to securely perform the separation of the cell by means of the magnetic field applied to the specific cells and the gravity exerted on the remainder cells other than the specific cells, the gap between the upper and lower plates of the cell mixture layer formed in the above process is maintained to be wide. However, in order to obtain the optimum cell separation state, the gap between the upper and lower plates is adjusted by operating a dial on which a layer forming location is marked, if necessary. The cells are then separated.

[5] As shown in Fig. 1, an inside of a conventional cell separation apparatus 10 is formed as a closed space to minimize an evaporation of moisture around the layer and a change of temperature. Accordingly, the cell separating process progresses while an optimal environment in which cells are alive is maintained. Also, an upper surface of the upper plate is covered with a magnet applying a magnetic field to the upper surface.

[6] In addition, as shown in Figs. 2 and 3, in a case where the same amount of cell mixture is respectively added into several cell mixture holding portions, the amounts of

cell mixtures added into the cell mixture holding portions may differ from each other, so that the gap between the upper and lower plates forming the optimum layer should be changed.

[7] In addition, as shown in Fig. 4, only by substituting the lower plate and the upper plate with another lower plate having the cell mixture holding portions the number of which differs and a new upper plate corresponding to the new lower plate, it is possible to perform a process for separating the cells having various conditions under the same circumstance in temperature, humidity, and the like at one time.

[8] In this case, according to the number of cell mixture holding portions, a gap between the upper and lower plates forming the layer differs from that between the other upper plate and lower plate. For example, on the lower plate having a certain area, as the number of cell mixture holding portions is increased, the diameter of cell mixture holding portion becomes smaller. Accordingly, as shown in Fig. 5, as the diameter of cell mixture holding portion is small, a gap between the upper and lower plates forming the optimum layer for a separation should become smaller.

[9] As described above, although the amount of cell mixture contained in the cell mixture holding portion of the lower plate and the diameter of the cell mixture holding portion differ from those of the other, the conventional cell separation apparatus has a problem in that in a state where it is impossible to verify the layer from the outside, the dial on which a location at which the layer is formed is marked should be manipulated to form the cell mixture layer and adjust the thickness of the formed layer.

[10] In order to solve the above problem, a transparent window is provided around the side surface on which the layer is formed, so that the user can verify a status of the layer through the transparent window from the outside.

[11] However, in consideration of a size or an installation location of the transparent window, the verification of the layer and the adjustment of the thickness of the layer through the transparent window can be performed with the intense concentration thereon in a state where the location of the transparent window should be coincided with user s eye, thereby causing inconvenience. Furthermore, the transparent window should be additionally provided for enabling a hermetically sealed interior to be maintained, thereby causing the manufacturing process to be more complicated.

[12] Meanwhile, in a case where the remainder cells other than the specific cells adhere to the upper plate after the cell separation process, such cells have a bad influence upon the subsequent experiment, so that there is need to enhance the purity of the separated specific cells to a maximum.

[13] To this end, as shown in Fig. 6, all the remainder cells other than the specific cells separated and placed on the lower plate are removed or the lower plate is substituted with new one, and only buffer solution containing no cell is then added to the lower

plate. Then, the specific cell mixture layer, which is formed by adjusting the gap between the upper and lower plates to be reduced, is shaken vertically to a maximum. Accordingly, the homogenization process, in which the remainder cells other than the specific cells, which adhere to the upper plate together with the specific cells, are detached and become a single cell state, is performed.

[14] In the above cell separation apparatus, however, the homogenization process is performed in a state where a magnetic field the magnitude of which is enough to overcome the gravity exerted on the specific cells is continuously applied to an upper surface of the upper plate. Accordingly, it is difficult to make all the cell lumps adhering to the upper plate and maintaining their shape by the magnetic force become the single cell state as shown in Fig. 7.

Disclosure of Invention

Technical Problem

[15] The present invention is conceived to solve the above problems, an object of the present invention is to provide an apparatus and method for separating biological materials, in which a status of biological material mixture layer can be conveniently and accurately verified through an upper surface of an upper plate exposed to the outside and a homogenization process can be performed in a state where a magnetic field is not applied to the upper plate to enhance the purity of the separated specific biological materials.

Technical Solution

[16] According to the present invention for achieving the objects, there is provided a biological material separation apparatus, comprising: a lower plate having a biological material mixture holding portion provided on an upper surface thereof, the biological material mixture holding portion accommodating a biological material mixture in an upwardly convex shape, the biological material mixture containing specific biological materials tagged with magnetic carriers; an upper plate positioned above the lower plate to face the lower plate, the upper plate having a lower surface to which the biological material mixture accommodated in the biological material mixture holding portion is adsorbed; a magnetic field applying means positioned on an upper surface of the upper plate, the magnetic field applying means being provided detachably from the upper plate; and a gap adjusting means for adjusting a gap between the upper and lower plates, the gap adjusting means being coupled to the upper plate or the lower plate, whereby the gap adjusting means controls the gap between the upper and lower plates to be decreased so that the biological material mixture accommodated in the biological material mixture holding portion is adsorbed to the lower surface of the upper plate and then formed into a biological material mixture layer, and the gap

between the upper and lower plates to be increased so that the specific biological materials moved toward the upper plate by means of a magnetic field applied to the upper plate of the created biological material layer through the magnetic field applying means and the remainder biological materials other than the specific biological materials moved toward the lower plate by means of gravity are separated from each other and respectively positioned on the lower surface of the upper plate and the biological material mixture holding portion of the lower plate, wherein the magnetic field applying means is provided so that the upper surface of the upper plate is exposed to the outside when the magnetic field applying means is detached from the upper plate and the magnetic field is not applied to the upper plate.

[17] Preferably, the biological material separation apparatus may further comprise an upper housing with an upper portion opened downward and a lower housing with a lower portion opened upward, wherein the magnetic field applying means and the upper plate having the upper surface on which the magnetic field applying means is placed are provided in the upper housing, the upper plate being configured so that the upper surface of the upper plate is exposed to the outside when the magnetic field applying means is detached from the upper plate, the lower plate and the gap adjusting means coupled with the lower plate are provided in the lower housing, and the gap adjusting means moves the lower plate vertically to adjust the gap between the upper and lower plates in a state where a lower portion of the upper housing and an upper portion of the lower housing are coupled with each other so that the lower surface of the upper plate and the biological material mixture holding portion of the lower plate are positioned to face each other.

[18] More preferably, the biological material separation apparatus may further comprise a housing with a front face partially opened, wherein the housing is provided therein with the magnetic field applying means, the upper plate having the upper surface on which the magnetic field applying means is placed, and the lower plate installed so that the biological material mixture holding portion thereof faces the lower surface of the upper plate, the gap adjusting means is coupled to the upper plate to move the upper plate vertically thereby adjusting the gap between the upper and lower plates, and the housing is formed so that the upper surface of the upper plate is exposed to the outside when the magnetic field applying means is detached from the upper plate.

[19] According to the present invention for achieving the objects, there is provided a method for separating biological material, comprising: a first step of forming a biological material mixture containing specific biological materials tagged with magnetic beads into a biological material mixture layer by adjusting a gap between upper and lower plates to be decreased so that the biological material mixture which is accommodated in a biological material mixture holding portion of the lower plate in an

upwardly convex shape can be adsorbed to a lower surface of the upper plate positioned to face the biological material mixture holding portion of the lower plate; a second step of moving the specific biological materials toward the upper plate side of the biological material mixture layer by applying a magnetic field to the upper plate side of the biological material mixture layer formed in the first step and simultaneously moving the remainder biological materials other than the specific biological materials toward the lower plate side of the biological material mixture layer by means of gravity; a third step of allowing the specific biological materials and the remainder biological materials other than the specific biological materials respectively moved toward the upper and lower plate sides of the biological material mixture layer in the second step to be separated from each other and respectively positioned on the lower surface of the upper plate and the biological material mixture holding portion of the lower plate while the biological material mixture layer is separated by adjusting the gap between the upper and lower plates to be increased; a fourth step of forming a specific biological material mixture layer by decreasing the gap between the upper and lower plates after removing the remainder biological materials other than the specific biological materials separated in the third step and positioned in the lower plate or replacing the lower plate with a new one and then adding a buffer solution containing no biological material in the lower plate; a fifth step of homogenizing the specific biological material mixture layer by eliminating the magnetic field applied to the specific biological material mixture layer formed in the fourth step and changing the gap between the upper and lower plates repeatedly several times; a sixth step of moving the specific biological materials toward the upper plate side of the specific biological mixture layer by applying a magnetic field to the upper plate side of the specific biological material mixture layer homogenized in the fifth step and simultaneously moving the remainder biological materials other than the specific biological materials toward the lower plate side of the specific biological material layer by means of gravity; and a seventh step of allowing the specific biological materials and the remainder biological materials other than the specific biological materials respectively moved toward the upper and lower plate sides of the specific biological material layer in the sixth step to be separated and respectively positioned on the lower surface of the upper plate and the biological material mixture holding portion of the lower plate when the specific biological material mixture layer is separated by increasing the gap between the upper and lower plates.

Advantageous Effects

[20] According to a biological material separation apparatus of the present invention having the above structure, an upper surface of an upper plate is exposed to the outside

and a magnetic field applying means is detached from the upper plate so as not to apply the magnetic field. Accordingly, when the magnetic field applying means mounted to the upper surface of the upper plate is detached from the upper plate, it is possible to conveniently and accurately verify the formation of biological material mixture layer and whether a gap is set to make the formed layer become an optimum biological material separating state through an exposed, transparent or semitransparent upper surface of the upper plate without forming an additional window. In addition, in a process for additionally separating the remainder biological materials other than the specific biological materials, which is contained in the separated specific biological material, when the magnetic field applying means mounted to the upper surface of the upper plate is detached from the upper plate, a homogenization process is performed in a state where the magnetic field is not applied to the upper plate, whereby a separation efficiency of the specific biological material can be enhanced.

Brief Description of the Drawings

[21] Fig. 1 shows a conventional cell separation apparatus in which a lower body is covered and coupled with an upper body;

[22] Fig. 2 shows one example in which cell mixture is contained in a lower plate of the lower body from which the upper body is separated in Fig. 1;

[23] Fig. 3 shows a state of a cell mixture layer according to the amount of cell mixture contained in the lower plate in Fig. 2;

[24] Fig. 4 shows another example in which cell mixture is contained in the lower plate of the lower body from which the upper body is separated in Fig. 1;

[25] Fig. 5 shows a status of the cell mixture layer according to the diameter of cell mixture contained in the lower plate in Fig. 4;

[26] Fig. 6 shows a state where buffer solution is added into the lower plate to enhance the purity of the specific cells remaining in the separated upper plate when the upper body is separated from the lower body after performing a cell separation process;

[27] Fig. 7 is a sectional view showing a process of homogenizing the specific cell mixture layer in a state where a magnetic field is applied to the specific cell mixture layer formed by covering the lower body with the upper body in Fig. 6;

[28] Fig. 8 is an exploded perspective view of a cell separation apparatus according to one embodiment of the present invention;

[29] Fig. 9 shows that cell mixture is added into a lower plate of a lower body a state where an upper body is divided into a magnet portion and an upper plate portion and the lower body is coupled with the upper body in Fig. 8;

[30] Fig. 10 shows a state where the upper plate portion of the upper body is coupled with the lower plate of the lower body to which the cell mixture is added;

[31] Fig. 11 comparatively shows, from a side surface and an upper surface of the upper plate, a status of a layer formed according to the amount of cell mixture added to the lower plate and a state where a thickness of the layer is optimally adjusted for separation in Fig. 10;

[32] Fig. 12 shows cross sections of peripheries of the upper and lower plates in a case where cells are separated by coupling the magnet portion to the upper plate portion of the upper body in Fig. 8;

[33] Fig. 13 shows a cross section of a periphery of the upper and lower plates in a case where buffer solution is added into a new lower plate to couple the upper body with the lower body in order to enhance the purity of the specific cells remaining in the separate d upper plate in Fig. 12 and the magnet portion of the upper body is then separated from the upper plate portion to eliminate the magnetic field;

[34] Fig. 14 shows cross sections for illustrating a process of homogenizing the specific cell mixture layer by vertically moving the lower plate repeatedly;

[35] Fig. 15 shows cross sections for illustrating a process where peripheries of the upper and lower plates are changed when the cell is separated by applying magnetic field, which is formed by coupling the magnet portion of the upper body with the upper plate portion, to the homogenized specific cell mixture layer;

[36] Figs. 16 and 17 show cell separation apparatuses according to other embodiments of the present invention in which an upper body is divided into a magnet portion and an upper plate portion; and

[37] Fig. 18 shows a cell separation apparatus according to a further embodiment of the present invention in which a magnet is separated from an upper plate.

