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[Continued on next page]

(54) Title: SAMPLE COLLECTION AND TRANSFER ASSEMBLY AND RELATED METHODS

(57) Abstract: A collection device includes a base housing member having at least one chamber. A cover housing member has at least one aperture therein, and the cover housing member is configured to cover the base housing member such that the at least one aperture is positioned in fluid communication with the at least one chamber. The cover housing member includes a cartridge holding interface configured to releasably engage with a cartridge that is configured to cover and receive a fluid from the at least one chamber via the at least one aperture.

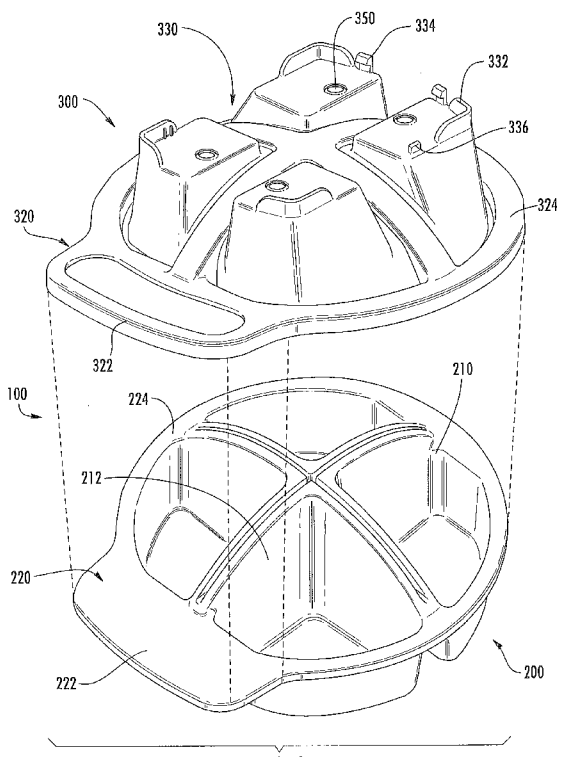

FIG. 3

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SAMPLE COLLECTION AND TRANSFER ASSEMBLY AND RELATED METHODS

RELATED APPLICATIONS

[0001] This application claims the benefit of United States Provisional Applications No. 61/662,158, filed June 20, 2012 (Docket No. 9903-9PR), No. 61/696,517, filed Sept. 4, 2012 (Docket No. 9903-13PR), and No. 61/757,466, filed January 28, 2013 (Docket No. 9903-9PR2), the disclosures of which are incorporated by reference herein in their entirety.

FIELD OF THE INVENTION

[0002] The present invention relates to sample collection assembly including a sample collection and transfer device and a sample cartridge.

BACKGROUND

[0003] Mastitis is the inflammation of the mammary gland caused by microorganisms that invade one or more quadrants of the bovine udder, multiply, and produce toxins that are harmful to the mammary gland. Economic loss to mastitis in the United States is estimated to be over $ 2 billion. This is approximately 10% of the total value of farm milk sales, and about two-thirds of this loss is due to reduced milk production in subclinically infected cows.

[0004] In subclinical mastitis, there may be no visible signs of the disease, and diagnosis of subclinical mastitis may be performed by a somatic cell count (SCC) of the milk. The SCC is the number of leukocytes or white blood cells per volume of milk and is also used as an index of milk quality. It has also been recognized that there are multiple types of leukocytes, each with its own significance. In milk from a healthy animal, the predominant cell types are lymphocytes, followed by lesser numbers of neutrophils and macrophages. The percentages of each kind of cell rise and fall as part of the immune response to infection. Those percentages, "the milk leukocyte differential", cell count represent the unique immune status of an individual quarter udder, at a specific point in time for better diagnosis of subclinical mastitis.

[0005] One method for detecting the milk leukocyte differential is using flow-

cytometry, which is an expensive, sophisticated tool typically only found in top research laboratories and generally not practical for the farmer. Another method for detecting the milk leukocyte differential is the "manual milk differential smear" (MMDS), which is a difficult and time consuming procedure, and is subject to great variability, even when performed by highly trained laboratory technologists. Both flow-cytometry and MMDS present practical difficulties for field research or a barn environment.

[0006] U.S. Patent Application Publication No. 2009/0233329 to Rodriguez discloses a wedge microfluidic slide chamber for detecting mastitis or other diseases from a body fluid of a mammal, such as from cow's milk. The wedge-shaped chamber uses capillary action to fill the chamber with the sample as a "self-preparing wet smear" with a meta-chromatic stain. The wedge-shaped microscope slide with the stained sample may be analyzed by visual identification and direct observation or by imaging instruments using computer-enhanced digital camera images. Accordingly, mastitis may be detected more easily with such a self-preparing wet smear.

[0007] Milk collection techniques for such a slide, however, may be time consuming and/or difficult. Typically, a sample from each quadrant of the cow's udder may be collected in different containers and pipetted by an operator into the wedge-shaped slide chamber for further analysis, for example, by an imaging instrument or reader. Moreover, it may be desirable to pipette the sample into the self-preparing wet smear relatively soon before placing the microscope slide into the imaging instrument or reader. The liquid samples may be stored in separate containers prior to analysis, and the tracking and/or storage of such samples may present various challenges, for example, to track which sample came from which cow and from which quadrant of the cow.

SUMMARY OF EMBODIMENTS OF THE INVENTION

[0008] In some embodiments, a collection device includes a base housing member having at least one chamber. A cover housing member has at least one aperture therein, and the cover housing member is configured to cover the base housing member such that the at least one aperture is positioned in fluid communication with the at least one chamber. The cover housing member includes a cartridge holding interface configured to releasably engage with a cartridge that is configured to cover and receive a fluid from the at least one chamber via the at least one aperture.

[0009] In some embodiments, the cartridge holding interface comprises a retaining

wall or pins configured to abut an outer perimeter of the cartridge. The cartridge holding interface may include at least one notch and/or groove that is configured to engage a corresponding notch and/or groove on the cartridge. The cartridge holding interface may include at least one hook member that is configured to engage and retain an edge portion of the cartridge.

[0010] In some embodiments, the cartridge holding interface is configured to interface with the cartridge in a single orientation.

[0011] In some embodiments, at least one chamber includes a plurality of chambers and the at least one aperture includes a plurality of apertures. The base housing member and the cover housing member include cooperating sealing members that are configured to seal each of the plurality of chambers. In some embodiments, the cooperating sealing members include a base sealing feature between the plurality of wells on the base housing member, and a cover sealing feature on the cover housing member configured to engage with the base sealing feature and to thereby fluidly seal each of the plurality of wells. One of the base sealing feature and the cover sealing feature may include a groove and the other of the base sealing feature and the cover sealing feature comprises a ridge that is configured to be received in the groove and form a snug fit. In some embodiments, the chambers overlap a central portion and a perimeter portion of the base housing member, and the chambers comprise a wall that has a height that is higher in the central portion than at the perimeter portion.

[0012] In some embodiments, a collection and transfer assembly includes a collection device. The collection device includes a base housing member having at least one chamber, and a cover housing member having at least one housing aperture therein. The cover housing member is configured to cover the base housing member such that the at least one housing aperture is positioned in fluid communication with the at least one chamber. The cover housing member further includes a cartridge holding interface. The collection and transfer assembly further includes a sample cartridge comprising at least one sample area having at least one cartridge aperture that is configured to receive a fluid from the at least one chamber via the at least one housing aperture. The cartridge holding interface on the cover housing member is configured to releasably engage with the sample cartridge.

[0013] In some embodiments, the sample cartridge has a first major surface opposite a second major surface, the at least one sample area is on the first major surface and faces away from the cover housing member, and the at least one cartridge aperture is on the second

major surface and fluidly connects the housing aperture to the sample area. In some embodiments, the sample cartridge has a first major surface opposite a second major surface, and the at least one sample area is on the second major surface and faces toward the cover housing member, and the at least one cartridge aperture is on the second major surface and fluidly connects the housing aperture to the sample area.

[0014] In some embodiments, the cartridge holding interface comprises a retaining wall or protruding pins or posts configured to abut an outer perimeter of the cartridge. The cartridge holding interface may include at least one notch and/or groove that is configured to engage a corresponding notch and/or groove on the cartridge. The cartridge holding interface may include at least one hook member that is configured to engage and retain an edge portion of the cartridge.

[0015] In some embodiments, the cartridge holding interface is configured to interface with the cartridge in a single orientation.

[0016] In some embodiments, the at least one chamber includes a plurality of chambers and the at least one aperture includes a plurality of apertures. The base housing member and the cover housing member may include cooperating sealing members that are configured to seal each of the plurality of chambers. In some embodiments, the cooperating sealing members include a base sealing feature between the plurality of wells on the base housing member, and a cover sealing feature on the cover housing member configured to engage with the base sealing feature and to thereby fluidly seal each of the plurality of wells. One of the base sealing feature and the cover sealing feature may include a groove and the other of the base sealing feature and the cover sealing feature may include a ridge that is configured to be received in the groove and form a snug fit. In some embodiments, the chambers overlap a central portion and a perimeter portion of the base housing member, and the chambers include a wall that has a height that is higher in the central portion than at the perimeter portion.

[0017] In some embodiments, a method of collecting a sample includes collecting a sample in at least one chamber of a base housing member of a collection device. A cover housing member is placed on the base housing member of the collection device. The cover housing member has at least one housing aperture therein. The cover housing member is configured to cover the base housing member such that the at least one housing aperture is positioned in fluid communication with the at least one chamber. The cover housing member further includes a cartridge holding interface. A sample cartridge is positioned on the

cartridge holding interface. The sample cartridge further includes at least one sample area having at least one cartridge aperture that is configured to receive a fluid from the at least one chamber via the at least one housing aperture. The cover and base housing members are inverted such that the sample flows from the at least one chamber to the at least one sample area via the at least one housing aperture and the at least one cartridge aperture.

[0018] In some embodiments, the method further includes placing the cartridge in an imaging reader.

[0019] In some embodiments, the method further includes storing the collection device and the sample cartridge before the inverting step.

BRIEF DESCRIPTION OF THE DRAWINGS

[0020] The accompanying drawings, which are incorporated in and constitute a part of the specification, illustrate embodiments of the invention and, together with the description, serve to explain principles of the invention.

[0021] **Figure 1** is a front perspective view of a collection and transfer device with a sample cartridge according to some embodiments.

[0022] **Figure 2** is a back perspective view of the collection and transfer device of **Figure 1**.

[0023] **Figure 3** is a front perspective view of a base housing and a cover housing of the collection and transfer device of **Figure 1** in an open configuration.

[0024] **Figure 4** is an interior perspective view of the cover housing of the collection and transfer device of **Figure 1**.

[0025] **Figure 5** is a cross-sectional perspective view of the cover housing of the collection and transfer device of **Figure 1**.

[0026] **Figure 6** is a top view of the base housing of the collection and transfer device of **Figure 1**.

[0027] **Figure 7** is a top view of the cover housing of the collection and transfer of **Figure 1**.

[0028] **Figure 8** is a bottom perspective view of the cartridge device of **Figure 1**.

[0029] **Figure 9** is a cross-sectional top perspective view of the cartridge device of **Figure 1**.

[0030] **Figure 10** is a cross-sectional perspective view of the collection and transfer device and the cartridge of **Figure 1**.

[0031] **Figure 11** is an exploded cross-sectional perspective view of the collection and transfer device interface with the cartridge device of **Figure 1**.

[0032] **Figure 12** is a perspective view of a collection and transfer device and a sample cartridge device according to some embodiments.

[0033] **Figure 13** is a cross-sectional perspective view of the collection and transfer device of **Figure 12**.

[0034] **Figure 14** is an exploded cross-cross sectional perspective view of the collection and transfer device interface with the cartridge device of **Figure 12**.

[0035] **Figure 15** is a bottom perspective view of the collection and transfer device base housing of **Figure 1**.

[0036] **Figure 16** is a cross-sectional perspective side view of two collection and transfer devices with corresponding sample cartridges in a nesting configuration according to some embodiments.

[0037] **Figure 17** is a perspective side view of a plurality of the base housings of collection and transfer devices in a nesting configuration according to some embodiments.

[0038] **Figure 18** is a flowchart illustrating operations according to some embodiments.

[0039] **Figure 19** is a front perspective view of a collection and transfer device with a sample cartridge according to some embodiments.

[0040] **Figure 20** is a front perspective view of a collection and transfer device with the sample cartridge removed according to some embodiments.

[0041] **Figure 21** is a front perspective view of the base housing of the collection and transfer device of **Figure 20**.

[0042] **Figure 22** is a bottom perspective view of the cover housing of the collection and transfer device of **Figure 20**.

[0043] **Figure 23** is an exploded perspective view of a sample cartridge according to some embodiments.

[0044] **Figure 24** is an assembled view of the sample cartridge of **Figure 23**.

[0045] **Figure 25** is a front perspective view of the base housing of the collection and transfer device with a cover housing or plug according to some embodiments.

DETAILED DESCRIPTION OF EMBODIMENTS OF THE INVENTION

[0046] The present invention now will be described hereinafter with reference to the accompanying drawings and examples, in which embodiments of the invention are shown. This invention may, however, be embodied in many different forms and should not be construed as limited to the embodiments set forth herein. Rather, these embodiments are provided so that this disclosure will be thorough and complete, and will fully convey the scope of the invention to those skilled in the art.

[0047] Like numbers refer to like elements throughout. In the figures, the thickness of certain lines, layers, components, elements or features may be exaggerated for clarity.

[0048] The terminology used herein is for the purpose of describing particular embodiments only and is not intended to be limiting of the invention. As used herein, the singular forms "a," "an" and "the" are intended to include the plural forms as well, unless the context clearly indicates otherwise. It will be further understood that the terms "comprises" and/or "comprising," when used in this specification, specify the presence of stated features, steps, operations, elements, and/or components, but do not preclude the presence or addition of one or more other features, steps, operations, elements, components, and/or groups thereof. As used herein, the term "and/or" includes any and all combinations of one or more of the associated listed items. As used herein, phrases such as "between X and Y" and "between about X and Y" should be interpreted to include X and Y. As used herein, phrases such as "between about X and Y" mean "between about X and about Y." As used herein, phrases such as "from about X to Y" mean "from about X to about Y."