[38] [Explanation of Reference Numerals for Major Portions Shown in Drawings]

[39] 100, 100', 100": Cell separation apparatus according to embodiments of the present invention

[40] 110: Upper body

[41] 110-1: Upper body magnet portion

[42] 110-2: Upper body upper plate portion 111, 230: Upper plate

[43] 112: Cell mixture adsorbing portion 113, 220: Magnet

[44] 114: Upper plate fixing means 115: Upper housing

[45] 120: Lower body 121, 250: Lower plate

[46] 122: Cell mixture holding portion 123: Lower plate support

[47] 124: Lower plate moving dial 126: Lower housing

[48] 200: Cell separation apparatus according to another embodiment of the present invention

[49] 210: Housing 240: Upper plate support

[50] 260: Lower plate support

[51] 270: Upper plate support moving dial

[52] 280: Upper opening of housing 41: Specific cell

[53] 42: Magnetic bead

[54] 43: Cells other than specific cell

Best Mode for Carrying Out the Invention

[55] Hereinafter, an apparatus for separating biological materials according to embodiments of the present invention will be described in detail with reference to the accompanying drawings, with cell separation as an example.

[56] Fig. 8 is an exploded perspective view of a cell separation apparatus according to one embodiment of the present invention; Fig. 9 shows that cell mixture is added into a lower plate of a lower body a state where an upper body is divided into a magnet portion and an upper plate portion and the lower body is coupled with the upper body in Fig. 8; Fig. 10 shows a state where the upper plate portion of the upper body is coupled with the lower plate of the lower body to which the cell mixture is added; Fig. 11 comparatively shows, from a side surface and an upper surface of the upper plate, a status of a layer formed according to the amount of cell mixture added to the lower plate and a state where a thickness of the layer is optimally adjusted for separation in Fig. 10; Fig. 12 shows cross sections of peripheries of the upper and lower plates in a case where cells are separated by coupling the magnet portion to the upper plate portion of the upper body in Fig. 8; Fig. 13 shows a cross section of a periphery of the upper and lower plates in a case where buffer solution is added into a new lower plate to couple the upper body with the lower body in order to enhance the purity of the specific cells remaining in the separated upper plate in Fig. 12 and the magnet portion of the upper body is then separated from the upper plate portion to eliminate the magnetic field; Fig. 14 shows cross sections for illustrating a process of homogenizing the specific cell mixture layer by vertically moving the lower plate repeatedly; Fig. 15 shows cross sections for illustrating a process where peripheries of the upper and lower plates are changed when the cell is separated by applying magnetic field, which is formed by coupling the magnet portion of the upper body with the upper plate portion, to the homogenized specific cell mixture layer.

[57] In the descriptions of cell separation apparatuses according to embodiments of the present invention, as compared with, for example, among embodiments of PCT/KR2006/001077, a lower plate support 123 which has a bolt-shaped connecting portion formed on a bottom thereof and is vertically moved by rotation of a lower plate support moving dial 124 having a nut-shaped connecting portion threadly engaged with the bolt-shaped connecting portion to thereby move a lower plate 121 vertically, the portions different from PCT/KR2006/001077, particularly verifying the formation

of a layer and the adjustment of a thickness of the formed layer and enhancing a separation efficiency of separated cells, will be mainly described on the basis of the configuration of an upper body including an upper housing, while the portions identical with PCT/KR2006/001077, which are applied as they are, will be omitted.

[58] As shown in Figs. 8 to 12, in a cell separation apparatus 100 according to one embodiment of the present invention comprising an upper body 110 and a lower body 120, a process of separating cells is executed in a state where the lower body 120 containing the cell mixture is covered with the upper body 110 to which a magnetic field is applied and then integrally coupled therewith. Such a process is the same as that disclosed in PCT/KR2006/001077.

[59] Unlike the apparatus disclosed in PCT/KR2006/001077, however, the upper housing, in which a magnet 113 and an upper plate 111 having an upper surface on which the magnet 113 is placed are installed, is formed so that the upper housing can be separated into a portion on which the magnet 113 is provided and the other portion on which the upper plate 111 is installed. In this case, if the magnet 113 is separated from the upper plate 111, an upper surface of the upper plate 111 is exposed to the outside and a magnetic field is not applied.

[60] Accordingly, the upper body 110 is formed such that an upper body magnet portion 110-1 on which the magnet 113 is fixedly provided is detachably coupled with an upper body upper plate portion 110-2 on which the upper plate 111 is fixedly provided by means of an upper plate fixing means 114.

[61] In this case, the upper housing 115 is divided into a magnet portion upper housing on which the magnet 113 is fixedly provided and an upper plate portion upper housing on which the upper plate 111 is fixedly provided by means of the upper plate fixing means 114.

[62] Therefore, as shown in Fig. 10, in the cell separation apparatus 100 according to one embodiment of the present invention, when the upper body magnet portion 110-1 is detached from the upper body upper plate portion 110-2, an upper surface of the upper plate 111 of a transparent or semi-transparent material is exposed to the outside, so that unlike the conventional process in which the separation of the cell can not be verified from the outside, it is possible to verify the formation of a cell mixture layer immediately and conveniently.

[63] In addition, when a gap between the upper plate 111 and a lower plate 121 is adjusted so that a separation of cell requiring the formed layer become an optimum state considering that the gap between the upper and lower plates forming a layer is changed according to the amount of the cell mixture or the diameter of a cell mixture, as shown in Fig. 11, it is more convenient and accurate to verify the diameter of the layer through an upper surface than to verify the thickness of the layer formed between

the upper and lower plates.

[64] That is, the cell separation apparatus 100 according to one embodiment of the present invention is disposed on an appropriate location and an upper surface of the upper plate 111 exposed to the outside is observed after detaching the magnet 111 from the upper plate 111, whereby it is possible to conveniently verify whether the layer is formed and the thickness of the formed layer is appropriately set. If a guide line for forming the optimum layer is marked on an upper surface of the upper plate 111, it is possible to more conveniently verify whether the layer is formed and the thickness of the formed layer is appropriately set. In this case, since the upper plate 111 which is exposed to the outside is made of a transparent or semi-transparent material, there is no need to provide an additional window, and the inside is maintained in a hermetically sealed state, whereby the apparatus can be easily manufactured.

[65] Accordingly, in a state where the cell mixture is added into a cell mixture holding portion 122 of the lower plate 121 placed in a lower housing 126 of the lower body 120, the lower body 120 is covered and coupled with the upper body 110 in which the upper body magnet portion 110-1 is detached from the upper body upper plate portion 110-2. In addition, a vertical location of the lower plate 121 is adjusted by moving vertically a lower plate support 123 with a lower plate support moving dial 124, whereby a user can observe an upper surface of the upper plate 111 exposed to the outside and verify the cell mixture layer being in contact with a cell mixture adsorbing portion 112 of the upper plate 111 and whether the thickness of the formed layer is optimally set.

[66] In addition, as shown in Fig. 12, the upper plate magnet portion 110-1 is coupled with the upper body upper plate portion 110-2, on which the thickness of the layer is set, thereby applying the magnetic field to the upper plate 111 for allowing the specific cells to be moved toward and placed on the upper plate 111. Simultaneously, the cell mixture layer is separated after a certain time, for example, after 7 to 15 minutes so that the remainder cells other than the specific cells are placed on the lower plate 121 by means of the gravity.

[67] As a result, the specific cells and the other cells of the separated cell mixture layer are placed on the upper plate 111 and the lower plate 121, respectively.

[68] Here, in order to enhance the purity of the specific cells remaining in the cell mixture adsorbing portion 112 of the upper plate 111, the mixture of the other cells of the lower plate 121 besides the specific cells is entirely removed or the lower plate is replaced with new one, and buffer solution is then added into the lower plate. Thereafter, the lower body 120 is covered and coupled with the upper body 110, and then, the lower plate 121 is caused to move, whereby the specific cell mixture layer is formed. Here, the magnetic field is still applied to the specific cells that are moved to

the upper plate 111 in the specific cell mixture and the moved specific cells are simultaneously in a state where the gravity is exerted downward to obviously separate the cells, so that the magnetic field is eliminated for effectively performing the homogenization process.

[69] In the embodiments of the present invention, the magnet 113 is detachably provided on the upper surface of the upper plate 111. As shown in Fig. 13, the upper body magnet portion 110-1 is separated from the upper body upper plate portion 110-2 in the upper body 110, so that the applied magnet field does not have influence on the upper plate 111. As a result, since only the gravity is exerted on the specific cell mixture layer, the cells are separated from the upper plate 111 and moved toward the lower plate 121.

[70] Then, as shown in Fig. 14, in a state where the magnetic field is eliminated from the upper plate 111, the lower plate support moving dial 124 is rotated from side to side to vertically move the lower plate left 121 repeatedly. By performing the above operation, the gap between the upper plate 111 and the lower plate 121 is changed repeatedly several times, for example, 10 times or less and the specific cell mixture layer is homogenized while the specific cell mixture layer is maintained.

[71] In such a case, vibration energy is transmitted to the respectively lumped cells to break the cohesion between the cells, so that the other cells adhering to the peripheries of the specific cells are separated. Consequently, all the cells become a single cell state and are uniformly distributed throughout the specific cell mixture layer.

[72] Here, in a case where the homogenization process in which the separation and formation of the specific cell mixture layer are repeated is carried out, the vibration energy transmitted to the respectively cells is increased. Accordingly, as compared with a case where the specific cell mixture layer is maintained, the single cell state can be easily obtained.

[73] Since the homogenization process can be verified through the upper surface of the upper plate 111 exposed to the outside, the magnitude of the vibration energy transmitted to the cells can be easily adjusted.

[74] After the homogenization process is performed, as shown in Fig. 15, the upper body magnet body 110-1 is again coupled with the upper body upper plate portion 110-2 and then left for 7 to 15 minutes as it is. As a result, the magnetic field applied to the upper plate side of the homogenized specific cell mixture layer through the magnet 113 causes the specific cells to move toward the upper plate side of the specific cell mixture layer, and the remainder cells other than the specific cells are simultaneously moved toward the lower plate side of the specific cell mixture layer by the gravity.

[75] In such a state, if the lower plate support moving dial 124 is rotated to separate the specific cell mixture layer, the specific cells the remainder cells other than the specific

cells are separated from each other and respectively placed on the upper plate 111 and the lower plate 121, so that the other cells contained in the separated specific cells are additionally removed, so that the purity of the necessary specific cells can be enhanced.

[76] The magnetic field applied when the homogenization process is performed does not have influence on the upper plate 111, so that the entire efficiency can be increased by 1 to 3 % as compared with a case where a magnetic field has influence on the upper plate in PCT/KR2006/001077.

[77] In addition, the homogenization process makes it possible to separate the upper body magnet portion 110-1 from the upper body upper plate portion 110-2 before forming the specific cell mixture layer.

[78] In this case, in a state where the cells in the specific cell mixture of the upper plate 111 is located in a lower side by the gravity, the lower plate 121 is moved, so that the specific cell mixture of the upper plate 111 is brought into contact with the buffer solution of the lower plate to form the specific cell mixture layer. Accordingly, the single cell state can be easily and rapidly achieved.

[79] Figs. 16 and 17 show cell separation apparatuses according to other embodiments of the present invention in which an upper body is divided into a magnet portion and an upper plate portion.

[80] Unlike the previous embodiment, the cell separation apparatuses 100' and 100" according to the other embodiments of the present invention are configured such that the upper body magnet portion 110-1 is accommodated in or drawn out of the upper body upper plate portion 110-2 or the upper body magnet portion 110-1 is lifted up or put on the upper body upper plate portion by rotation thereof about a hinge provided at one side. Like the previous embodiment, the magnetic field is eliminated in a state where the upper surface of the upper plate 111 is exposed to the outside or the magnetic field is applied while the upper surface of the upper plate 111 is covered.

[81] Fig. 18 shows a cell separation apparatus according to a further embodiment of the present invention in which a magnet is separated from an upper plate.

[82] In a cell separation apparatus 200 according to the further embodiment of the present invention, in a state where an upper plate 230 and a lower plate 250 are provided in one housing 210, the upper plate 230 is moved to adjust a gap between the upper plate 230 and the lower plate 250, thereby separating the cells.