[0049] Unless otherwise defined, all terms (including technical and scientific terms) used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs. It will be further understood that terms, such as those defined in commonly used dictionaries, should be interpreted as having a meaning that is consistent with their meaning in the context of the specification and relevant art and should not be interpreted in an idealized or overly formal sense unless expressly so defined herein. Well-known functions or constructions may not be described in detail for brevity and/or clarity.

[0050] It will be understood that when an element is referred to as being "on," "attached" to, "connected" to, "coupled" with, "contacting," etc., another element, it can be directly on, attached to, connected to, coupled with or contacting the other element or intervening elements may also be present. In contrast, when an element is referred to as

being, for example, "directly on," "directly attached" to, "directly connected" to, "directly coupled" with or "directly contacting" another element, there are no intervening elements present. It will also be appreciated by those of skill in the art that references to a structure or feature that is disposed "adjacent" another feature may have portions that overlap or underlie the adjacent feature.

[0051] Spatially relative terms, such as "under," "below," "lower," "over," "upper" and the like, may be used herein for ease of description to describe one element or feature's relationship to another element(s) or feature(s) as illustrated in the figures. It will be understood that the spatially relative terms are intended to encompass different orientations of the device in use or operation in addition to the orientation depicted in the figures. For example, if the device in the figures is inverted, elements described as "under" or "beneath" other elements or features would then be oriented "over" the other elements or features. Thus, the exemplary term "under" can encompass both an orientation of "over" and "under." The device may be otherwise oriented (rotated 90 degrees or at other orientations) and the spatially relative descriptors used herein interpreted accordingly. Similarly, the terms "upwardly," "downwardly," "vertical," "horizontal" and the like are used herein for the purpose of explanation only unless specifically indicated otherwise.

[0052] It will be understood that, although the terms "first," "second," etc. may be used herein to describe various elements, these elements should not be limited by these terms. These terms are only used to distinguish one element from another. Thus, a "first" element discussed below could also be termed a "second" element without departing from the teachings of the present invention. The sequence of operations (or steps) is not limited to the order presented in the claims or figures unless specifically indicated otherwise.

[0053] Embodiments according to the present invention will now be described with respect to **Figures 1-24**. As illustrated in **Figures 1-2**, a collection and transfer assembly **10** includes a collection device **100** and a sample cartridge **400**. The collection device **100** includes a base housing **200** and cover housing **300**. The base housing **200** includes four chambers **210** that are configured to collect a sample. The sample cartridge **400** includes four sample areas **430**, which are configured to receive the sample from the chambers **210** for further analysis. In some embodiments, the collection and transfer assembly **10** may be used to collect a liquid biological fluid sample, for example, a milk sample from a milk producing animal, such as a cow or goat. Accordingly, the milk sample may be conveniently collected and transferred to the microscope slide for further analysis, for example, in a microscope or

imaging reader. The sample areas **430** may include a wedge-shaped microscope slide such that capillary action fills the slide **430** with the sample as a "self-preparing wet smear" with a meta-chromatic stain. The stain may be preloaded onto the sample areas **430**. The wedge-shaped microscope slide with the stained sample may be analyzed by visual identification and direct observation or by imaging instruments using computer-enhanced digital camera images. Examples of suitable imaging readers and wedge-shaped slides are described in U.S. Patent Application Publication No. 2009/0233329 to Rodriguez. The cartridge for imaging a specimen on an automated microscope may include a substrate, a chamber or generally planar imaging surface on or in the substrate for containing or supporting the specimen; a plurality of exogeneous targets in the chamber or on the surface; and (optionally but in some embodiments preferably) at least one optically transparent wall formed on or forming the chamber to facilitate imaging the contents thereof.

[0054] As illustrated in **Figures 3-7**, the base housing **200** defines four chambers **210** having centrally elevated splash-guard walls **212**, a groove **214**, and an outer lip **220** that includes a handle portion **222** and a perimeter portion **224**. The cover housing **300** includes a plurality of apertures **316**, an outer lip **320** includes a handle portion **322** and a perimeter portion **324**. The cover housing **300** further defines a cartridge holding interface **330** that is sized and configured to releasably engage with and/or abut the cartridge **400** (**Figures 1-2**). As shown, for example, in **Figure 3**, the cartridge holding interface **330** includes various retaining features, including retaining walls **332**, retaining members or hooks **334**, and interlocking member or notch **336**, and retaining pins **338**.

[0055] As illustrated, for example, in **Figure 4**, the cover housing **300** includes chamber covering sealing **310** that correspond to the chambers **210** of the base housing **200**. The chamber sealing features **310** are configured to seal and/or fluidly isolate different samples that are collected in each of the four chambers **210** of the base housing **200**. As illustrated, the chamber sealing features **310** include a sealing rib **314** on a ridge interface **315**, and sealing protrusions **318**. The sealing rib **314** is configured to mate with the groove **214** of the base housing **200**, and the sealing protrusions **318** are configured to extend into the chambers **210** of the base housing **200** such that a fluid sample in each of the chambers **210** is generally prevented from leaking into other ones of the chambers **210**.

[0056] In this configuration, and as illustrated in **Figure 10**, the base housing **200** and the cover housing **300** interlock with on another to generally seal each of the sample chambers **210** to reduce or prevent leaking between chambers **210**. Therefore, the samples

from the quadrants of the cow may be separately stored and/or tested separately. In particular, the lips **220**, **320** may engage with one another (such as in a snap- or press-fit) to form an outer seal. The sealing protrusions **318** of the cover housing **300** extend into the chambers **210** to further seal the sample chambers **210**. The sealing rib **314** and ridge interface **315** of the cover housing **300** mate with the central groove **214** of the base housing **200** to form a sufficiently snug fit to generally seal the four chambers **210** and/or reduce or prevent sample fluid from one chamber **210** leaking to another chamber **210**.

[0057] As shown in **Figures 8-9**, the sample cartridge **400** includes an outer perimeter **420**, sample areas **430**, and apertures **450**. The outer perimeter **420** has an asymmetric shape and orientation features, such as notches **422**, for interacting with the cartridge holding interface **330** of the collection device cover housing **300**. The sample areas **430** include a transparent or translucent slide **432**, such as a microscope slide, that is configured to hold and retain the fluid sample for analysis by a slide reader (not shown). The apertures **450** are sized and configured to provide a fluid connection with the apertures **350** of the cover housing **300**.

[0058] As illustrated in **Figures 10-11**, a fluid sample collected in the chambers **210** may be transferred to the sample areas **430** of the sample cartridge **400** by inverting the collection and transfer chamber **10**. In an inverted position, the fluid sample flows from the base chamber **210** to the cover housing **300** and into the sample area **430** via the cover housing apertures **350** and the sample cartridge apertures **450**.

[0059] Accordingly, corresponding interlocking features, such as notches and/or grooves **336** and **422** may be included in the outer perimeter **420** of the cartridge **400**. In addition, the cartridge holding interface **330** and the outer perimeter **420** of the cartridge **400** may be asymmetric such that the cartridge **400** fits into the cartridge holding interface **330** in only a single orientation. Moreover, the hooks **334** may further hold the cartridge **400** in position such that the assembly **10** may be inverted or transported without the cartridge **400** becoming dislodged from the cartridge holding interface **330**.

[0060] Embodiments according to the invention are described above with respect to a configuration in which the fluid enters the cartridge **400** via the bottom (or the major side opposite the sample areas **430**) such that a user may view the sample in the sample areas **430** when the cartridge **400** is in position on the cover housing **300**. It should be understood, however, that other configurations may be used to provide a fluid pathway from the sample chambers **210** to a sample region, such as on a microscope slide. For example, as illustrated **Figures 12-14**, a sample cartridge **500** includes an outer perimeter **520**, sample areas **530**,

and apertures **550**. The sample areas **530** include a transparent or translucent slide **532** and sample entry passages **534**. The slide **532** may be a microscope slide that is configured to hold and retain the fluid sample for analysis by a slide reader (not shown). The entry passages **534** are positioned so as to provide a fluid passageway from the apertures **350** of the cover housing **300**. Accordingly, when the device **100** is inverted, the sample flows from the chambers **210** to the sample areas **530** via the apertures **350** and the passages **534**. As illustrated, the slide **532** is on a side of the cartridge **500** that faces the collection device cover housing **300**.

[0061] In a similar manner as described above with respect to the cartridge **400**, the outer perimeter **520** of the cartridge **500** has an asymmetric shape and orientation features, such as notches **522**, for interacting with the cartridge holding interface **330** of the collection device cover housing **300**. Therefore, the cartridges **400, 500** may be configured to mate with the cartridge holding interface **330** in a single orientation. In this configuration, the sample may be collected in the chambers **210** corresponding to predefined quadrant of the cow or other sample source. The cartridges **400, 500** may be configured to fit on the holding interface **330** in a single orientation so that the source of the sample from one of the predefined quadrants is known. The cartridges **400, 500** may also fit into a reader (not shown) in a single orientation so that the reader associates the sample results with a predefined quadrant of the cow or other sample source. The sample results may be recorded by a user or by a computer processor associate with the reader. The sample results may be recorded together with the sample source, such as a cow identification number and a quadrant number based on the particular chamber **210** and corresponding sample area **430, 530**.

[0062] As shown in **Figure 15**, the base housing **200** may include nesting features **260** that generally correspond to the cartridge holding interface **330** or other top features of the cover housing **300**. As illustrated in **Figure 16**, the nesting features **260** permit the stacking of two or more collection and transfer assemblies **10** by resting the nesting features **260** against the cartridge holding interface **330** of an adjacent assembly **10**. As shown in **Figure 16**, the cartridge holding interface **330** is shaped such that it fits inside the nesting features **260** of an adjacent assembly **10**. In some embodiments, the collection devices **100** may be filled with fluid samples, and then stored in the stacked configuration. The fluid samples may be collected in the sample chambers **210** without being added to the cartridges **400** during storage. Accordingly, contact between the fluid sample and the assay and/or stain

in the sample area **430** may be avoided until just prior to analyzing the sample in the cartridge **400** when the collection and transfer assembly **10** may be agitated and inverted to thereby cause fluid to flow from the chamber **210** to the sample area **430** via the cover apertures **350** and the cartridge apertures **450**. Therefore, the user may collect samples from multiple bovine animals prior to analysis and store the samples without contacting the sample with the assay and/or stain in the sample area **430** until the user chooses to analyze the cartridges **400**.

[0063] The chambers **210** of the base housing **200** may be tapered to permit nesting of the sample collection base housing **200** for ease of transport and/or shipping as illustrated in **Figure 17**.

[0064] Operations according to some embodiments are shown in **Figure 18**. A fluid sample, for example, milk from a cow, goat or other milk producing animal, may be collected in the chambers of the base housing (Block **1000**; **Figure 18**). As shown in **Figures 1-7** and **10**, the chambers **210** of the base housing **200** include splash-guard walls **212** that are sufficiently high that splatter may be reduced between the chambers **210**. Fluid milk samples may be collected directly from each of the four quadrants or teats of the cow generally without mixing samples collected from other quadrants of the milk-producing animal due, in part, to the high splash-guard walls **212**. The cover housing may then be placed on the base housing (Block **1002**; **Figure 18**). In some embodiments, however, the cover housing may be a temporary plug housing, such as the housing **1500** of **Figure 25** that is placed on the base housing **1200** for storage, transportation and/or agitation of the sample. The device assembly may be stored (Block **1006**). In some embodiments, the assembly may be stored prior to analysis and/or filing the cartridge sample areas. However, in some embodiments, the sample areas of the cartridge may be filed with the fluid sample prior to storage. Before the sample cartridge is filled, the plug (if used) is replaced with the transfer cover and the sample cartridge is positioned on the cartridge holding interface of the cover housing.

[0065] When the user wishes to initiate sample analysis, the sample may be agitated, for example, in the base housing, because fluids such as milk may separate into high- and low-fat components (Block **1008**). If the cover housing **1500** of **Figure 25** was used, then the cover housing **1500** is first replaced with the cover housing **1200** and cartridge **1400** (Block **1007**). The cover and the base housing may be inverted such that the sample flows into the cartridge sample areas via the cover housing apertures and the cartridge apertures (Block **1010**). The sample cartridge may then be removed from the cover housing and placed in a reader (Block **1012**) for further analysis.

[0066] Although embodiments according to the present invention are described herein with respect to four sample areas **430** and corresponding cartridge filling apertures **450,** cover housing filling apertures **350**, and collection chambers **210**, it should be understood that any number of sample areas **430** may be provided. In some embodiments, a single sample area may be used with corresponding filling apertures and a single collection chamber.

[0067] As illustrated in **Figure 19**, a collection and transfer assembly **1010** includes a collection device **1100** and a sample cartridge **1400**. The collection device **1100** includes a base housing **1200** and a cover housing **1300**. The base housing **1200** includes four generally cylindrical chambers **1210** and on a base or main portion **1220**. The chambers **1210** include a wall portion that extends away from the main portion **1220** and into corresponding, cooperating cover sealing features **1310** on the cover housing **1300**. The sealing features **1310** cover the chambers **1210** and may form a tight fit to substantially isolate and reduce or prevent leakage from the chambers **1210**. The cover housing sealing features **1310** further include apertures **1350**. The sample cartridge **1400** includes sample areas **1430**, which are defined by sample slides **1432**. The sample areas **1430** include apertures **1452** that cooperate with the apertures **1350** of the cover housing **1300**. In this configuration, a milk sample may be collected in the chambers **1210** of the base housing **1200** and sealed by the cover housing **1300**. The milk sample may be received in the sample areas **1430** of the sample cartridge **1400** when the assembly **1010** is inverted, and the milk sample flows from the chambers **1210** via the apertures **1350** and **1452** and into the sample area **1430**.

[0068] As illustrated in **Figure 20**, the cover housing **1300** includes a holding interface **1330** having various retaining features **1332, 1334** and **1336** for retaining the sample cartridge **1400**. In some embodiments, the retaining features **1332, 1334** and **1336** may be assymetric such that the cartridge **1400** fits into the interface **1330** in a single orientation. As shown in **Figure 21**, the chambers **1210** of the base housing **1200** are illustrated in additional detail. As illustrated, the chambers **1210** include asymmetric or angled walls that may reduce splashing or contamination from one chamber to another because the wall height of the chambers **1210** is greater toward the central region of the base housing **1200**.

[0069] In some embodiments, the base housing **1200** and the cover housing **1300** may include various features that interact for stability of assembly and/or so that the base housing **1200** and the cover housing **1300** fit together in a single orientation. For example, the base housing **1200** further includes protrusions **1212** and **1214** that interact with corresponding features of the cover housing **1300** as shown in **Figure 22**. As illustrated in **Figure 22**, the

cover portion includes a notch **1312**, an aperture **1314** and a stabilization arm **1316**. In an assembled configuration, the protrusion **1212** of the base housing **1200** is received in the notch **1312**, and the protrusion **1214** is received in the aperture **1314**. When assembled, the stabilization arm **1316** rests on the main portion **1220** of the base housing **1200** for stability.

[0070] As shown in **Figures 23-24**, the cartridge **1400** includes a glass plate **1410** having microscope cover slips **1432** affixed thereto, and a frame member **1420** that defines sample collection apertures **1450**. The slides **1432** may be configured in a wedge-shape such that capillary action files the slides **1432** with the sample as a "self-preparing we smear" with a meta-chromatic stain, which may be preloaded into the sample areas **1430**.

[0071] As illustrated, the apertures **1350** protrude from the cover housing member **1300** such that the apertures **1350** form a fluid connection with a corresponding feature in the cartridge **1400**, such as the sample collection apertures **1450** to thereby fill the sample area **1430**. For example, the apertures **1350** may protrude from the cover housing **1300** and fit into a corresponding well or other collection feature (such as the apertures **1450**) on the cartridge **1400** to reduce or prevent leaking between the chambers **1210**, the sealing features **1310** and the sample areas **1430**.

[0072] In some embodiments, the cover housing used for transfer may be substituted with a plug assembly that caps the fluid in the base housing for storage and/or transportation. In some embodiments the plug may be configured to allow stackability. In some embodiments, the fluid inside the base housing may be agitated before removing the plug assembly and replacing it with the cover housing used for transfer of the fluid to the cartridge. For example, as illustrated in **Figure 25**, a cover housing or plug **1500** is optionally positioned on top of the base housing **1200** for storage and/or transport. The plug **1500** includes a directional arrow **1510** for indicating to the user the correct orientation of the plug **1500**. Accordingly, the sample may be stored in the base housing **1200** and protected by the plug **1500**, and the plug **1500** may be removed and the cover housing **1300** positioned thereon as described herein to transfer the sample to the cartridge **1400**.

[0073] In some embodiments, various tracking techniques may be used to identify a particular sample with an animal. For example, the collection and transfer devices and/or the sample cartridges described herein may include a write-on label and/or a bar code label and/or an RFID tag for purposes of identifying the origin of the sample, such as a cow identification number.

[0074] In some embodiments, the sample cartridges described herein may be placed in a reader or imager for further analysis. When the sample comprises cells to be imaged and/or counted by the reader, the cells may be stained by a suitable stain, including fluorescent stains such as acridine orange (*see, e.g.,* US Patent No. 3,883,247). In some embodiments, the cartridges described herein may use exogeneous targets as discussed below.

[0075] **Microscopes**

[0076] The present invention can be carried out with any suitable manual or automated microscope. Automated microscopes generally include a specimen support stage (e.g., configured for holding or securing a sample cartridge as described above), an objective lens, a camera operatively associated with the objective lens, at least one drive assembly operatively associated with said support stage and/or said objective lens. Examples of such microscopes include but are not limited to those described in US Patents Nos. 4,810,869; 5,483,055; 5,647,025; 5,790,710; 6,869,570; 7,141,773; and 8,014,583. In general, such apparatus includes a controller that is operatively associated with the camera and the at least one drive assembly which controller is configured through hardware and/or software to carry out an autofocus method as described herein (generally prior to acquisition of an image of the specimen or sample through the camera), typically through calculating a focus score. The focus score can be calculated by any suitable technique, including but not limited to those described in F. Groen et al., *A comparison of different focus functions for use in autofocus algorithms,* Cytometry **6,** 81-91 (1985). Difference from the background, given a uniform background, can be calculated a number of ways, including but not limited to differences in contrast, gradient, and variance.

[0077] **Exogeneous targets.**

[0078] General considerations for selecting the exogeneous target are as follows: The exogenous target should be visible by the particular optical system in use. This will depend on the magnification, excitation wavelength, size of field of view, etc. This will influence decisions on which size, shape, emission wavlengths, etc. of the texture. In addition, the exogenous target should be distinguishable from the target objects. Preferably, the The exogeneous target reside at sustantially the same (or a known distance from) the focal plane of the target objects (e.g., be mixed with a biological sample suspected of containing cells to be imaged and/or counted, and/or placed in the same chamber as will contain a biological sample comprising cells to be imaged and/or counted). The exogeneous target should be of a

size, shape, and number so as to not substantially obscure the view of the intended target objects, such as cells to be imaged and/or counted. And,the exogenous target should provide sufficient contrast with an empty field of view so as to provide an adequate focal peak and allow for reliable, reasonably rapid, and/or robust focusing.

[0079] The exogenous targets may be formed of any suitable material, including organic polymers, inorganic materials (including crystalline materials, amorphous materials, metals, etc.) and composites thereof.

[0080] The exogenous targets may be contained loosely within the chamber, fixed to one wall of the chamber, or surface to be imaged (e.g., by adhesive, by electrostatic, hydrophilic, or hydrophobic interaction, covalent bond directly or through a linking group, etc.), and/or formed on one wall of the chamber (e.g., by molding, etching, painting, silk-screening, lithography, etc.).

[0081] The exogenous targets may be opaque or transparent. When transparent the targets may be "tinted" so as to transmit light therethrough at a predetermined wavelength (for example, so that they appear red, green, blue, yellow, etc., to a human observer).

[0082] The exogenous targets may be regular or irregular in shape (for example, cylinders, spheres, cubes, pyramids, prisms, cones, rods, etc.). In some embodiments, the targets have an average diameter of from 0.1, 0.5 or 1 micrometers up to 2, 5, or 10 micrometers.

[0083] The number of exogenous targets is not critical, but in some embodiments the speed of the autofocus process can be increased by increasing, at least to a point, the number of exogenous targets in the chamber so that the targets are readily located in the automated microscope. Where a plurality of targets are included in the sample chamber (e.g., 2, 4, 6, 8 or 10 targets, up to 100, 200, 400, 600 or 800 exogenous targets, or more), in some embodiments that plurality preferably consists of or consists essentially of targets having substantially the same size, shape, and optical characteristics.

[0084] In some embodiments, the targets are beads, such as fluorescent microbeads. Such microbeads are commonly available and used for calibrating flow cytometers or fluorescent microscopes (see, e.g., US Patents Nos. 4,698,262; 4,714,682; and 4,868,126).

[0085] The targets are preferably optically distinguishable from cells to be counted (and hence would not be useful as calibration standards for the particular cells to be counted and/or imaged by the methods described herein). Optically distinguishable may be achieved by any suitable technique, such as by utilizing targets of a different and distinguishable shape

from the cells to be counted, by utilizing targets that emit, transmit, and/or reflect light at a different wavelength from the cells to be counted when under the same illumination conditions, and combinations thereof.

[0086] The foregoing is illustrative of the present invention and is not to be construed as limiting thereof. Although a few exemplary embodiments of this invention have been described, those skilled in the art will readily appreciate that many modifications are possible in the exemplary embodiments without materially departing from the novel teachings and advantages of this invention. Accordingly, all such modifications are intended to be included within the scope of this invention as defined in the claims. Therefore, it is to be understood that the foregoing is illustrative of the present invention and is not to be construed as limited to the specific embodiments disclosed, and that modifications to the disclosed embodiments, as well as other embodiments, are intended to be included within the scope of the appended claims. The invention is defined by the following claims, with equivalents of the claims to be included therein.

THAT WHICH IS CLAIMED IS:

1. A collection device comprising:

a base housing member having at least one chamber; and

a cover housing member having at least one aperture therein, the cover housing member being configured to cover the base housing member such that the at least one aperture is positioned in fluid communication with the at least one chamber, the cover housing member further comprising a cartridge holding interface configured to releasably engage with a cartridge that is configured to cover and receive a fluid from the at least one chamber via the at least one aperture.

2. The collection device of Claim 1, wherein the cartridge holding interface comprises a retaining wall, pins and/or retaining features configured to abut an outer perimeter of the cartridge.

3. The collection device of Claim 2, wherein the cartridge holding interface further comprises at least one notch and/or groove that is configured to engage a corresponding notch and/or groove on the cartridge.

4. The collection device of Claim 2, wherein the cartridge holding interface further comprises at least one hook member that is configured to engage and retain an edge portion of the cartridge.

5. The collection device of Claim 1, wherein the cartridge holding interface is configured to interface with the cartridge in a single orientation.

6. The collection device of Claim 1, wherein the at least one chamber comprises a plurality of chambers and the at least one aperture comprises a plurality of apertures, wherein the base housing member and the cover housing member comprises cooperating sealing members that are configured to seal each of the plurality of chambers.

7. The collection device of Claim 6, wherein the cooperating sealing members comprise a base sealing feature between the plurality of wells on the base housing member, and a cover sealing feature on the cover housing member configured to engage with the base sealing feature and to thereby fluidly seal each of the plurality of wells.

8. The collection device of Claim 7, wherein one of the base sealing feature and the cover sealing feature comprises a groove and the other of the base sealing feature and the cover sealing feature comprises a ridge that is configured to be received in the groove and form a snug fit.

9. The collection device of Claim 6, wherein the chambers overlap a central portion and a perimeter portion of the base housing member, and the chambers comprise a wall that has a height that is higher in the central portion than at the perimeter portion.

10. The collection device of Claim 1, wherein the at least one chamber comprises a plurality of chambers and the at least one aperture comprises a plurality of corresponding apertures, wherein each of the plurality of chambers comprises wall portion that extends away from the base housing member and defines the chamber and, in an assembled position, is configured to extend into a corresponding sealing feature on the cover housing member to seal a sample therein.

11. The collection device of Claim 10, wherein the base housing member comprises a central portion and a perimeter portion, and the wall portion of the plurality of chambers has a height that is higher in the central portion than at the perimeter portion of the base housing member.

12. The collection device of Claim 10, wherein the plurality of chambers comprises an fluid transfer feature comprising a protrusion having an aperture therein that is configured to fluidly transfer a sample from one of the respective plurality of chambers to a corresponding sample area of a sample cartridge.

13. The collection device of Claim 12, wherein aperture of the fluid transfer feature is configured to fit inside a corresponding well of the sample cartridge.

14. The collection device of Claim 1, further comprising a label, bar code and/or RFID that identifies an origin of a sample.

15. A collection and transfer assembly comprising:

a collection device comprising:

a base housing member having at least one chamber; and

a cover housing member having at least one housing aperture therein, the cover housing member being configured to cover the base housing member such that the at least one housing aperture is positioned in fluid communication with the at least one chamber, the cover housing member further comprising a cartridge holding interface; and

a sample cartridge comprising:

at least one sample area having at least one cartridge aperture that is configured to receive a fluid from the at least one chamber via the at least one housing aperture, wherein the cartridge holding interface on the cover housing member is configured to releasably engage with the sample cartridge.

16. The collection and transfer assembly of Claim 15, wherein the sample cartridge has a first major surface opposite a second major surface, the at least one sample area is on the first major surface and faces away from the cover housing member, and the at least one cartridge aperture is on the second major surface and fluidly connects the housing aperture to the sample area.

17. The collection and transfer assembly of Claim 15, wherein the sample cartridge has a first major surface opposite a second major surface, the at least one sample area is on the second major surface and faces toward the cover housing member, and the at least one cartridge aperture is on the second major surface and fluidly connects the housing aperture to the sample area.

18. The collection and transfer assembly of Claim 15, wherein the cartridge holding interface comprises a retaining wall or retaining pins configured to abut an outer perimeter of the cartridge.

19. The collection and transfer assembly of Claim 18, wherein the cartridge holding interface further comprises at least one notch and/or groove that is configured to engage a corresponding notch and/or groove on the cartridge.

20. The collection and transfer assembly of Claim 18, wherein the cartridge holding interface further comprises at least one hook member that is configured to engage and retain an edge portion of the cartridge.

21. The collection and transfer assembly of Claim 15, wherein the cartridge holding interface is configured to interface with the cartridge in a single orientation.

22. The collection and transfer assembly of Claim 15, wherein the at least one chamber comprises a plurality of chambers and the at least one aperture comprises a plurality of apertures, wherein the base housing member and the cover housing member comprises cooperating sealing members that are configured to seal each of the plurality of chambers.

23. The collection and transfer assembly of Claim 22, wherein the cooperating sealing members comprise a base sealing feature between the plurality of wells on the base housing member, and a cover sealing feature on the cover housing member configured to engage with the base sealing feature and to thereby fluidly seal each of the plurality of wells.

24. The collection and transfer assembly of Claim 23, wherein one of the base sealing feature and the cover sealing feature comprises a groove and the other of the base sealing feature and the cover sealing feature comprises a ridge that is configured to be received in the groove and form a snug fit.

25. The collection and transfer assembly of Claim 22, wherein the chambers overlap a central portion and a perimeter portion of the base housing member, and the chambers comprise a wall that has a height that is higher in the central portion than at the perimeter portion.

26. The collection and transfer assembly of Claim 15, wherein the at least one chamber comprises a plurality of chambers and the at least one aperture comprises a plurality of corresponding apertures, wherein each of the plurality of chambers comprises wall portion that extends away from the base housing member and defines the chamber and, in an assembled position, is configured to extend into a corresponding sealing feature on the cover housing member to seal a sample therein.

27. The collection and transfer assembly of Claim 15, wherein the base housing member comprises a central portion and a perimeter portion, and the wall portion of the plurality of chambers has a height that is higher in the central portion than at the perimeter portion of the base housing member.

28. The collection and transfer assembly of Claim 15, wherein the plurality of chambers comprises an fluid transfer feature comprising a protrusion having an aperture therein that is configured to fluidly transfer a sample from one of the respective plurality of chambers to a corresponding sample area of a sample cartridge.

29. The collection and transfer assembly of Claim 28, wherein aperture of the fluid transfer feature is configured to fit inside a corresponding well of the sample cartridge.

30. The collection and transfer assembly of Claim 15, further comprising a label, bar code and/or RFID that identifies an origin of a sample.