[83] Specifically, as shown in Fig. 18, in the cell separation apparatus 200 according to the further embodiment of the present invention, an upper plate support 240 and a lower plate support 260 are provided in the housing 210 with a front face partially opened so that they can be moved forward and rearward.

[84] Here, the lower plate support 260 is fixedly installed to a lower portion of the

housing 210, and the upper plate support 240 is placed at an upper portion to be spaced from the lower plate support 260 by a certain distance. In addition, the upper plate support is installed to be moved in the housing 210 upward and downward.

[85] The upper plate 230 and the lower plate 250 are provided such that a cell mixture adsorption portion of the upper plate 230 and a cell mixture receiving portion of the lower plate 250 are positioned to face each other in grooves formed on the upper plate support 240 and the lower plate support 260.

[86] In addition, the magnetic 220 is installed above the upper plate support 240 to face the upper surface of the upper plate 230. The magnet 220 is installed to be detachable from the housing 210 forward and rearward, thereby causing the magnet to be completely detached from the upper plate 230.

[87] In this case, the upper portion of the housing 210 is opened to form an upper opening 280 of the housing or the housing is made of transparent material so that the upper surface of the upper plate 230 is exposed to the outside when the magnet 220 is detached from the upper surface of the upper plate 230.

[88] In addition, the upper plate support 240 is meshed (not shown in the drawing) with an upper plate support moving dial 270 provided at one outside of the housing 210, thereby being moved upward and downward according to the rotation of the upper plate support moving dial 270. Here, in a case where the upper plate support 240 is moved upward and downward, the magnet 220 and the upper plate 230 are moved together.

[89] The present invention is not limited to the aforementioned embodiments and various modifications can be made thereto within the scope of the invention defined by the appended claims. Such modifications fall within the scope of the present invention.

[90] In the cell separation apparatus according to the embodiments of the present invention, although the upper or lower plate is moved or the magnet is detached and coupled by the direct manipulation of a user, a driving means, such as a motor, and a microprocessor for controlling the driving means by a program may be utilized to perform the automatic operation based on the predetermined algorithm instead of a direct operation of the user.

[91] In this case, the upper plate and the lower plate are automatically operated for a pre-determined time and the number of times by a control program to increase or reduce the gap between the upper and lower plates within a range in which the cell mixture layer is maintained, so that the remainder cells other than the specific cells placed on the upper plate are moved to the lower plate by the gravity to enable the separation efficiency to be further increased.

Industrial Applicability

[92] An apparatus and method for separating biological materials of the present invention can be utilized as a multi-functional cell separator capable of verifying the process for separating biological materials conveniently and accurately and enhancing the separation efficiency.

Claims

[1] A biological material separation apparatus, comprising:
a lower plate having a biological material mixture holding portion provided on an upper surface thereof, the biological material mixture holding portion accommodating a biological material mixture in an upwardly convex shape, the biological material mixture containing specific biological materials tagged with magnetic carriers;
an upper plate positioned above the lower plate to face the lower plate, the upper plate having a lower surface to which the biological material mixture accommodated in the biological material mixture holding portion is adsorbed;
a magnetic field applying means positioned on an upper surface of the upper plate, the magnetic field applying means being provided detachably from the upper plate; and
a gap adjusting means for adjusting a gap between the upper and lower plates, the gap adjusting means being coupled to the upper plate or the lower plate, whereby the gap adjusting means controls the gap between the upper and lower plates to be decreased so that the biological material mixture accommodated in the biological material mixture holding portion is adsorbed to the lower surface of the upper plate and then formed into a biological material mixture layer, and the gap between the upper and lower plates to be increased so that the specific biological materials moved toward the upper plate by means of a magnetic field applied to the upper plate of the created biological material layer through the magnetic field applying means and the remainder biological materials other than the specific biological materials moved toward the lower plate by means of gravity are separated from each other and respectively positioned on the lower surface of the upper plate and the biological material mixture holding portion of the lower plate,
wherein the magnetic field applying means is provided so that the upper surface of the upper plate is exposed to the outside when the magnetic field applying means is detached from the upper plate and the magnetic field is not applied to the upper plate.

[2] The biological material separation apparatus as claimed in claim 1, further comprising an upper housing with an upper portion opened downward and a lower housing with a lower portion opened upward,
wherein the magnetic field applying means and the upper plate having the upper surface on which the magnetic field applying means is placed are provided in the upper housing, the upper plate being configured so that the upper surface of the

upper plate is exposed to the outside when the magnetic field applying means is detached from the upper plate,

the lower plate and the gap adjusting means coupled with the lower plate are provided in the lower housing, and

the gap adjusting means moves the lower plate vertically to adjust the gap between the upper and lower plates in a state where a lower portion of the upper housing and an upper portion of the lower housing are coupled with each other so that the lower surface of the upper plate and the biological material mixture holding portion of the lower plate are positioned to face each other.

[3] The biological material separation apparatus as claimed in claim 1, further comprising a housing with a front face partially opened,

wherein the housing is provided therein with the magnetic field applying means, the upper plate having the upper surface on which the magnetic field applying means is placed, and the lower plate installed so that the biological material mixture holding portion thereof faces the lower surface of the upper plate,

the gap adjusting means is coupled to the upper plate to move the upper plate vertically thereby adjusting the gap between the upper and lower plates, and

the housing is formed so that the upper surface of the upper plate is exposed to the outside when the magnetic field applying means is detached from the upper plate.

[4] A method for separating biological material, comprising:

a first step of forming a biological material mixture containing specific biological materials tagged with magnetic beads into a biological material mixture layer by adjusting a gap between upper and lower plates to be decreased so that the biological material mixture which is accommodated in a biological material mixture holding portion of the lower plate in an upwardly convex shape can be adsorbed to a lower surface of the upper plate positioned to face the biological material mixture holding portion of the lower plate;

a second step of moving the specific biological materials toward the upper plate side of the biological material mixture layer by applying a magnetic field to the upper plate side of the biological material mixture layer formed in the first step and simultaneously moving the remainder biological materials other than the specific biological materials toward the lower plate side of the biological material mixture layer by means of gravity;

a third step of allowing the specific biological materials and the remainder biological materials other than the specific biological materials respectively moved toward the upper and lower plate sides of the biological material mixture layer in the second step to be separated from each other and respectively

positioned on the lower surface of the upper plate and the biological material mixture holding portion of the lower plate while the biological material mixture layer is separated by adjusting the gap between the upper and lower plates to be increased;

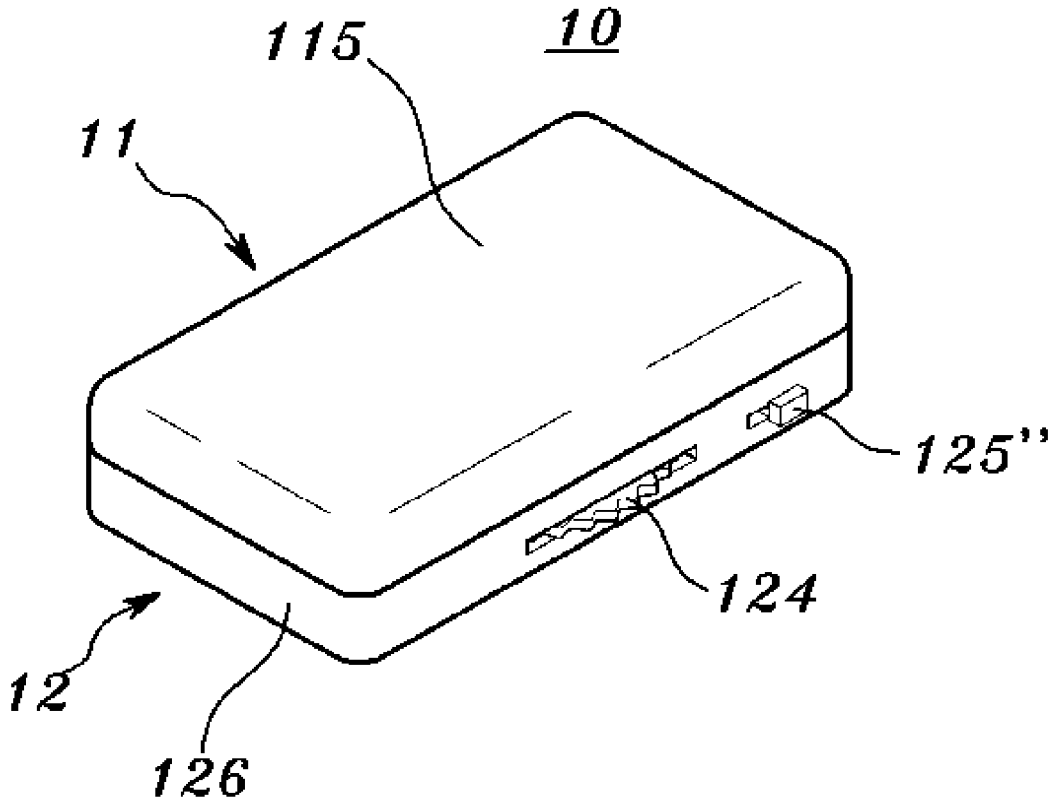
a fourth step of forming a specific biological material mixture layer by decreasing the gap between the upper and lower plates after removing the remainder biological materials other than the specific biological materials separated in the third step and positioned in the lower plate or replacing the lower plate with a new one and then adding a buffer solution containing no biological material in the lower plate;

a fifth step of homogenizing the specific biological material mixture layer by eliminating the magnetic field applied to the specific biological material mixture layer formed in the fourth step and changing the gap between the upper and lower plates repeatedly several times;

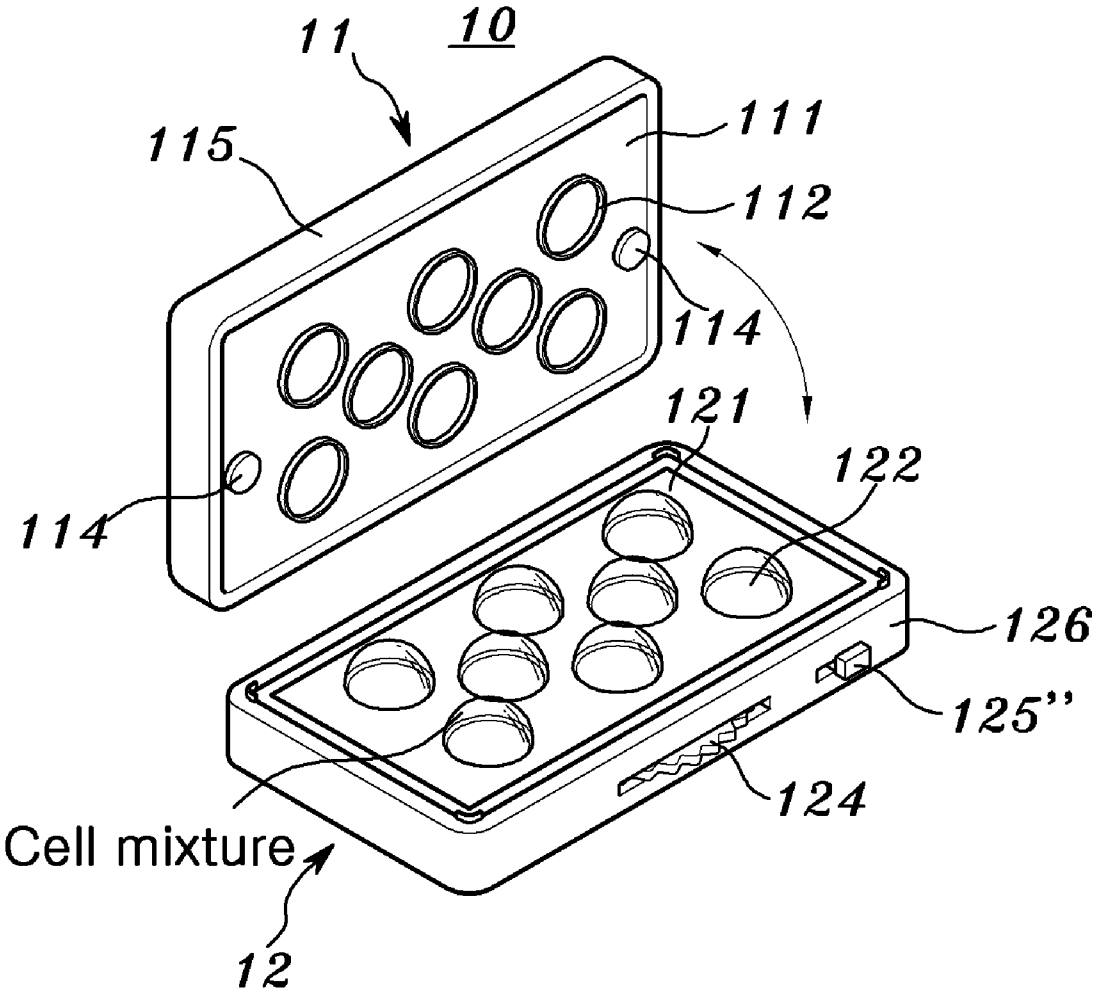
a sixth step of moving the specific biological materials toward the upper plate side of the specific biological mixture layer by applying a magnetic field to the upper plate side of the specific biological material mixture layer homogenized in the fifth step and simultaneously moving the remainder biological materials other than the specific biological materials toward the lower plate side of the specific biological material layer by means of gravity; and

a seventh step of allowing the specific biological materials and the remainder biological materials other than the specific biological materials respectively moved toward the upper and lower plate sides of the specific biological material layer in the sixth step to be separated and respectively positioned on the lower surface of the upper plate and the biological material mixture holding portion of the lower plate when the specific biological material mixture layer is separated by increasing the gap between the upper and lower plates.

[Fig. 1]



[Fig. 2]



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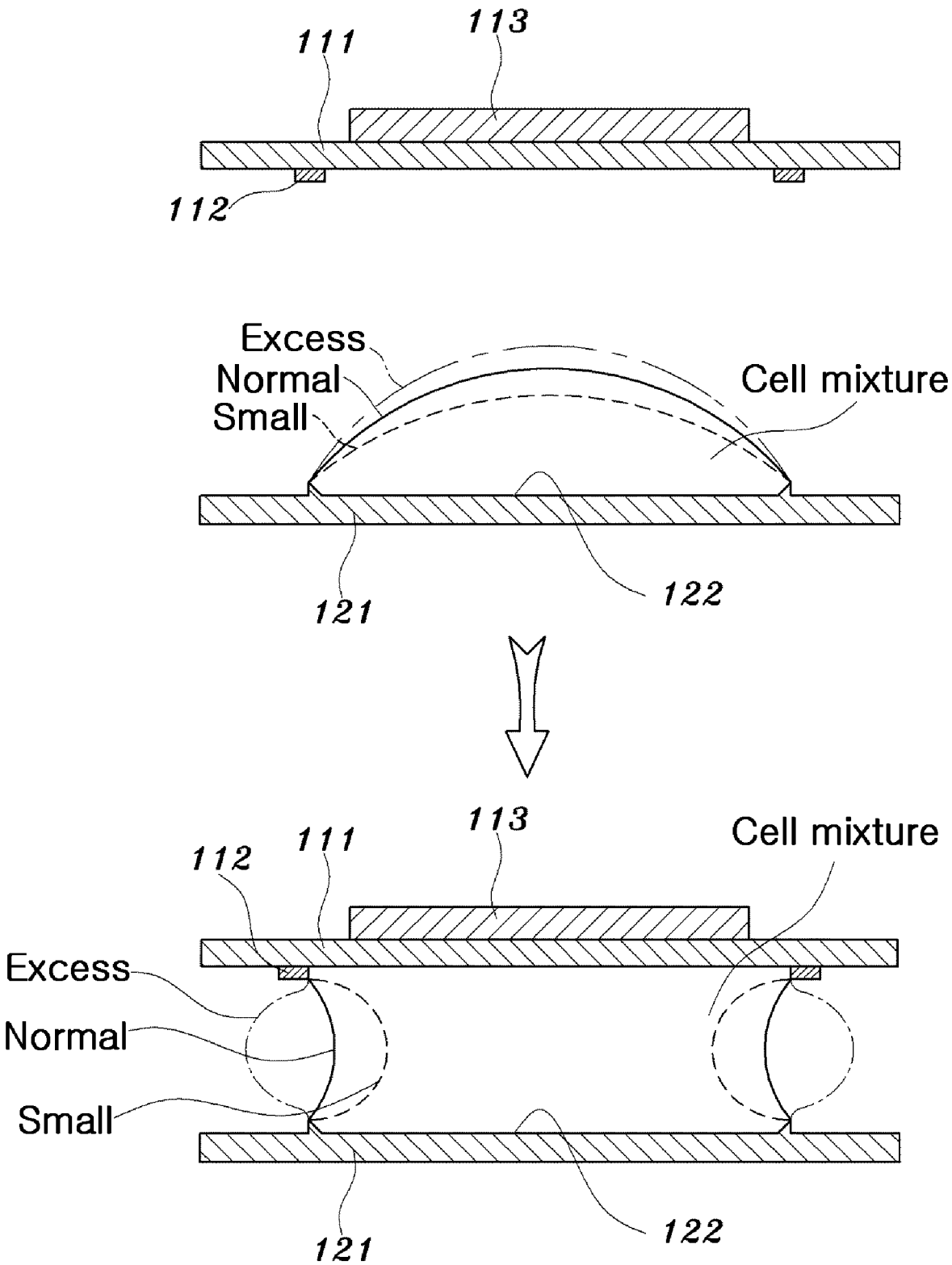
111
113
112
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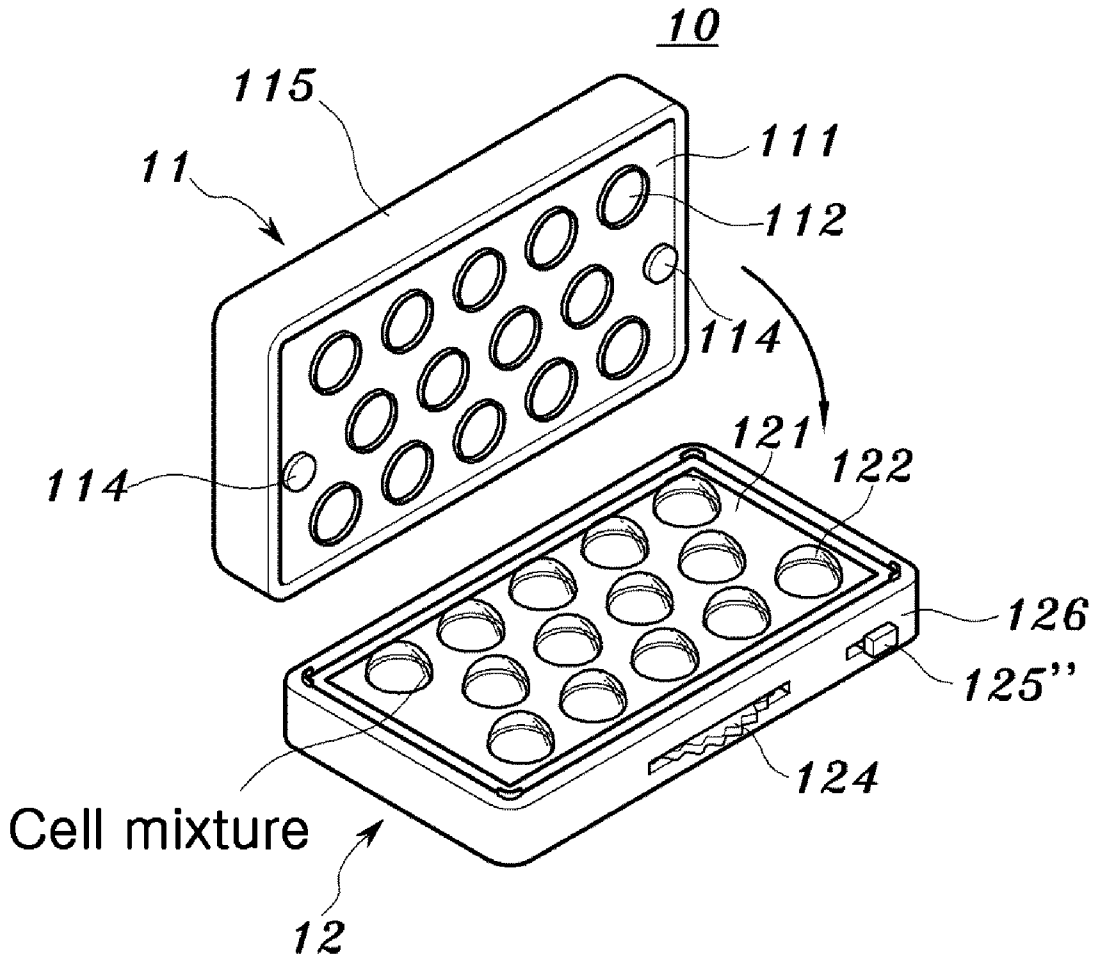
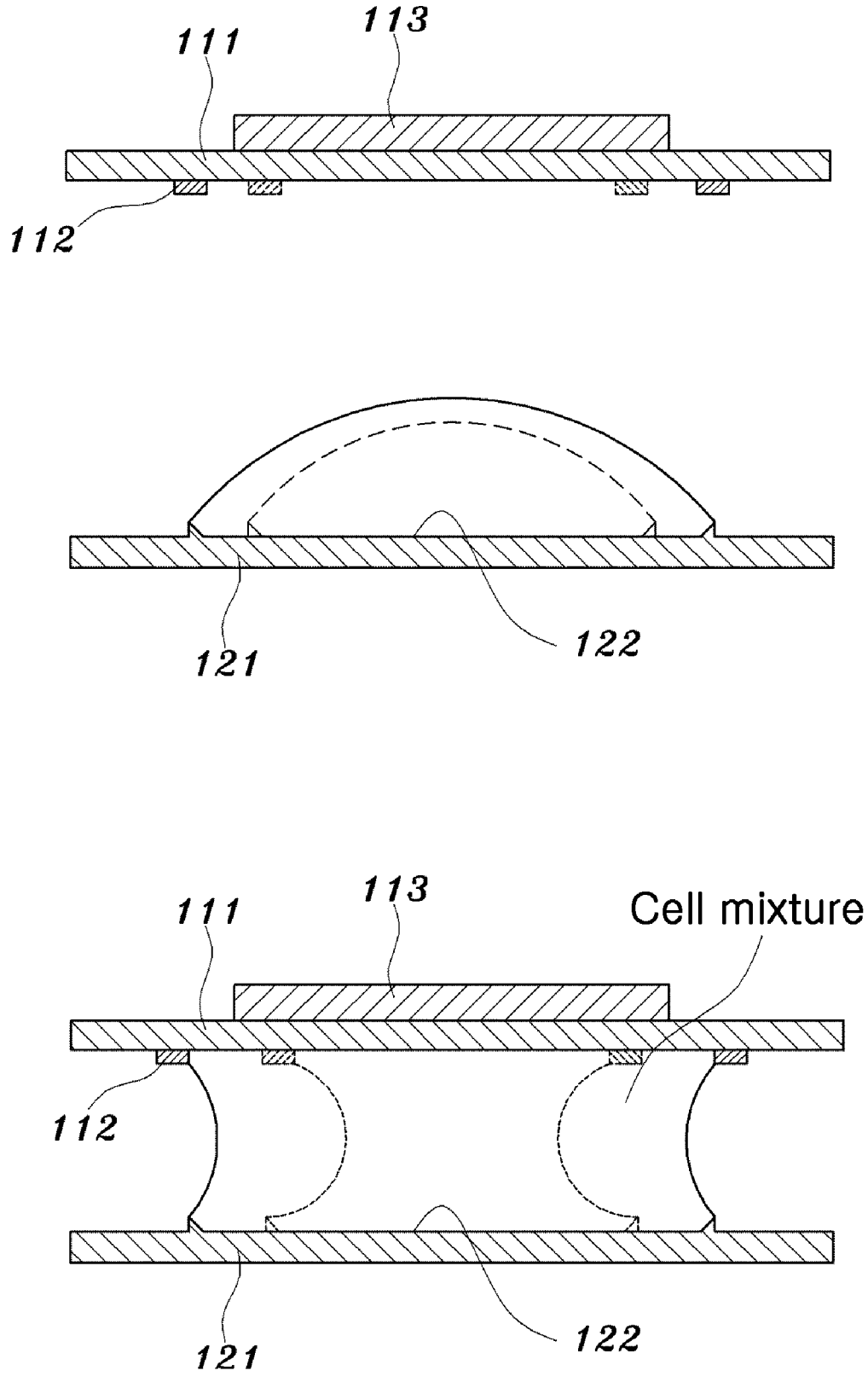
Normal
Small
Cell mixture
121
122
113
112
111
Cell mixture
Excess
Normal
Small
121
122