31. The collection and transfer assembly of Claim 15, further comprising a plurality of exogeneous targets in the at least one sample area, wherein said exogenous targets are particles.

32. The collection and transfer assembly of Claim 31, wherein said exogenous targets have an average diameter of from 0.1 micrometers up to 10 micrometers.

33. The collection and transfer assembly of Claim 31-32, wherein said exogenous targets are fluorescent.

34. The collection and transfer assembly of Claim 31, wherein said exogenous targets:

fluoresce at a peak absorption wavelength of at least 420 nanometers and at not more than 540 nanometers;

fluoresce at a peak emission wavelength of at least 450 nanometers and not more than 590 nanometers;

and wherein said peak absorption wavelength and said peak emission wavelength differ by at least 10 nanometers.

35. A method of collecting a sample, the method comprising:

collecting a sample in at least one chamber of a base housing member of a collection device;

place a cover housing member on the base housing member of the collection device, the cover housing member having at least one housing aperture therein, the cover housing member being configured to cover the base housing member such that the at least one housing aperture is positioned in fluid communication with the at least one chamber, the cover housing member further comprising a cartridge holding interface; and

position a sample cartridge on the cartridge holding interface, the sample cartridge comprising at least one sample area having at least one cartridge aperture that is configured to receive a fluid from the at least one chamber via the at least one housing aperture; and

inverting the cover and base housing members such that the sample flows from the at least one chamber to the at least one sample area via the at least one housing aperture and the at least one cartridge aperture.

36. The method of Claim 35, further comprising placing the cartridge in an imaging reader.

37. The method of Claim 35, further comprising storing the collection device and the sample cartridge before the inverting step.