[Fig. 4]



[Fig. 5]



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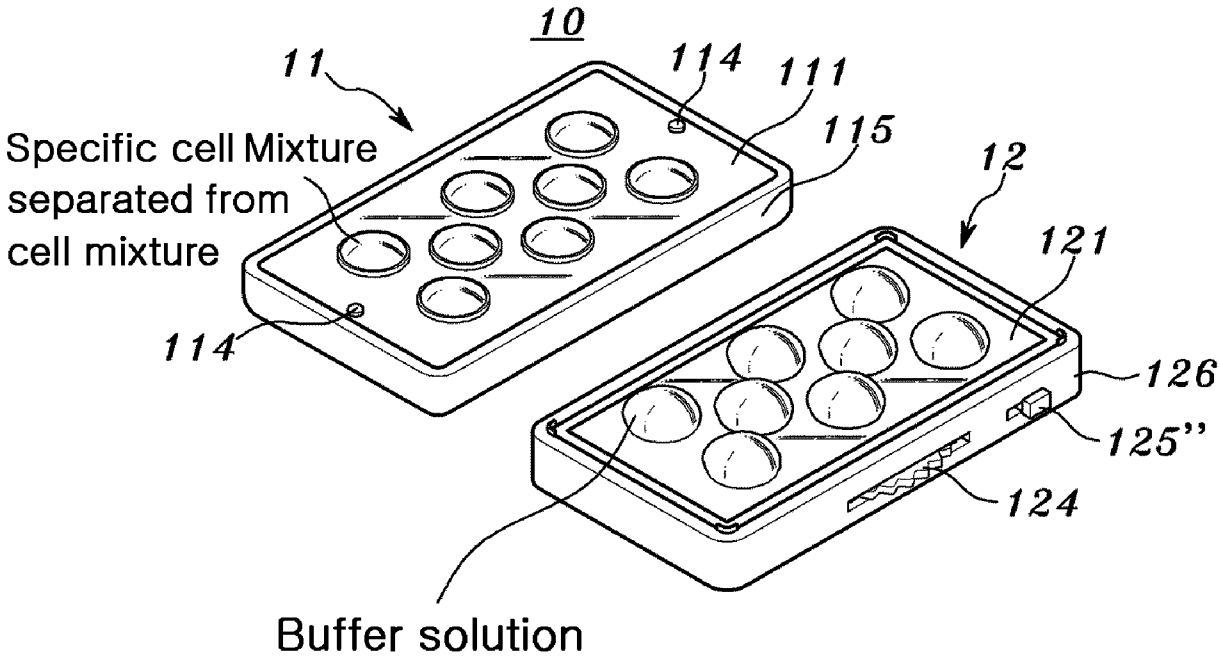
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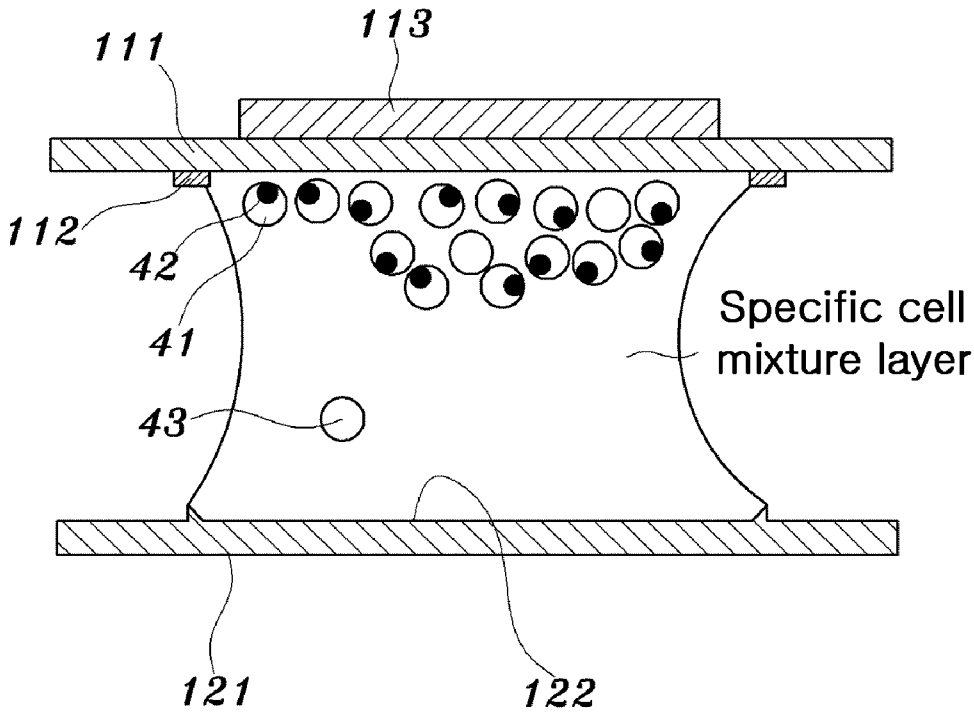
114
111
115
12
121
Specific cell Mixture separated from cell mixture
114
126
125''
124
Buffer solution

[Fig. 7]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41
43
Specific cell
mixture layer
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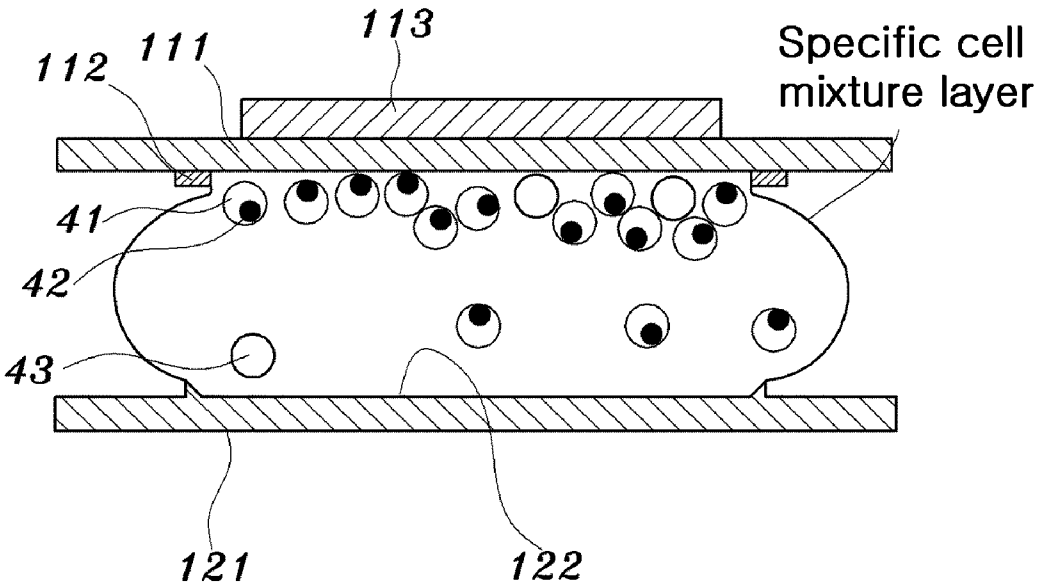
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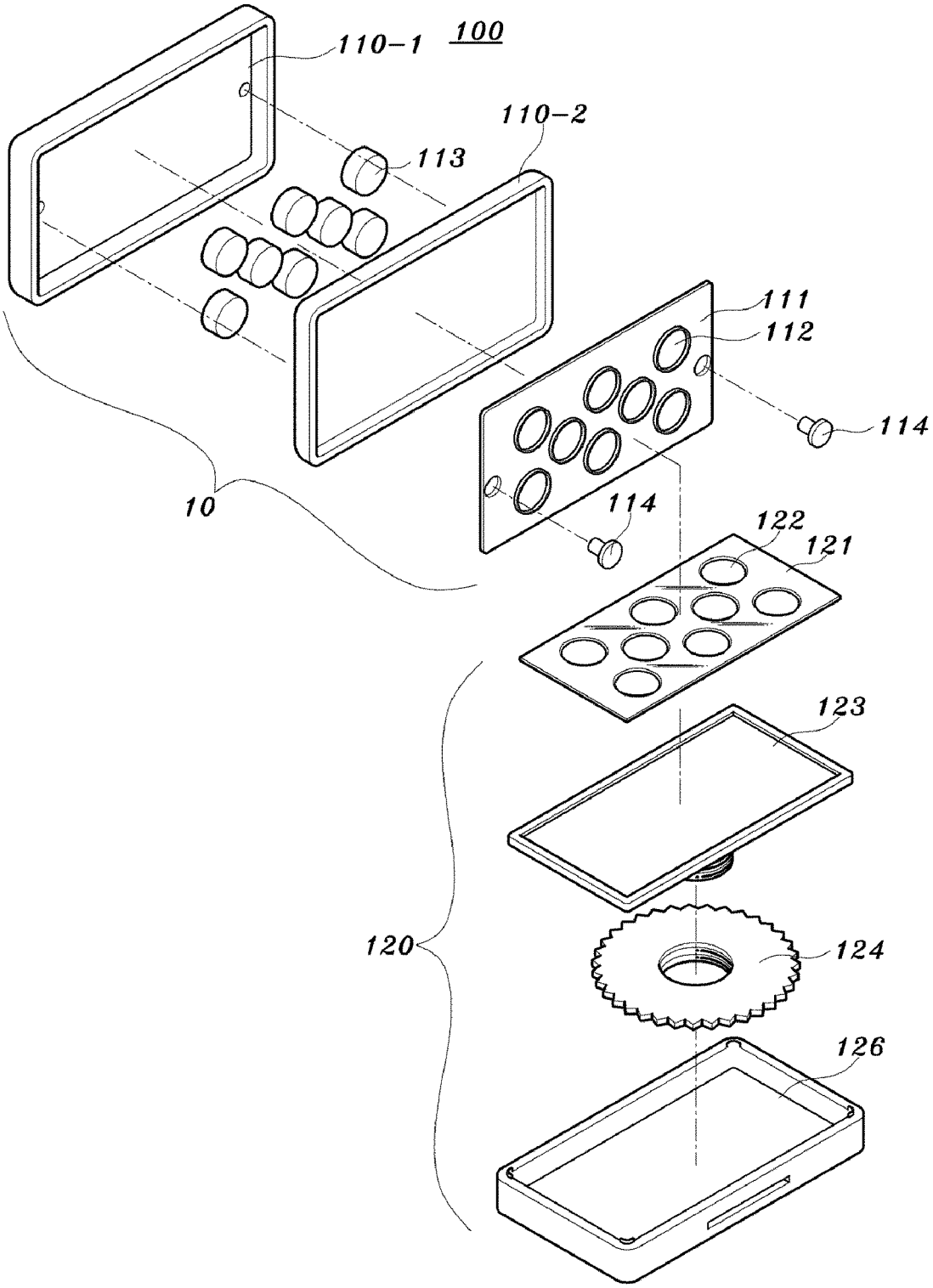
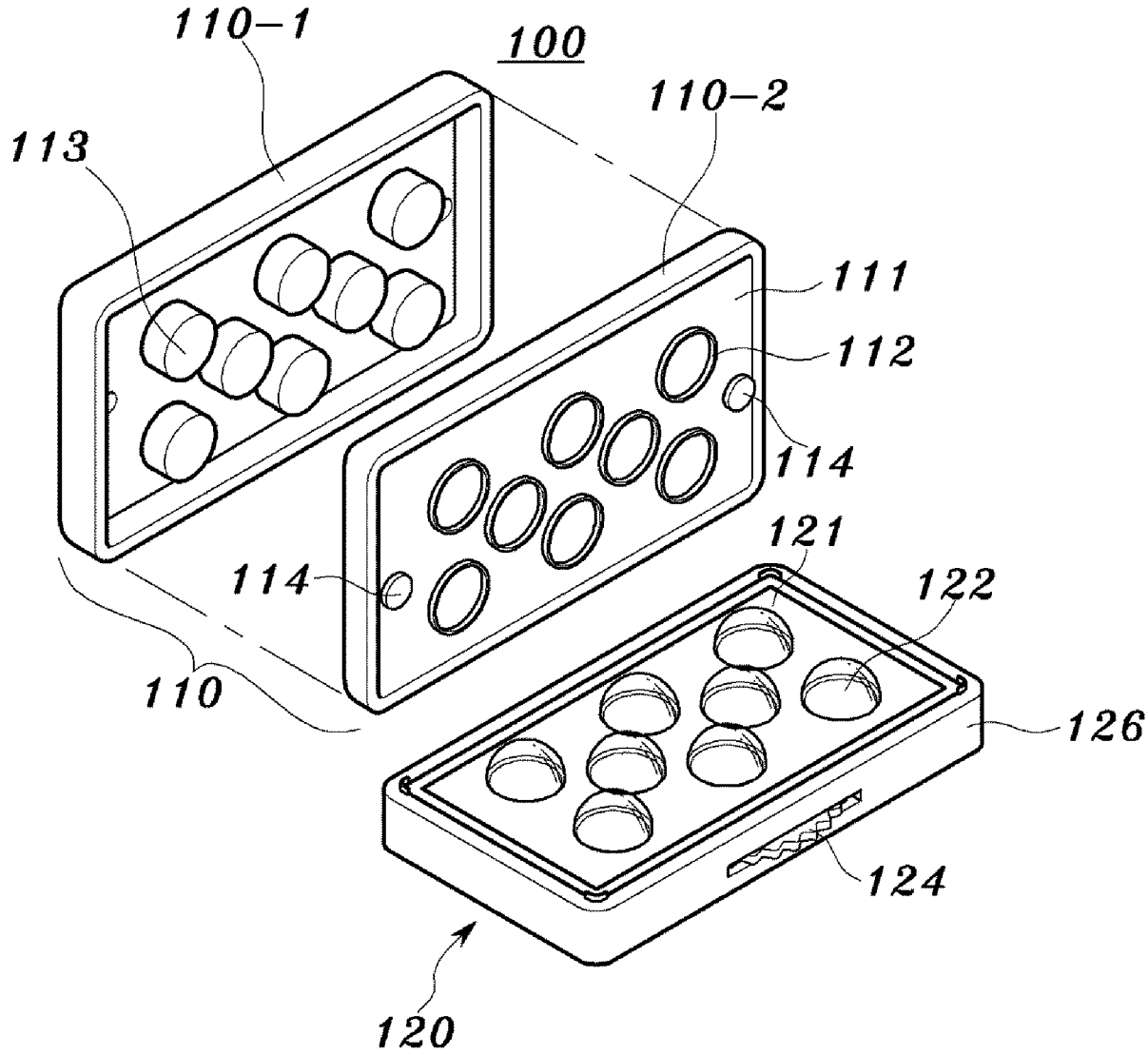
112
111
Specific cell
mixture layer
41
42
43
121
122

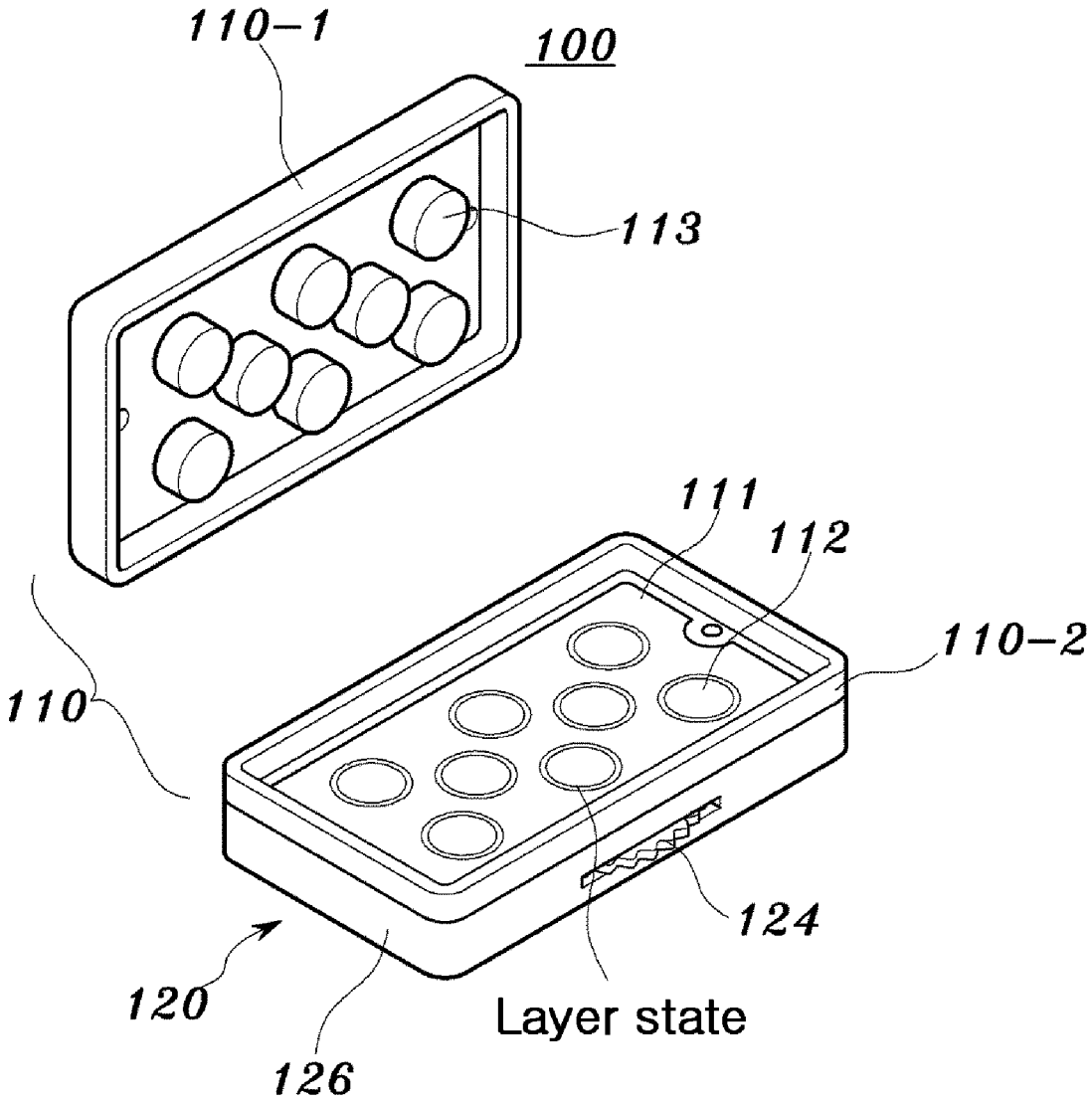
[Fig. 8]



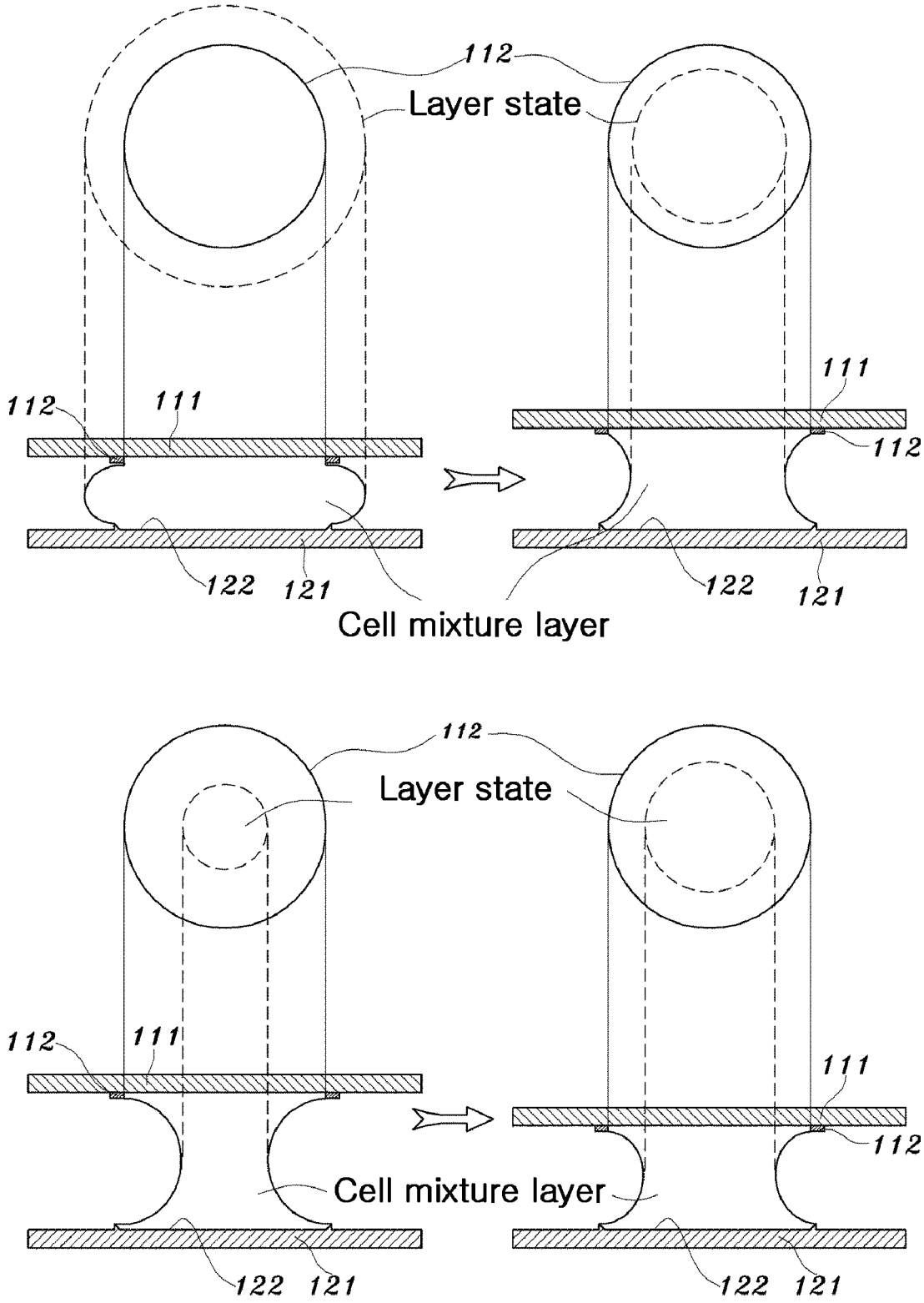
[Fig. 9]



[Fig. 10]



[Fig. 11]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111
111
112
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121
Cell mixture layer
112
Layer state
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111
112
Cell mixture layer
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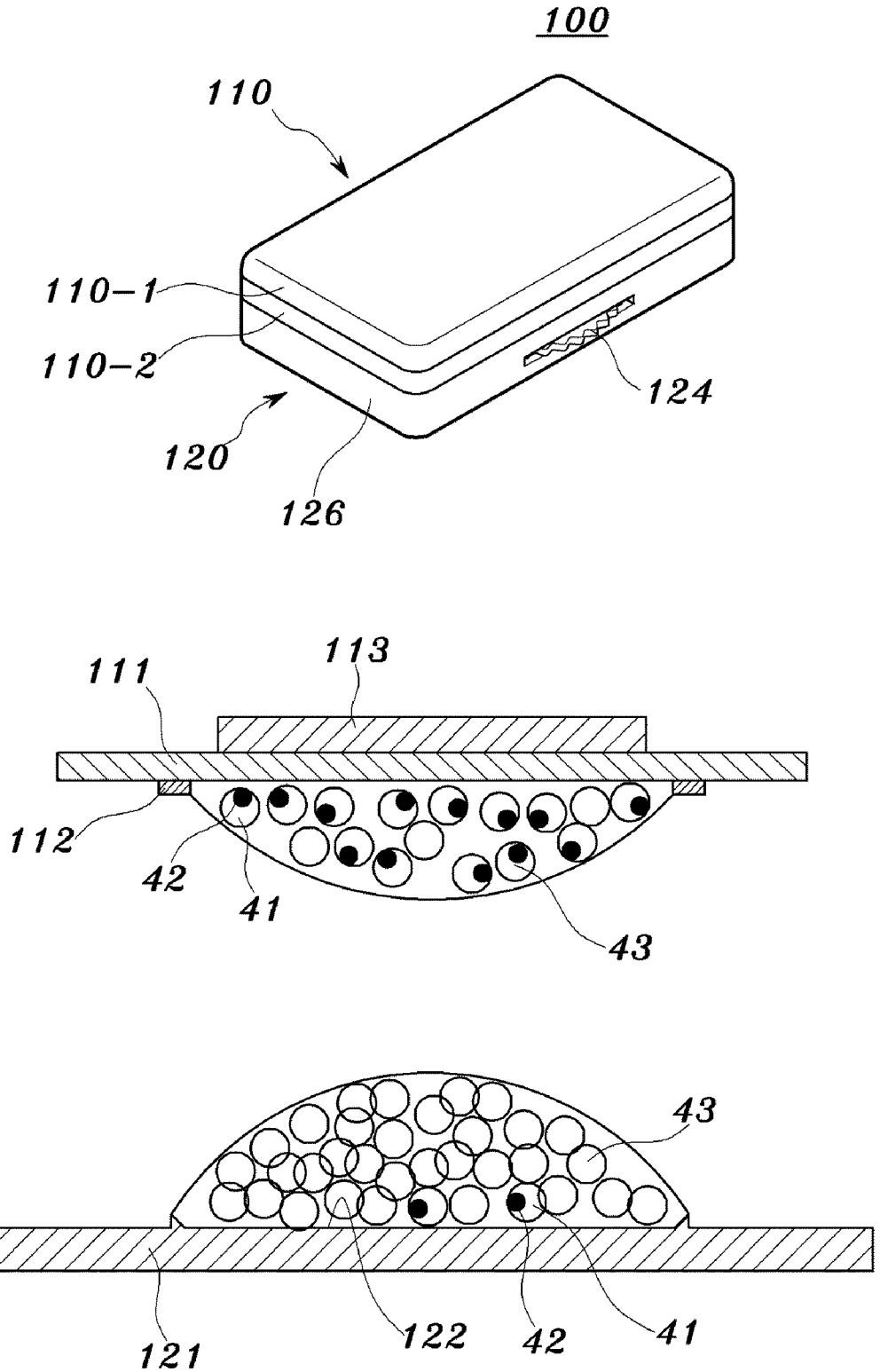
121
122
121