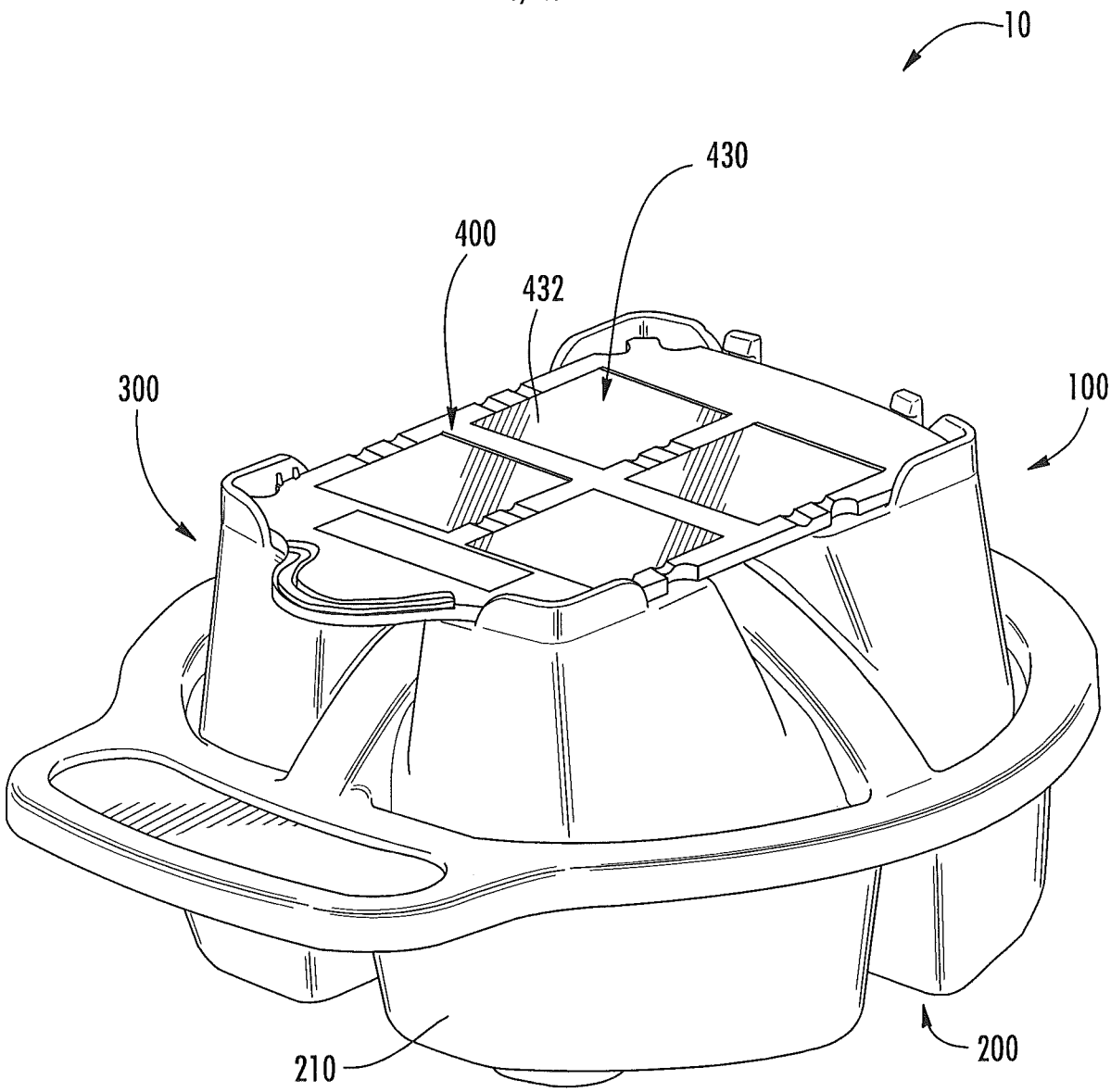

FIG. 1

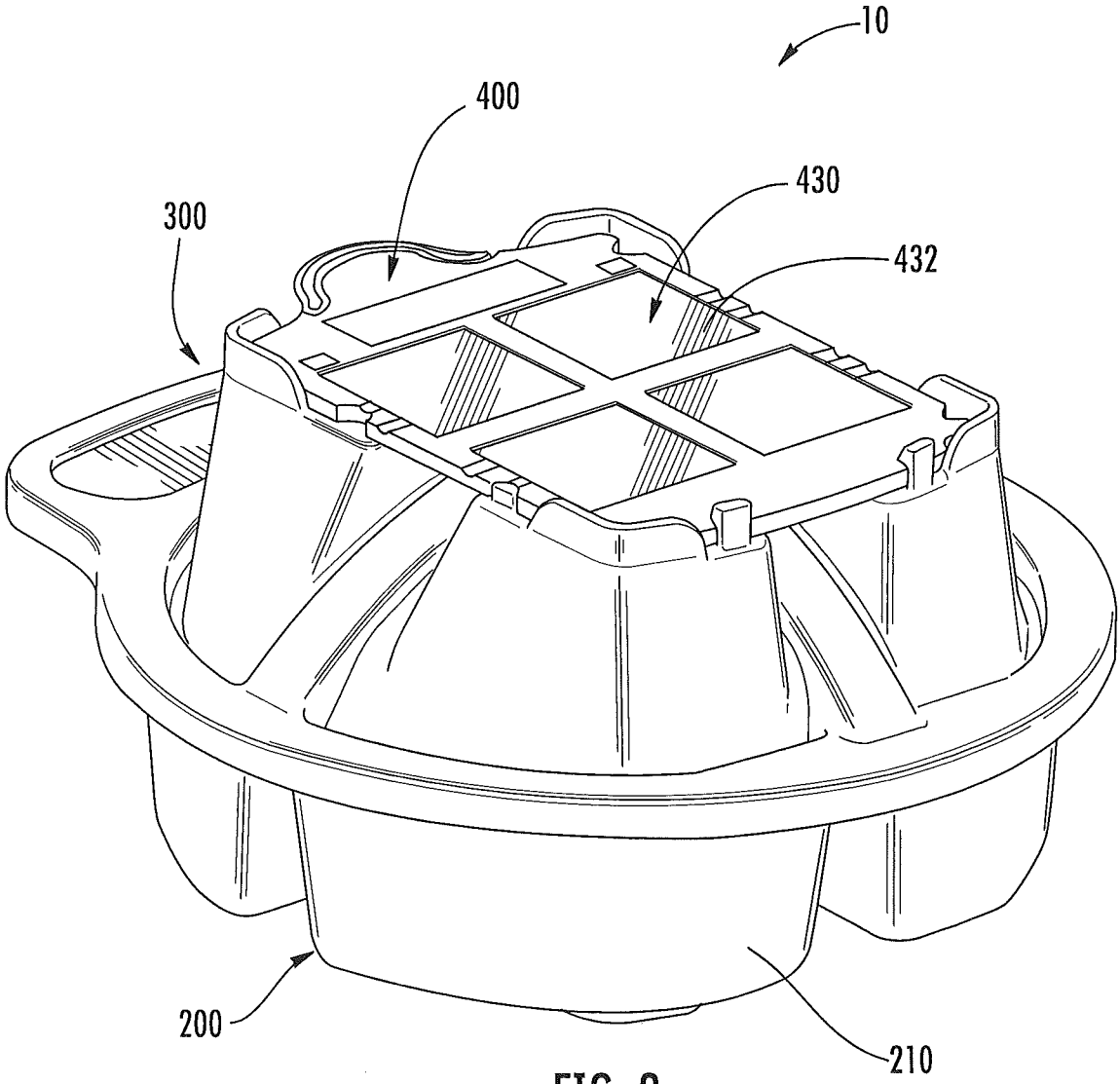

FIG. 2

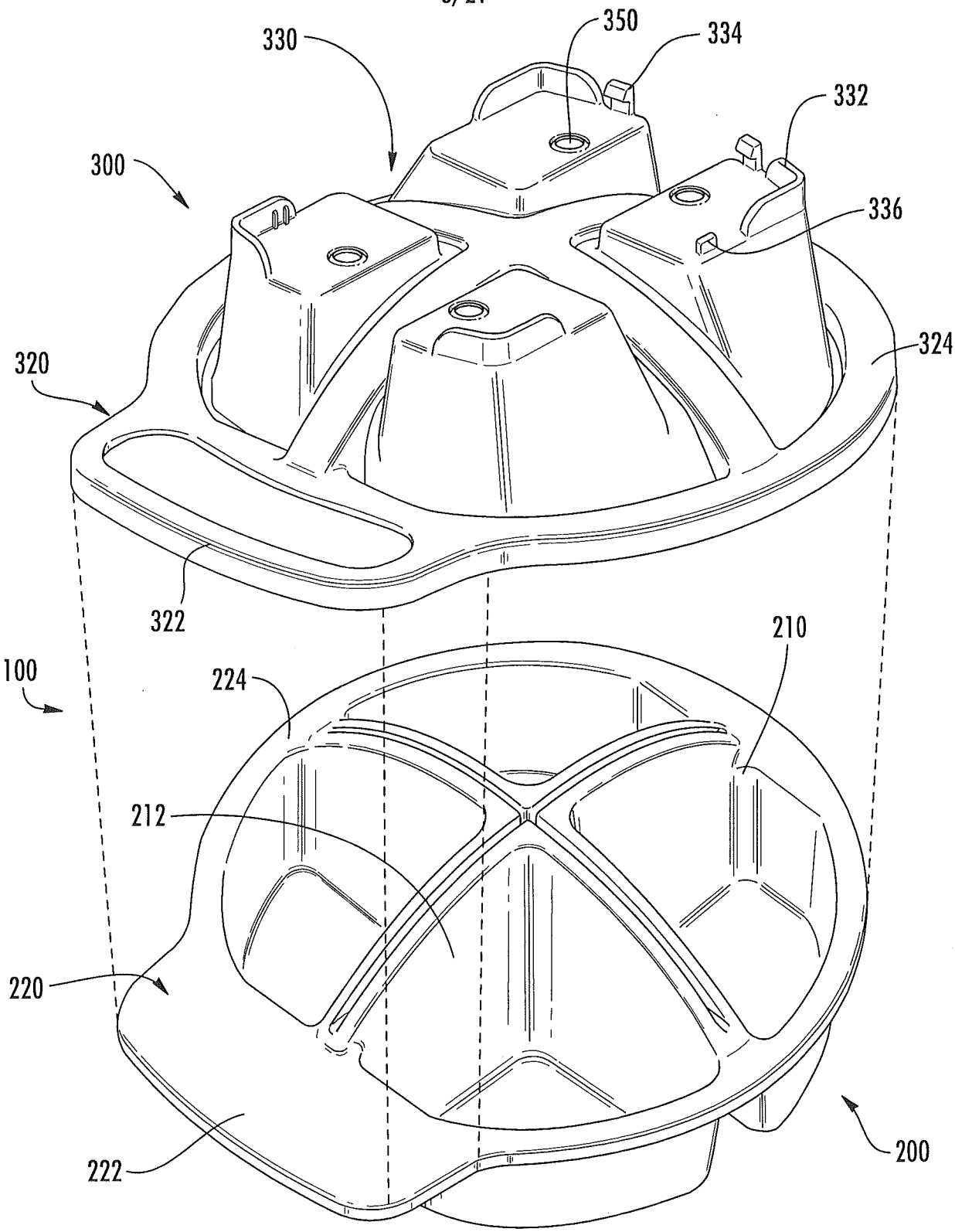

FIG. 3

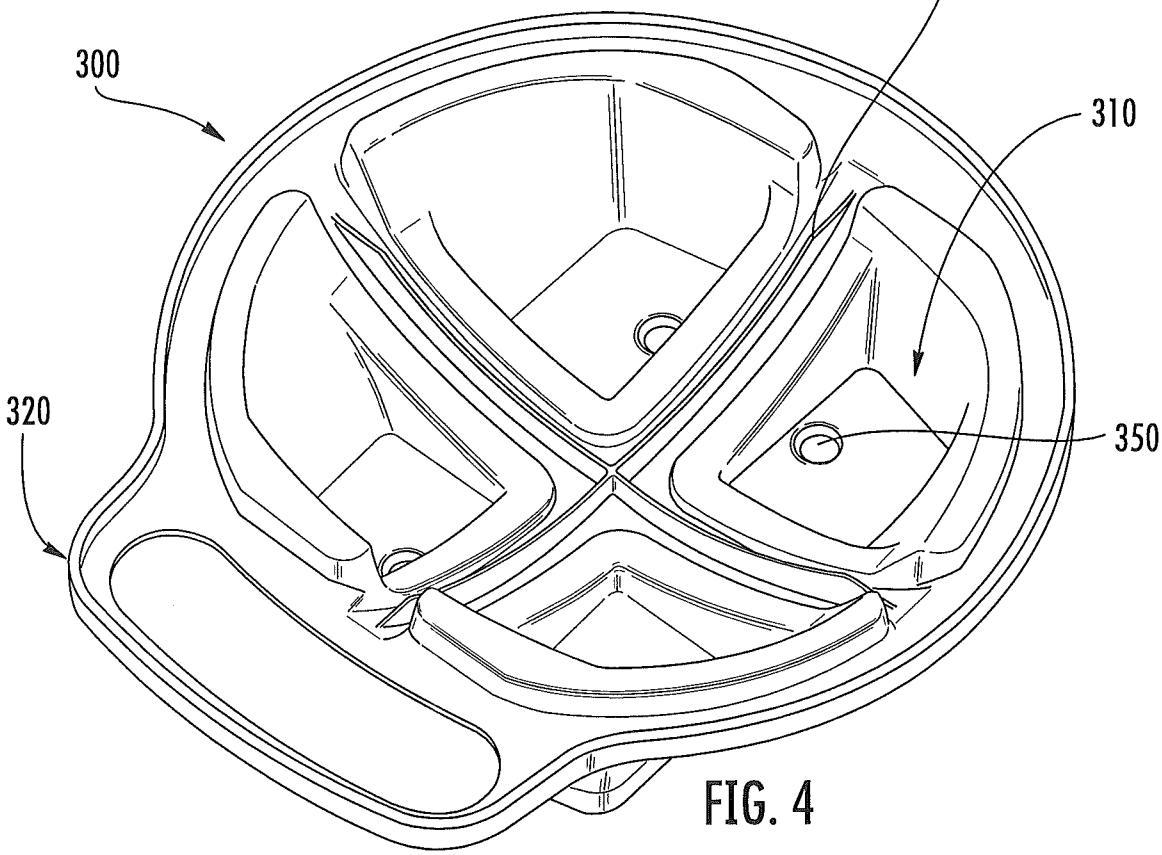

FIG. 4

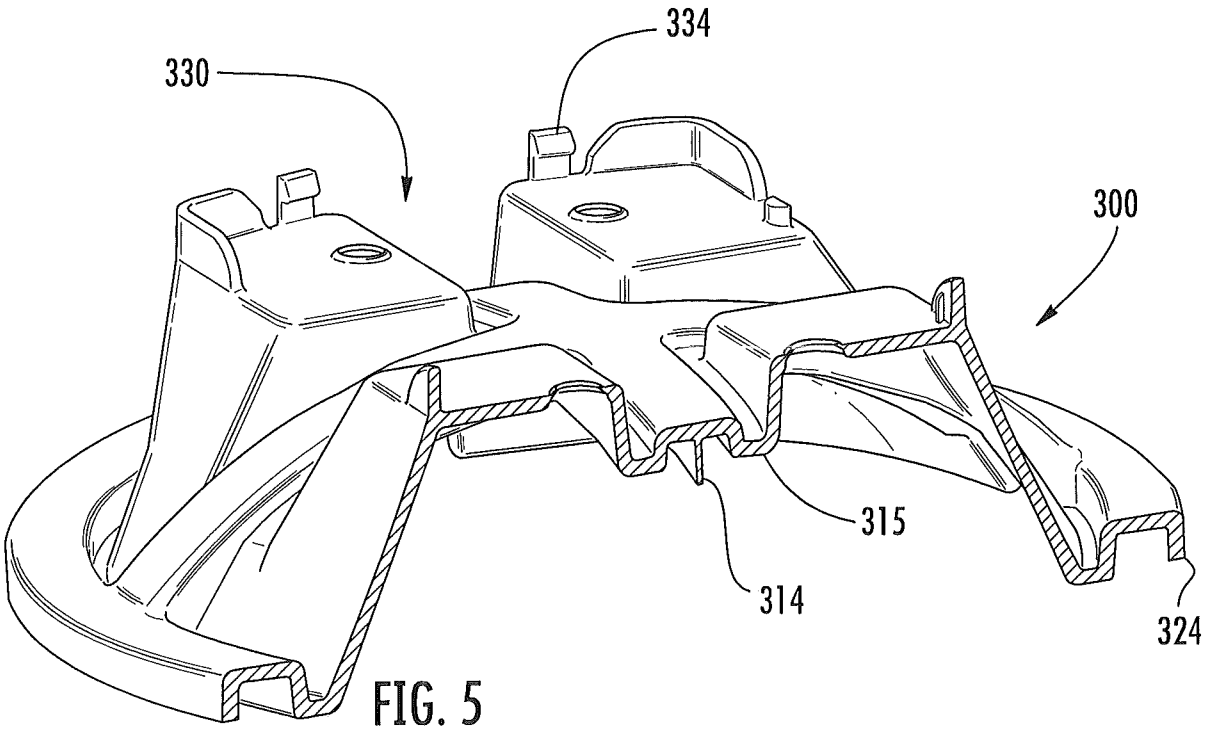

FIG. 5

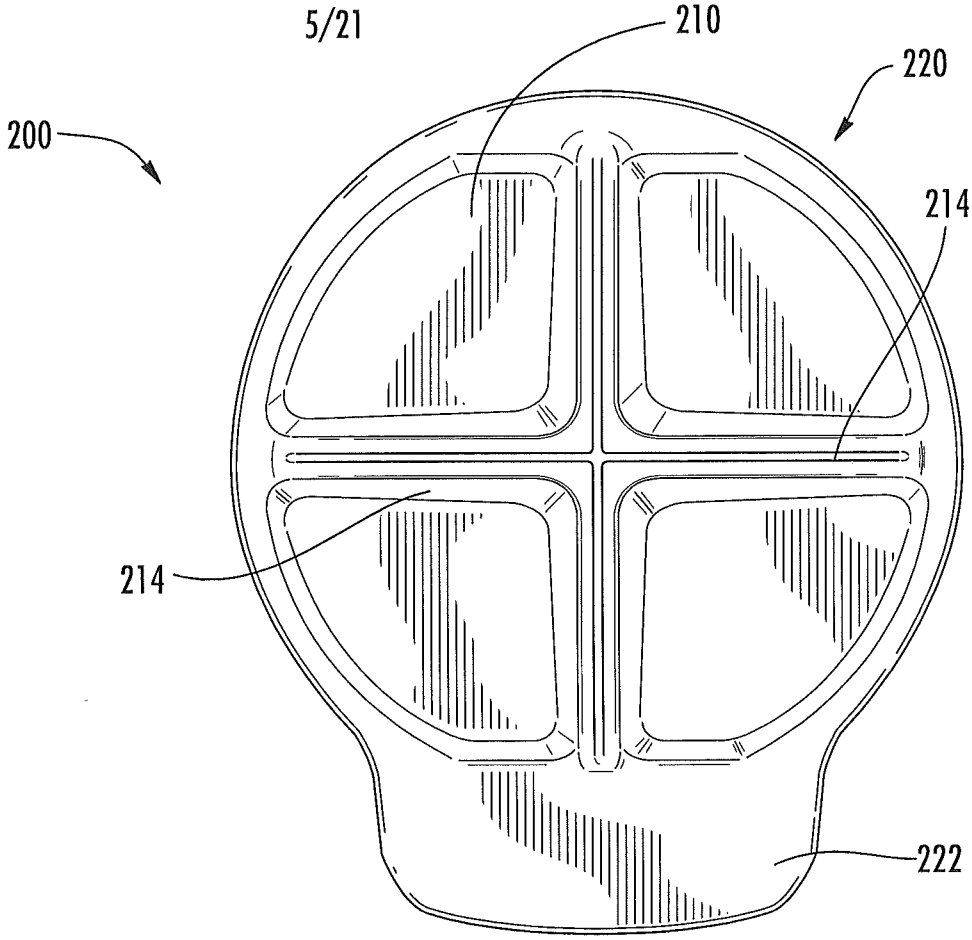

FIG. 6

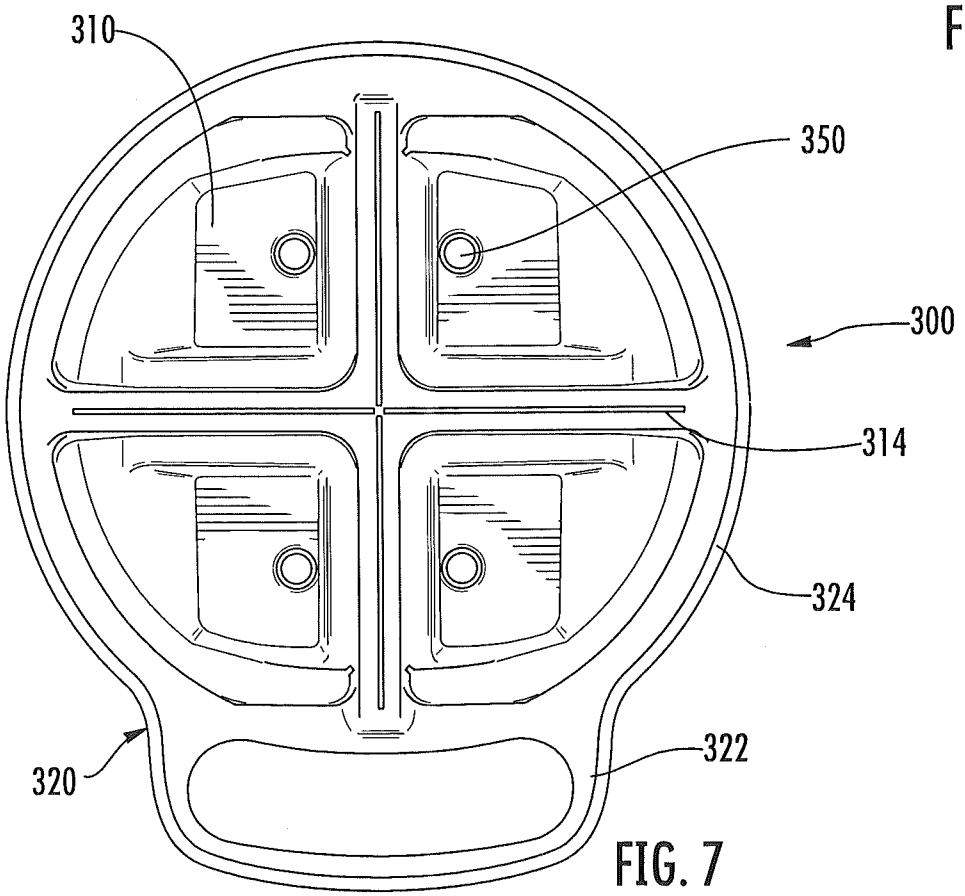

FIG. 7

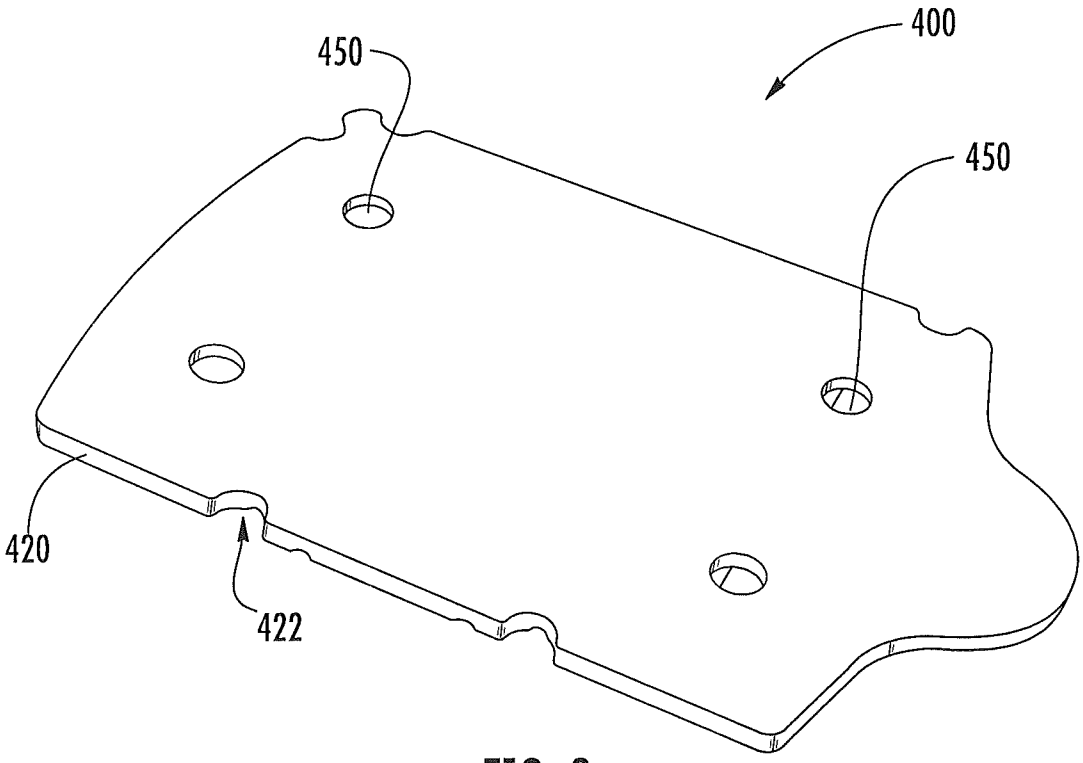

FIG. 8

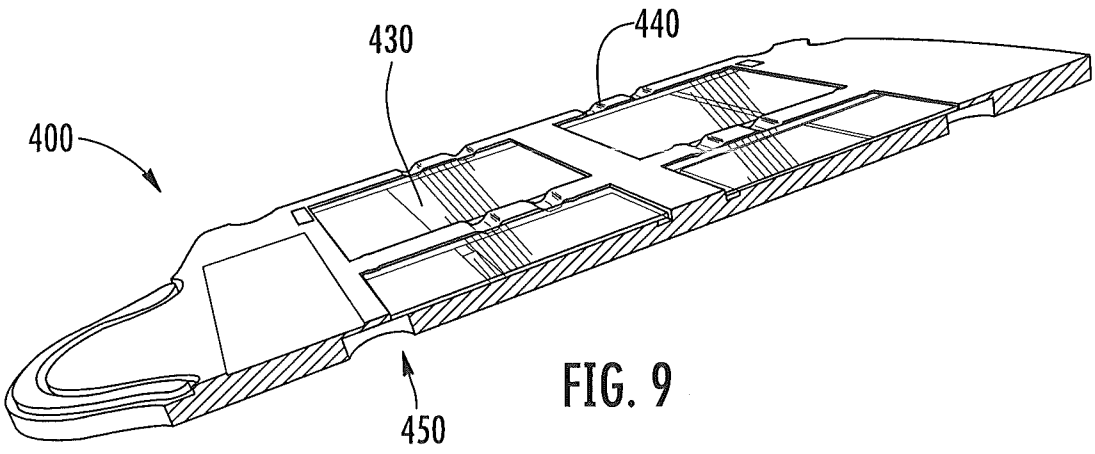

FIG. 9

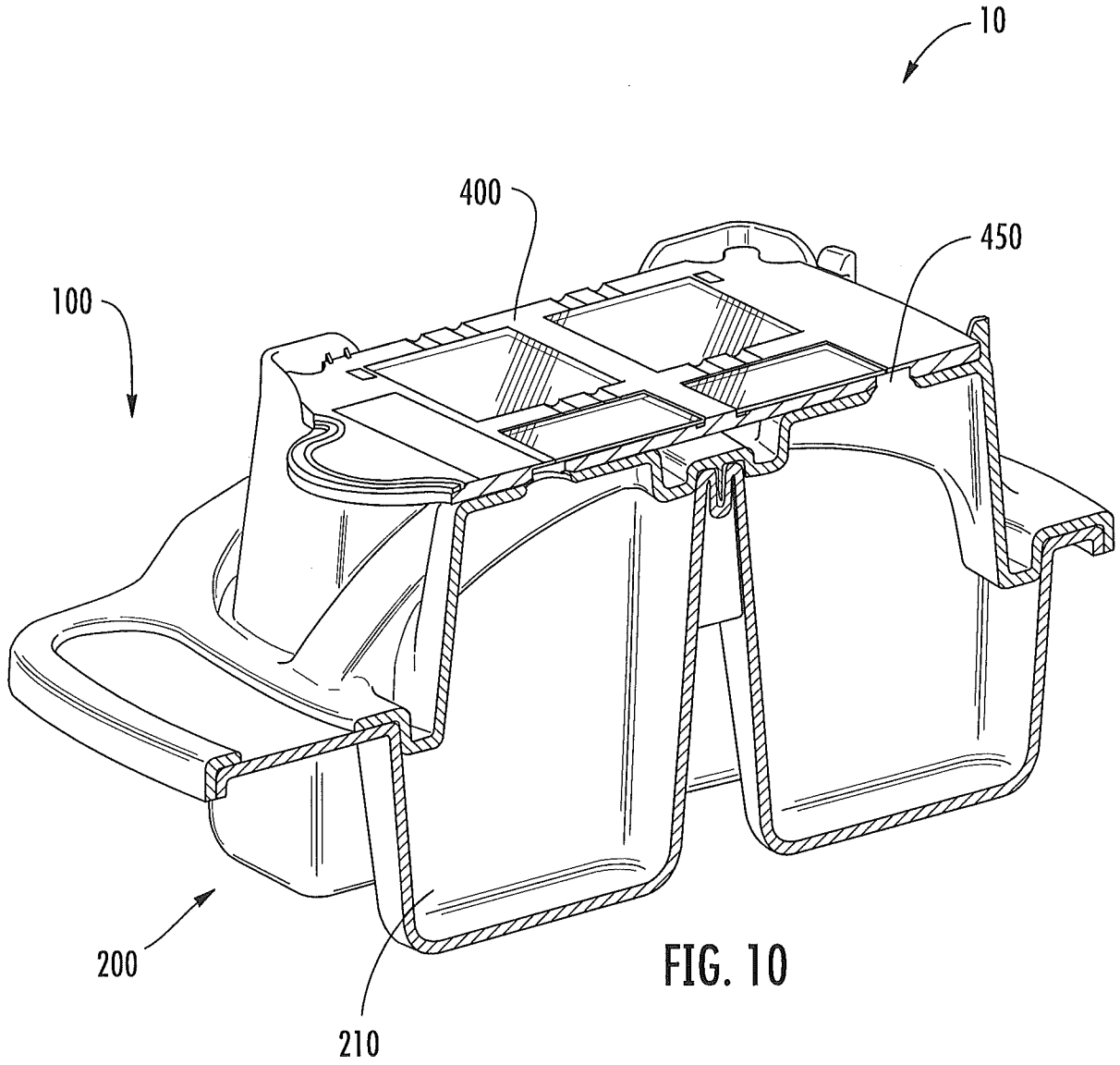

FIG. 10

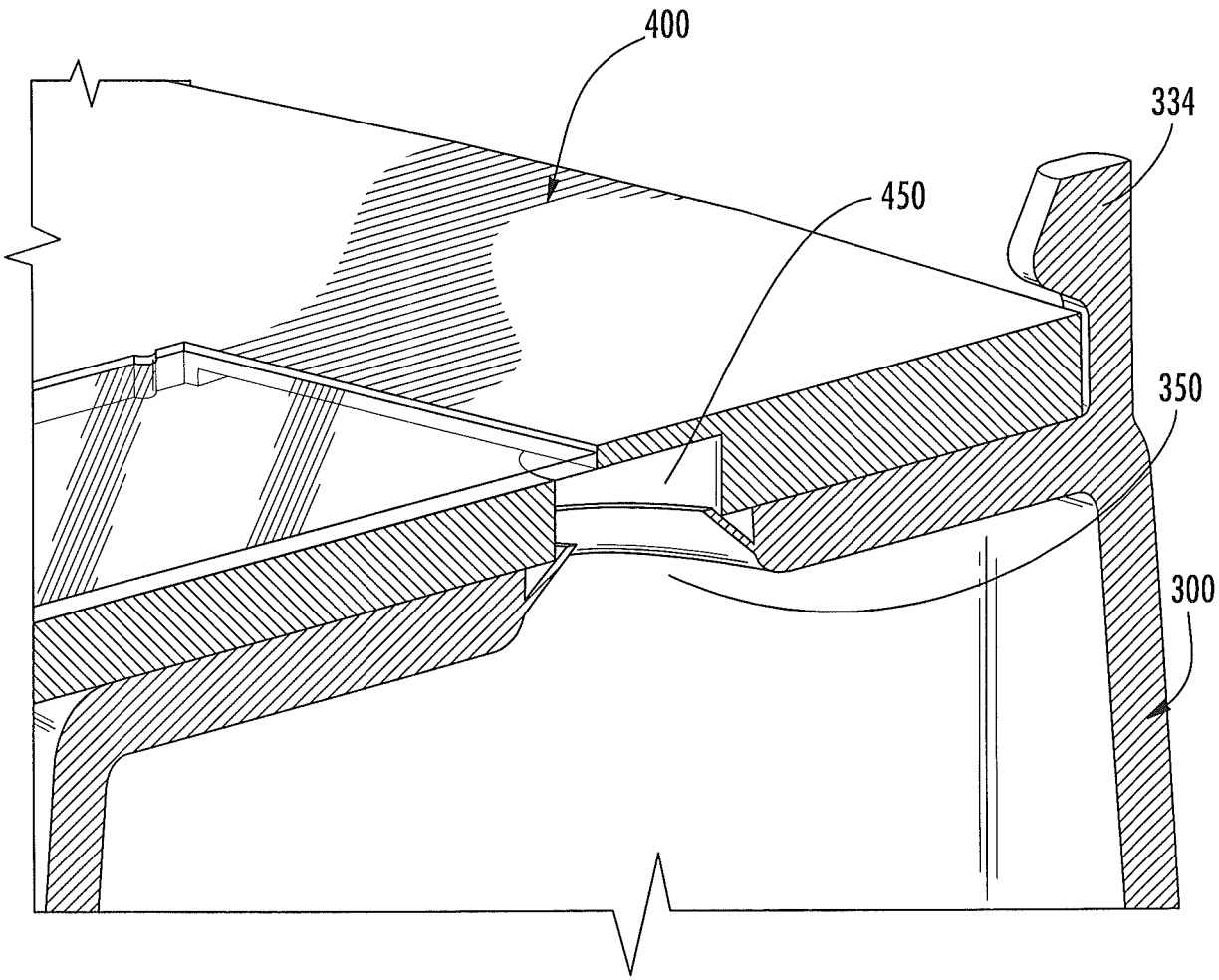

FIG. 11

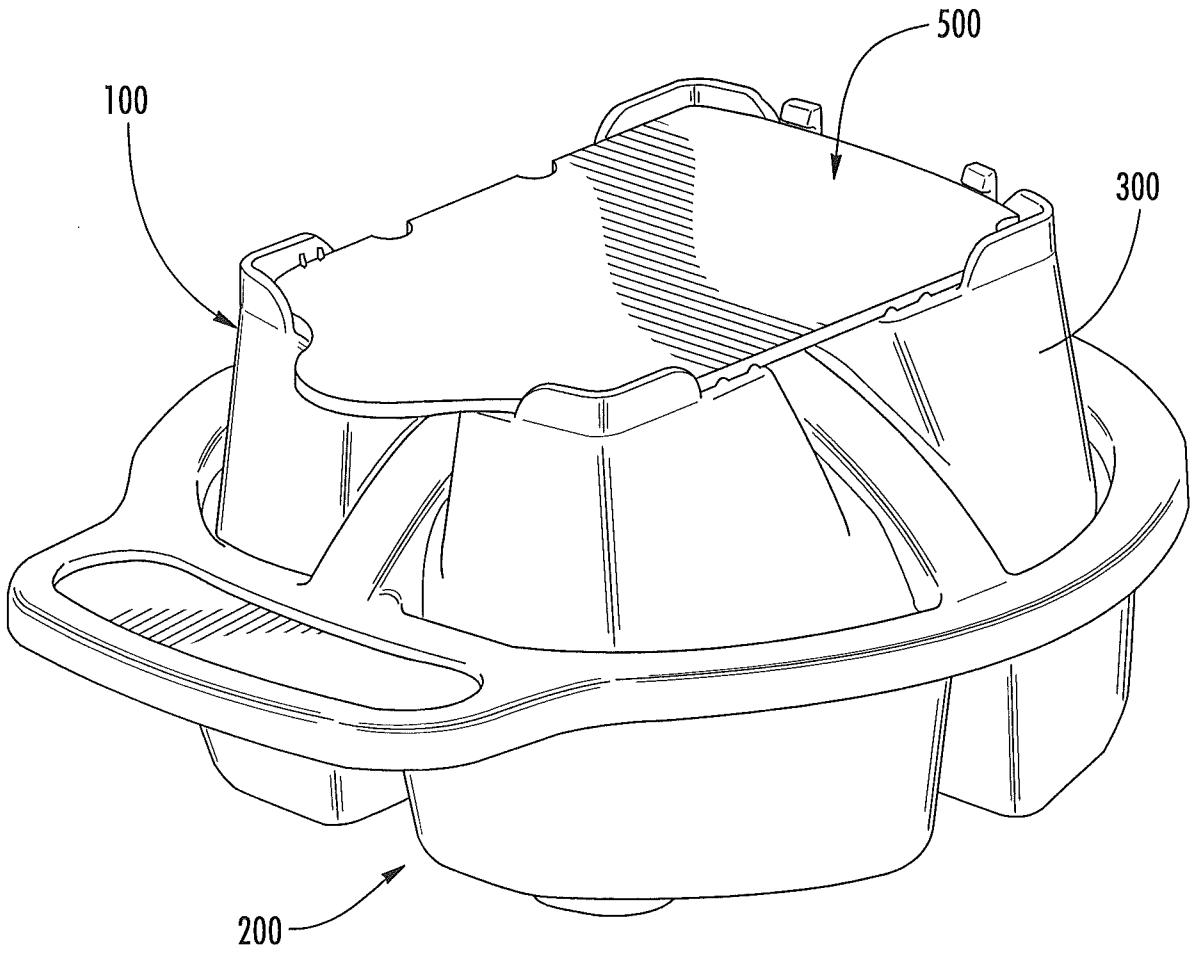

FIG. 12

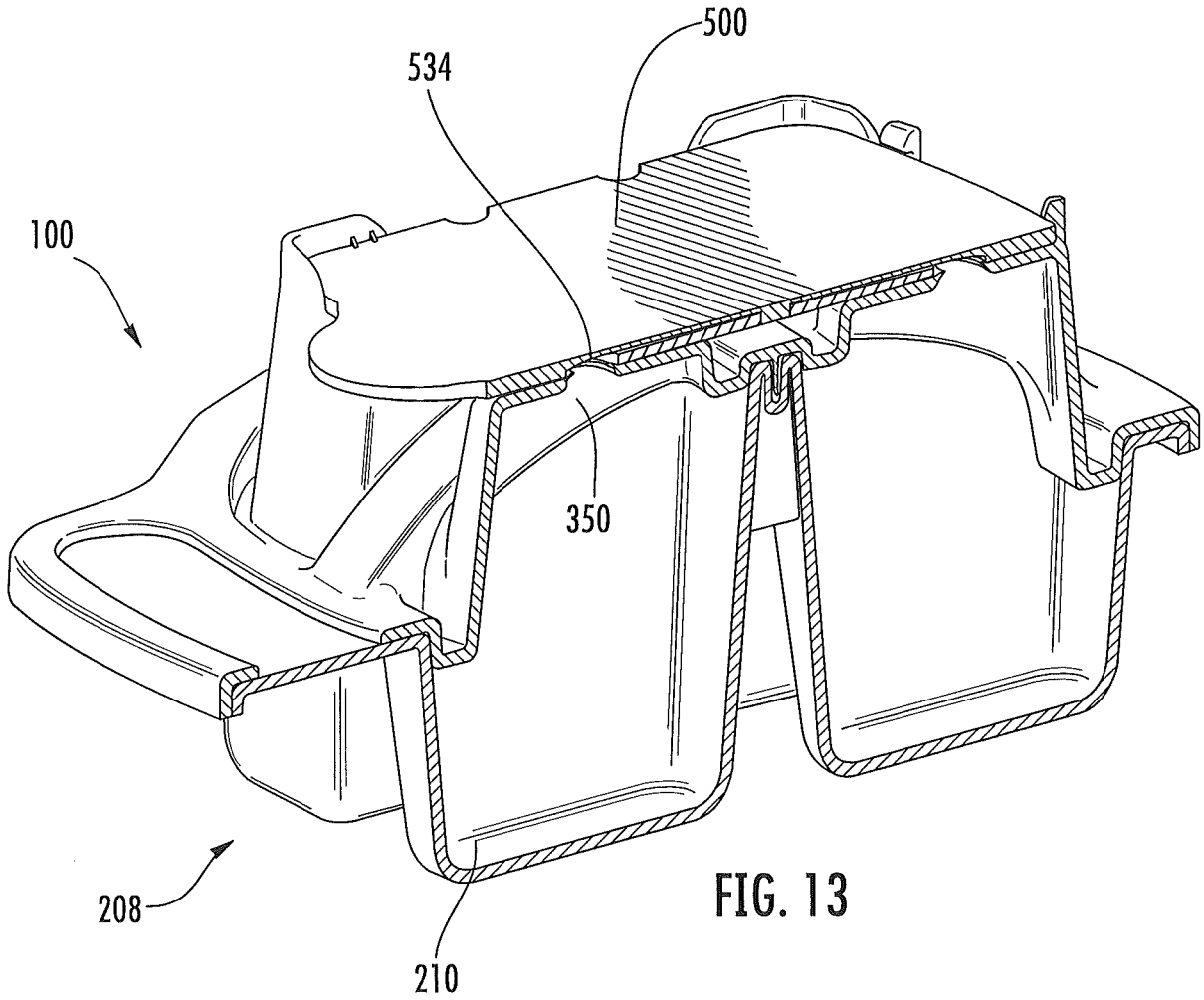

FIG. 13

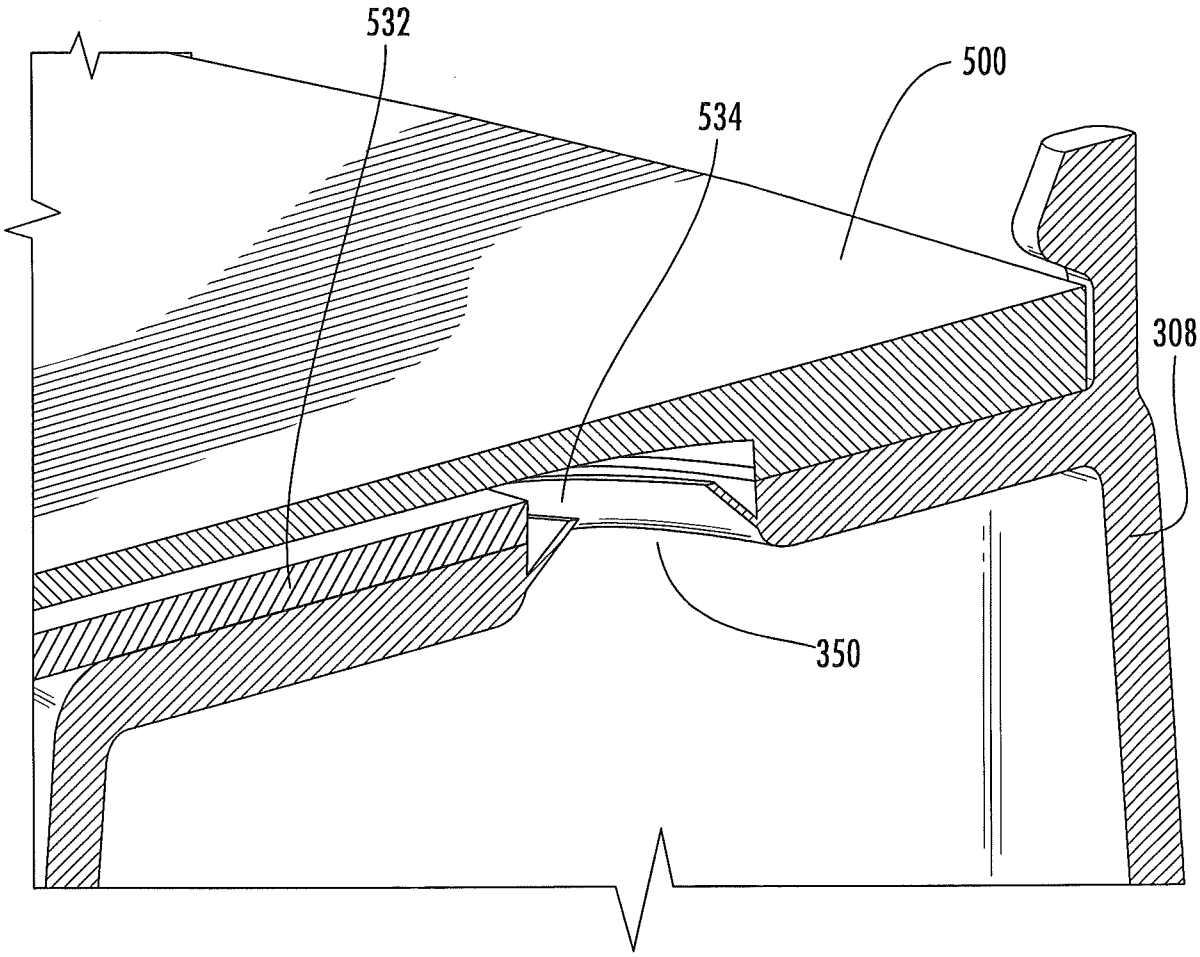