[Fig. 12]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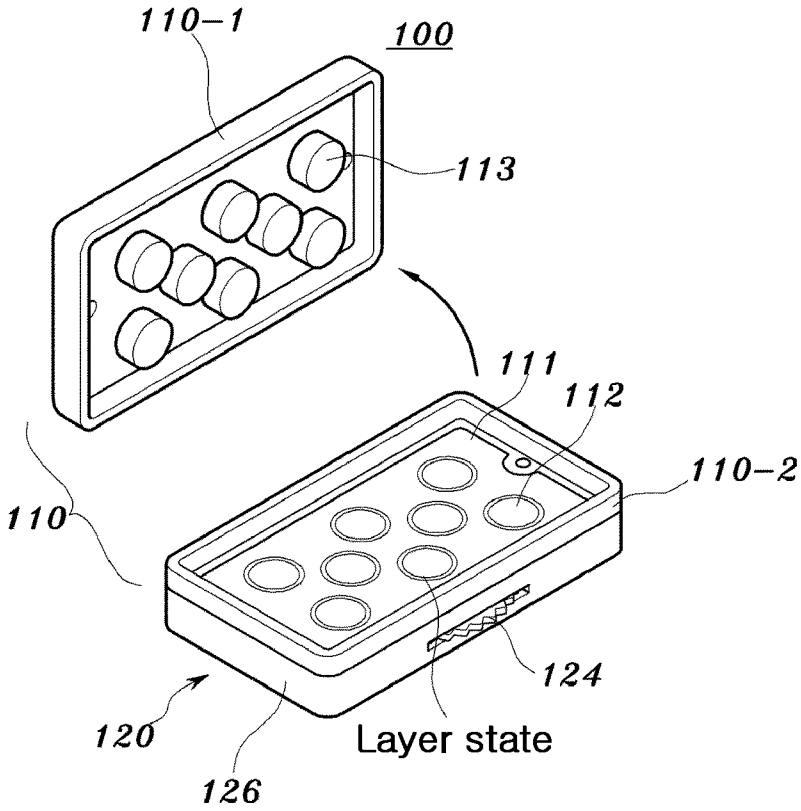
42
41

[Fig. 13]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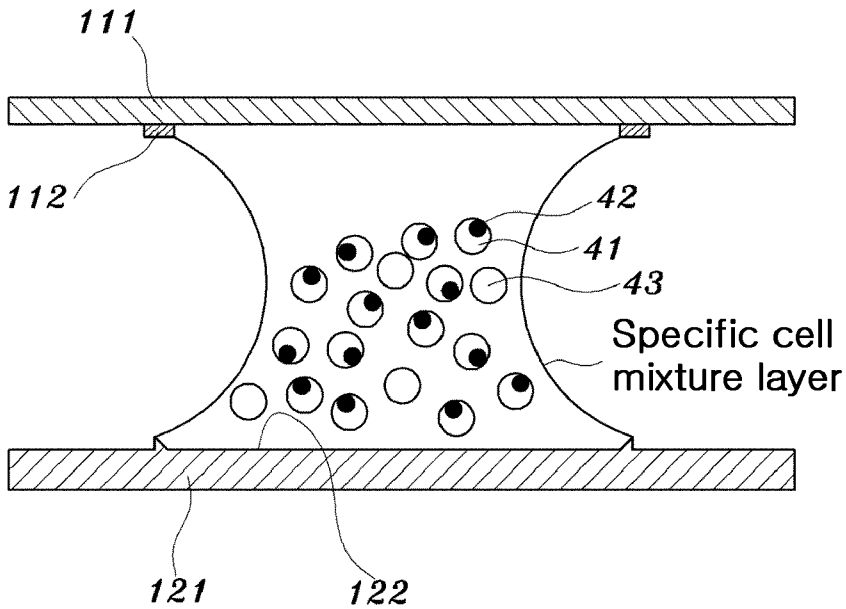
Layer state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112
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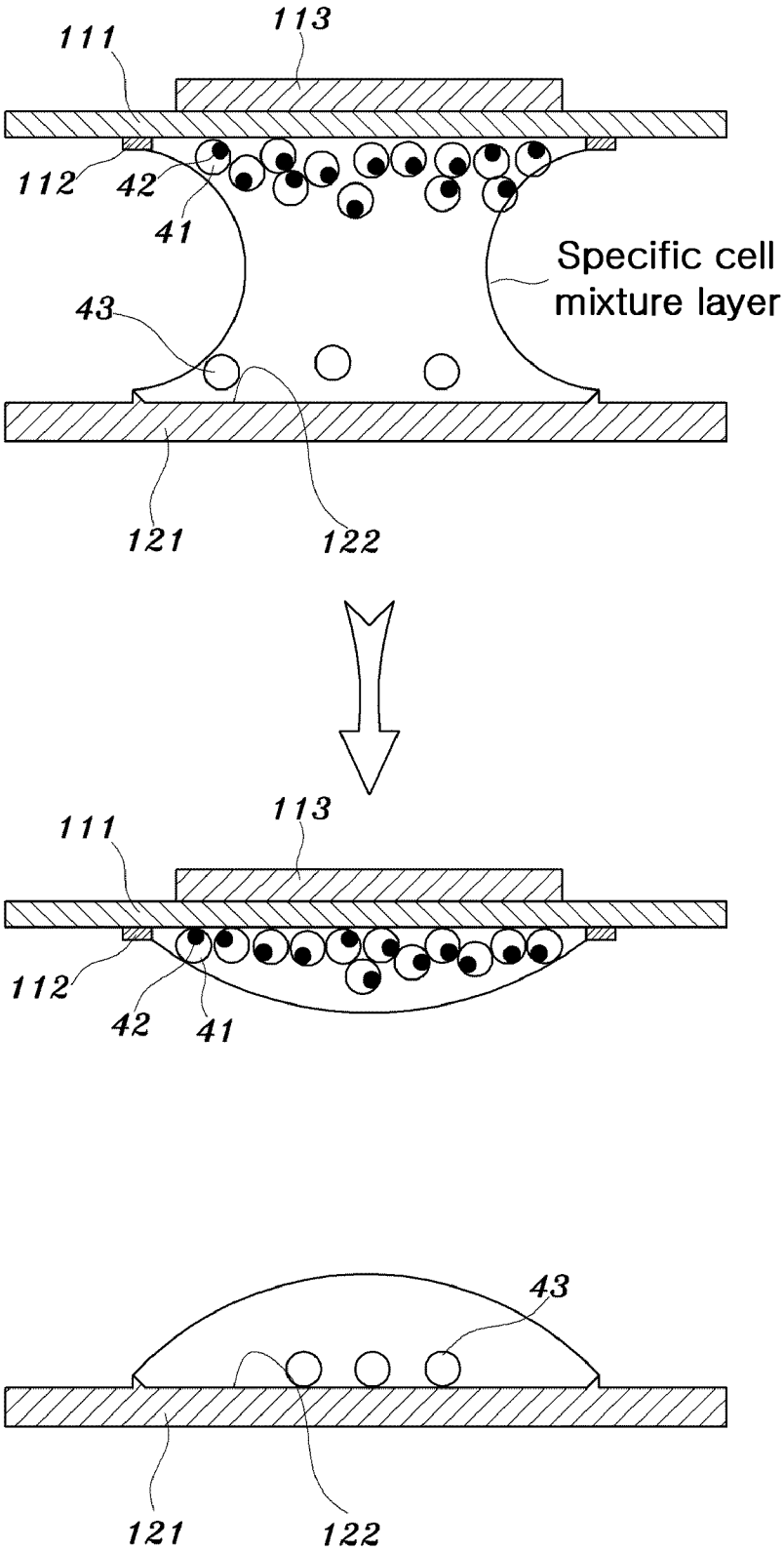
41
43
Specific cell
mixture layer
121
122

[Fig. 14]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41
Specific cell
mixture layer
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122
111
113
112
42
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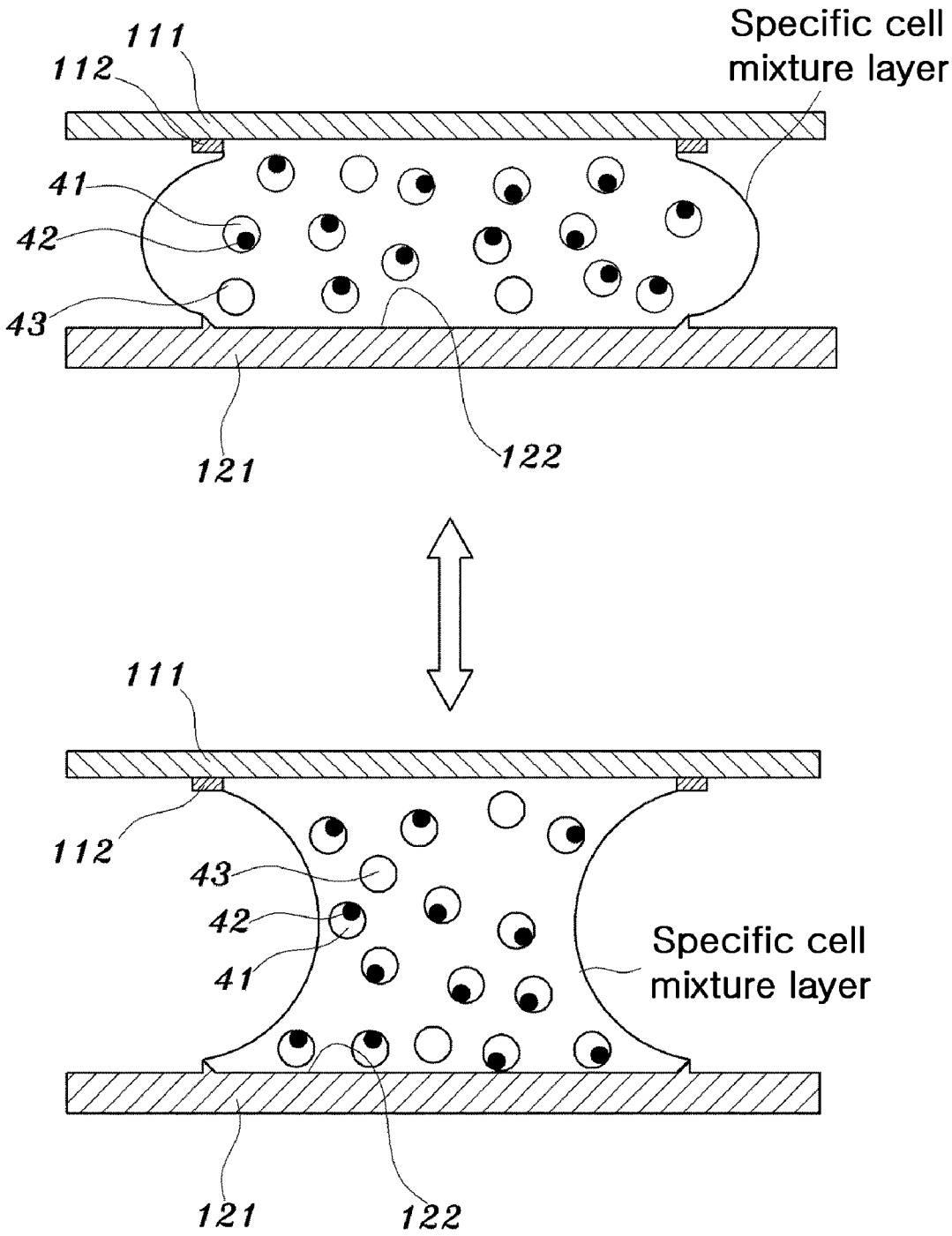
43
121
122