FIG. 14

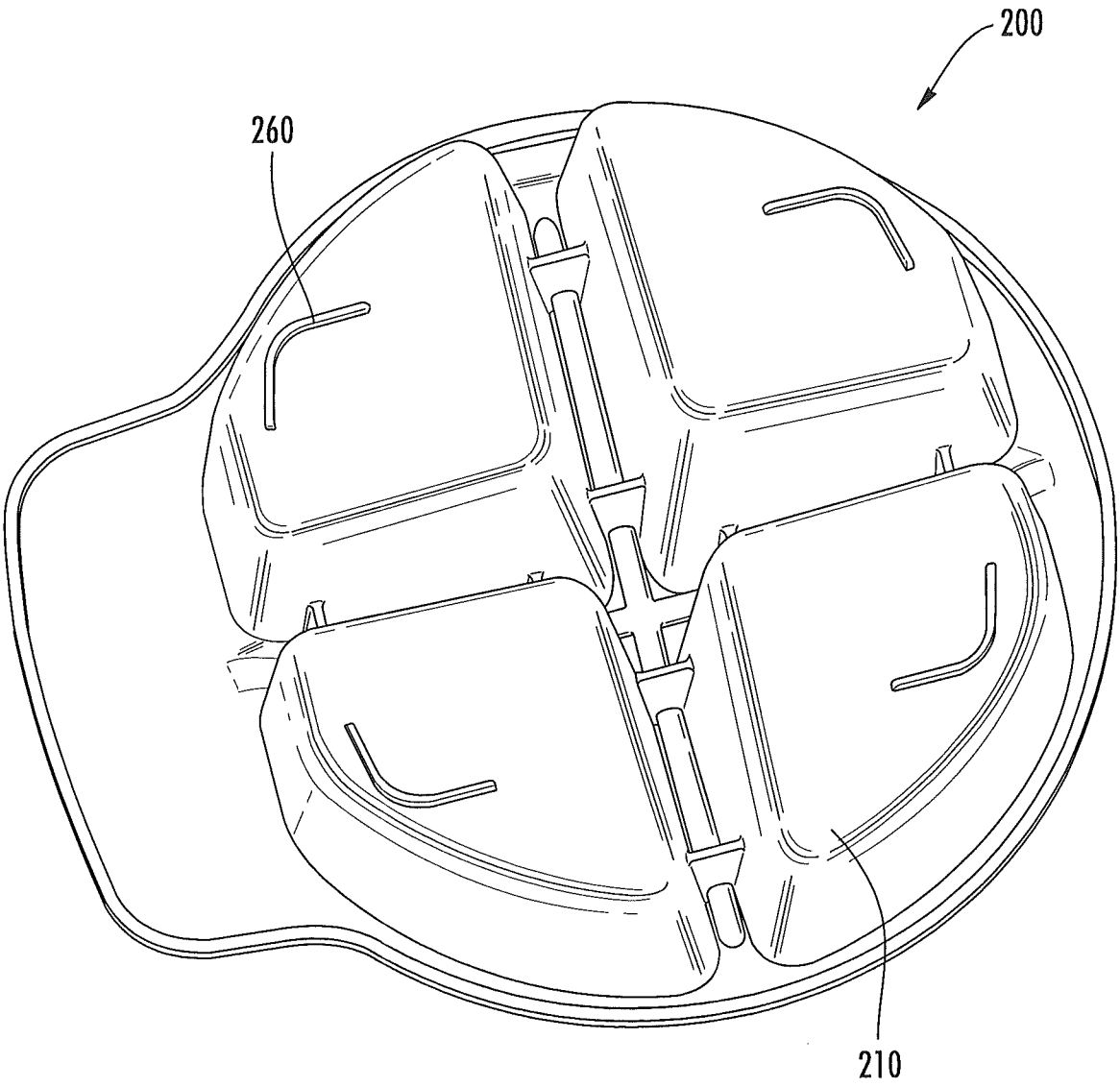

FIG. 15

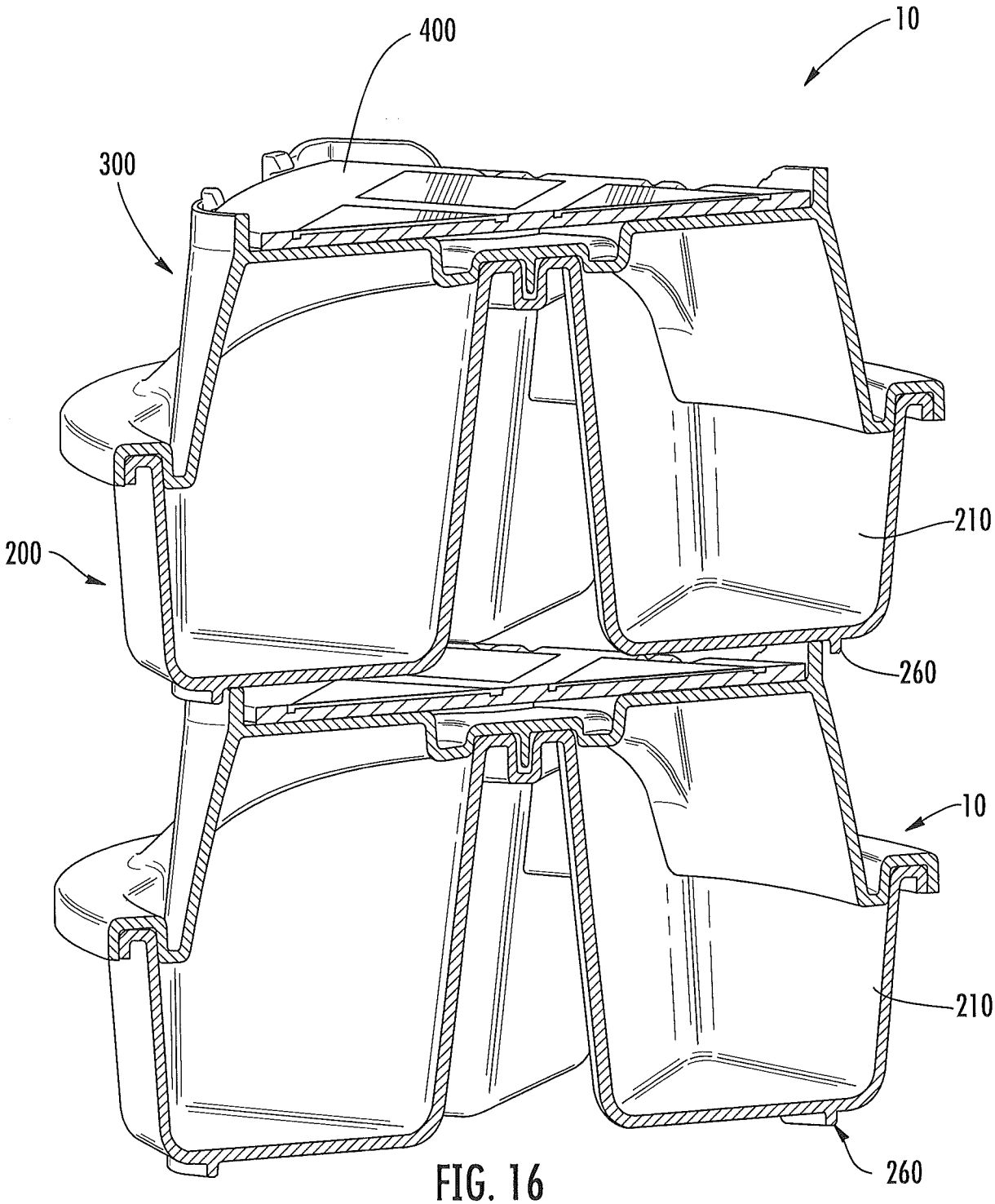

FIG. 16

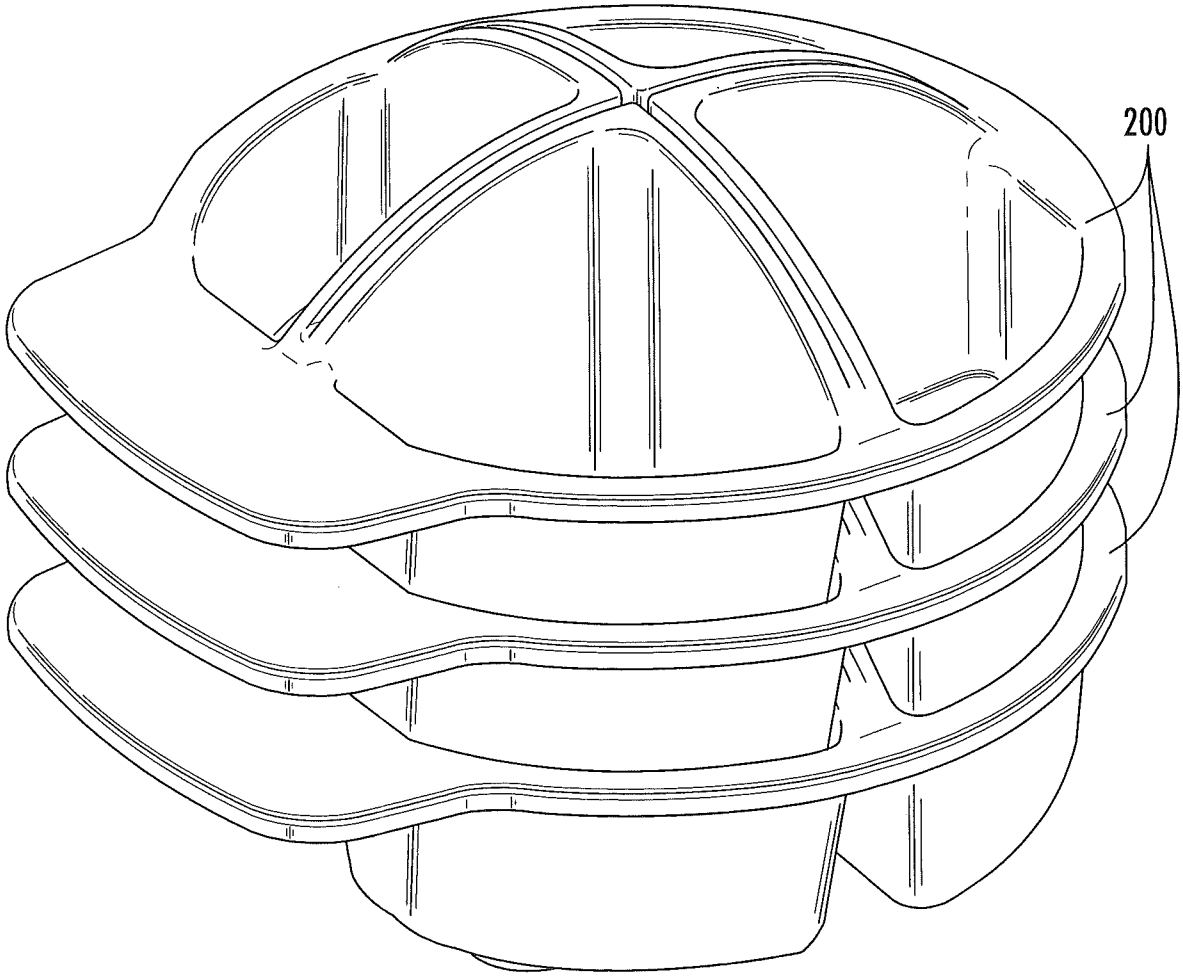

FIG. 17

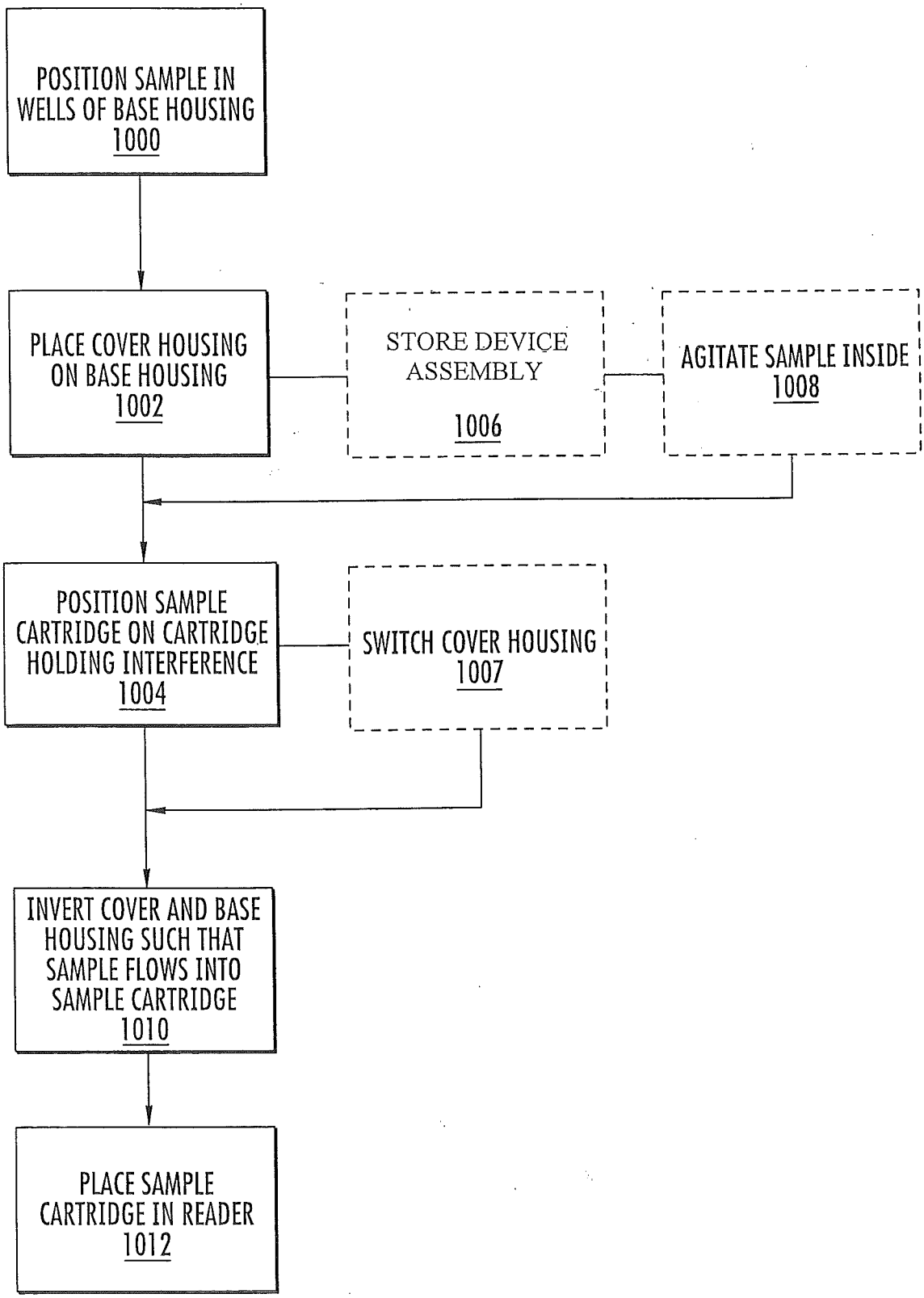
POSITION SAMPLE IN WELLS OF BASE HOUSING 1000
PLACE COVER HOUSING ON BASE HOUSING 1002
STORE DEVICE ASSEMBLY 1006