[Fig. 15]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112
Specific cell mixture layer
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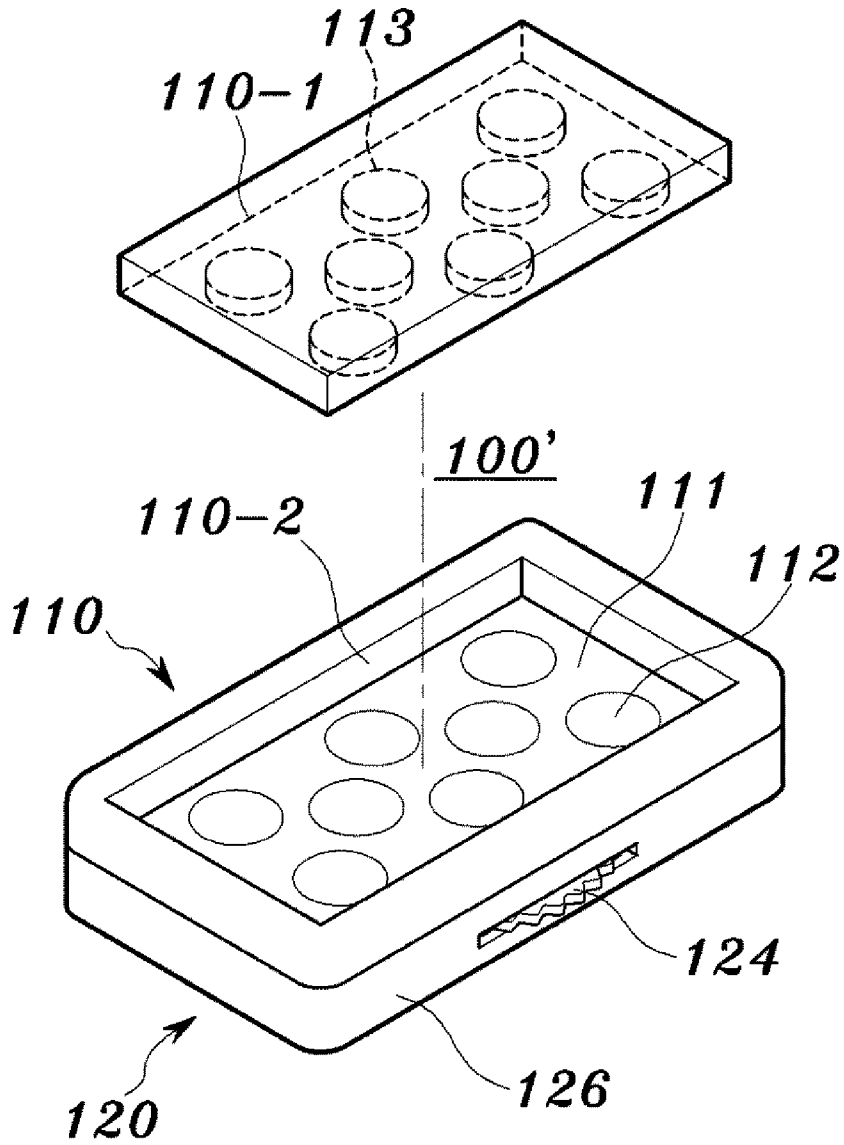
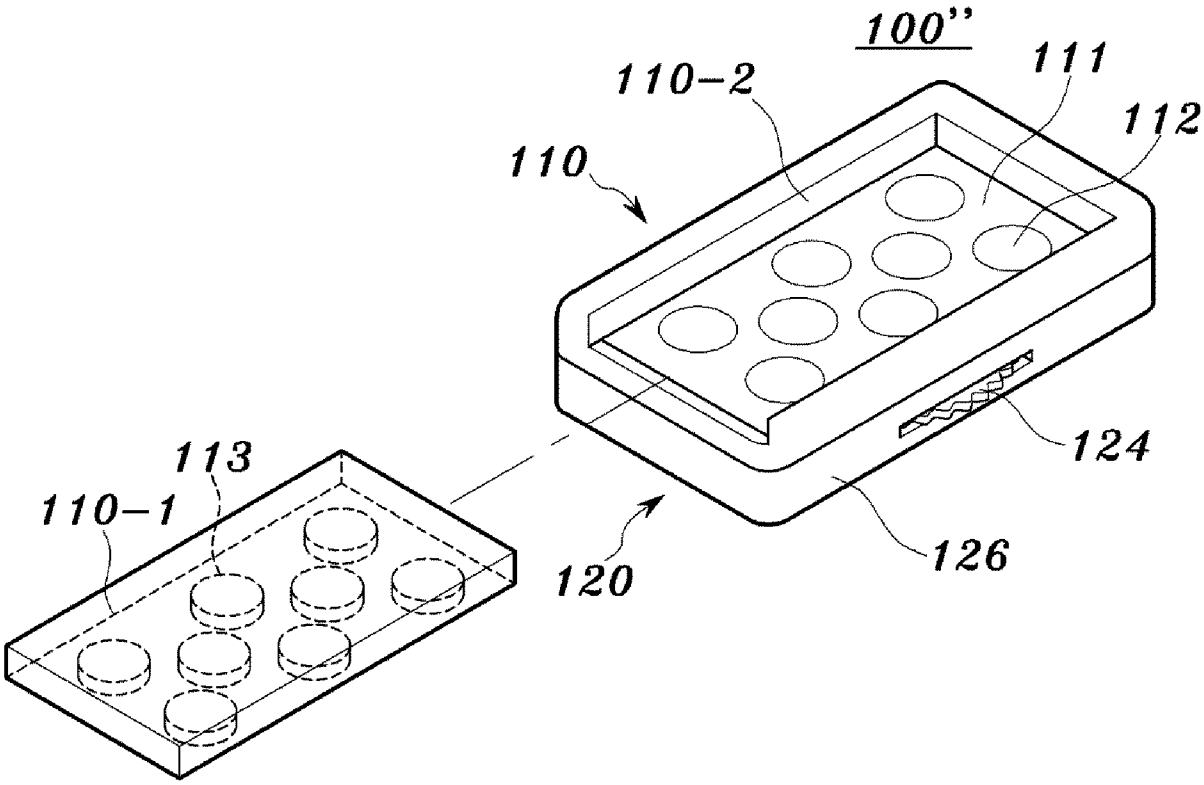
121
122
111
112
43
42
41
Specific cell mixture layer
121
122

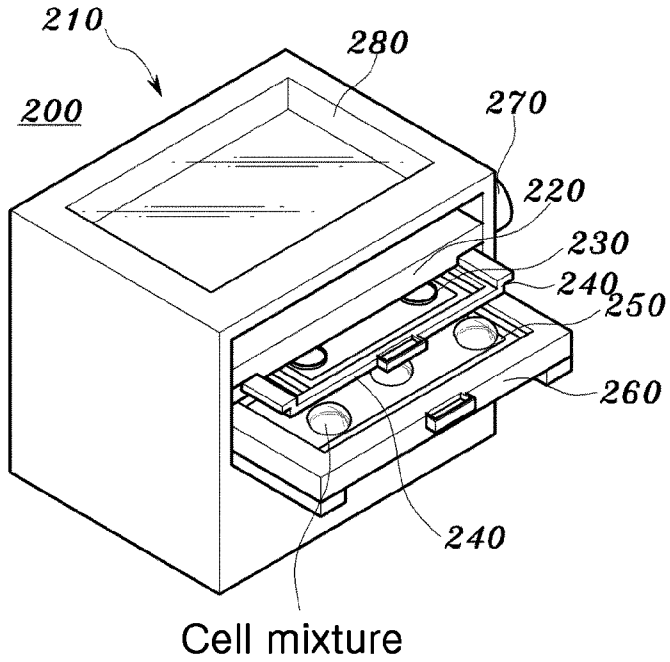
[Fig. 16]



[Fig. 17]



[Fig. 18]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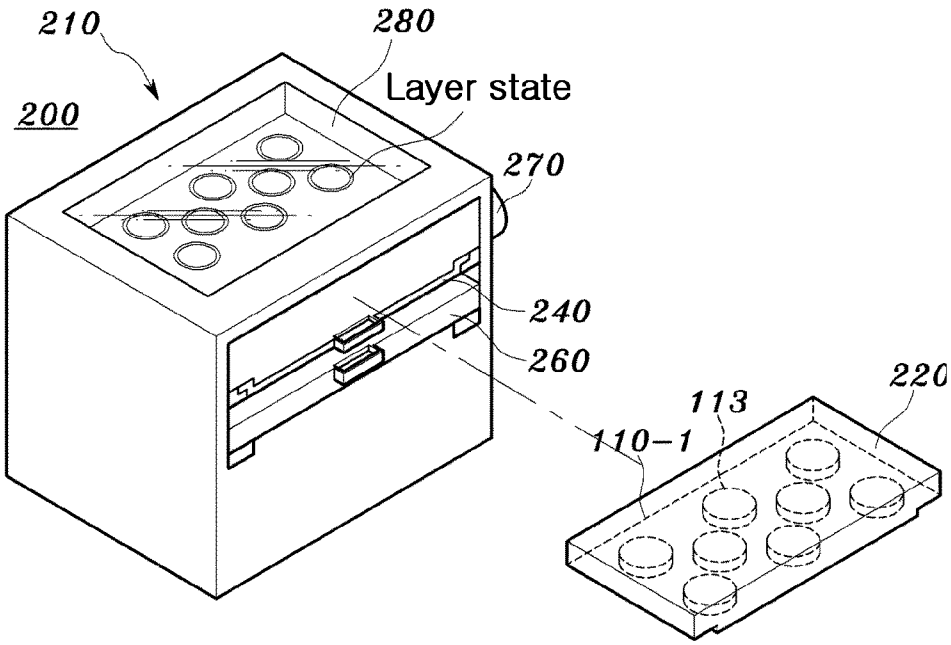
220
230
240
250
260
240
Cell mixture


210
280
Layer state
200
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110-1

INTERNATIONAL SEARCH REPORT

International application No.
PCT/KR2007/005042

A. CLASSIFICATION OF SUBJECT MATTER

C12M 1/42(2006.01)i

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 8 : C12M 1/42, C12M 1/34, G01N 33/553, C12N 5/00, C12N 13/00,

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

Esp@cenet, e-KIPASS "magnetic, separation, isolation, cell, biological, particle or bead"

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	WO 2006091060A1 (KIM, Y. et al.) 31 AUG. 2006 -see Abstract, Figs.1-29, & Claims 1-15.	1-4
A	US 5,602,042A (CYTYC CORP.) 11 FEB. 1997 -see Abstract, Fig.1-3, & Claims 1-31.	1-4
A	US 6,482,328B1 (DAVIDSON et al.) 19 NOV. 2002 -see Claims 1-22.	1-4
A	US 5,536,475A (BAXTER INTERNATIONAL INC.) 16 JUL. 1996 -see Abstract.	1-4
A	US 5,968,820A (CLEVELAND CLINIC FOUNDATION) 19 OCT. 1999 -See Abstract.	1-4
A	US 20050227349A1 (Korea Institute of Science Technology) 13 OCT. 2005 -see Abstract, Figs.1, 2, 4, & Claims 1-20.	1-4

☐ Further documents are listed in the continuation of Box C.

☒ See patent family annex.

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"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
11 JANUARY 2008 (11.01.2008)	**11 JANUARY 2008 (11.01.2008)**

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JEONG, JAE CHEOL

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INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No.

PCT/KR2007/005042

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