AGITATE SAMPLE INSIDE 1008
POSITION SAMPLE CARTRIDGE ON CARTRIDGE HOLDING INTERFERENCE 1004
SWITCH COVER HOUSING 1007
INVERT COVER AND BASE HOUSING SUCH THAT SAMPLE FLOWS INTO SAMPLE CARTRIDGE 1010
PLACE SAMPLE CARTRIDGE IN READER 1012

FIG. 18

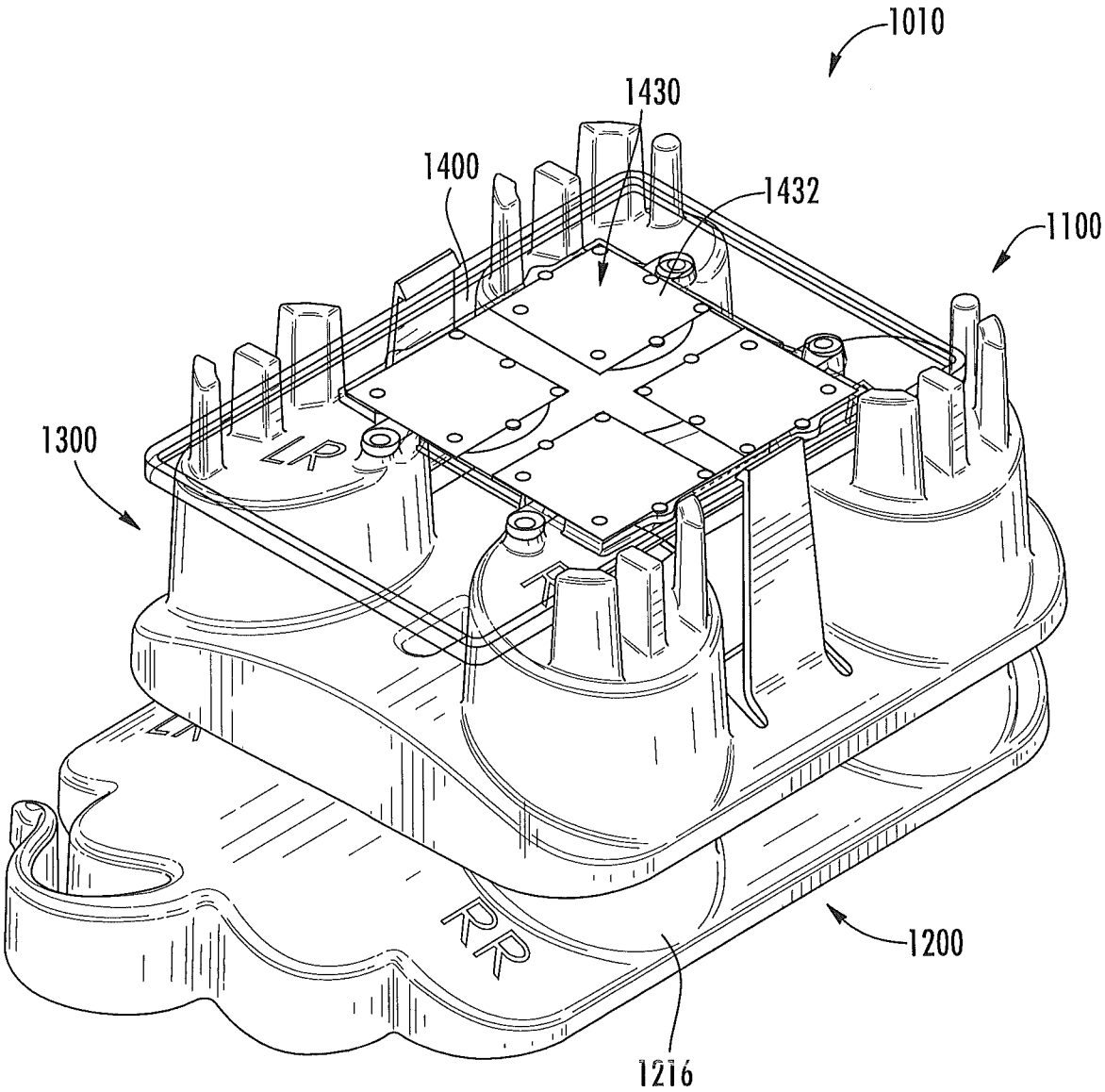

FIG. 19

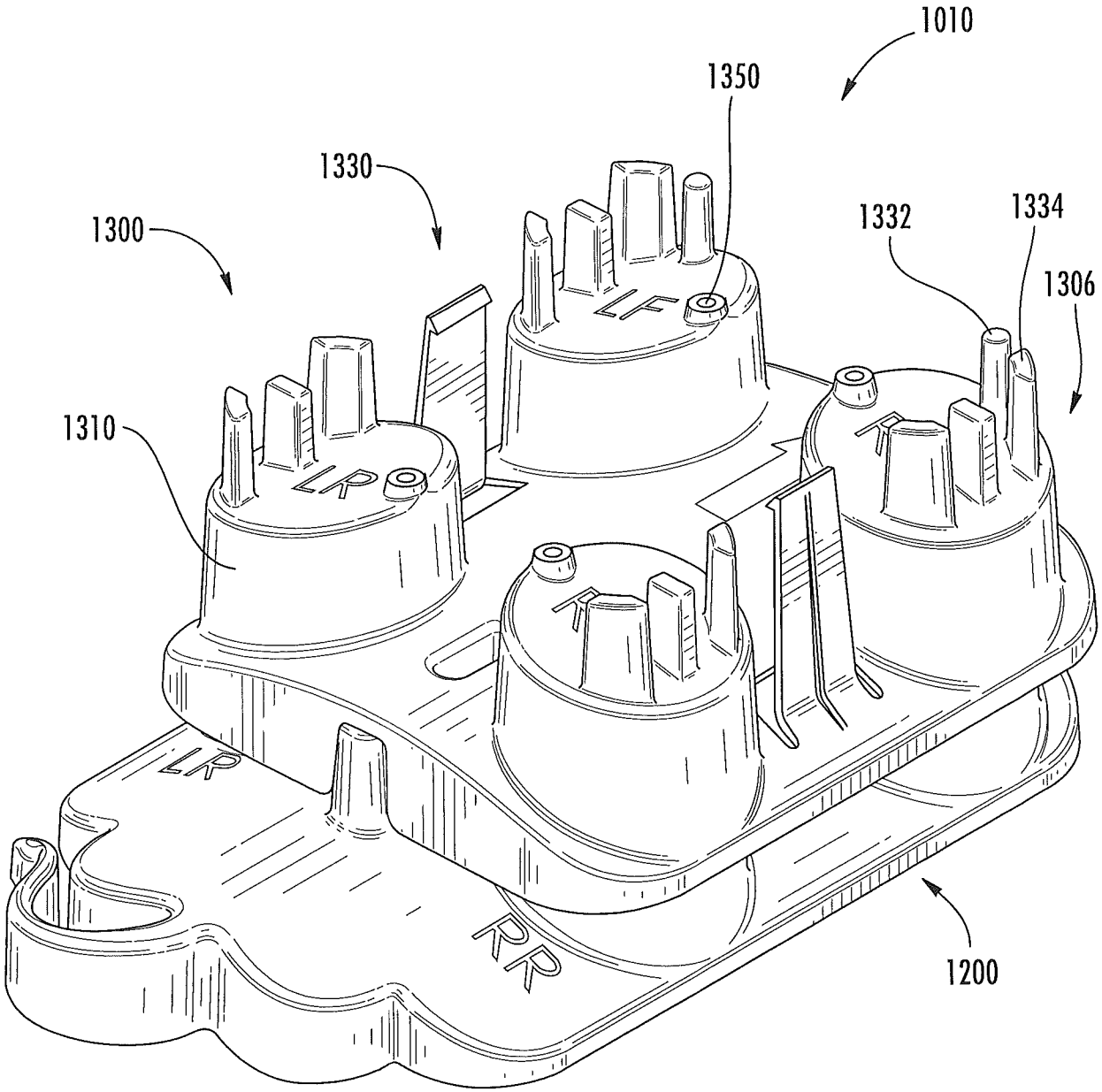

FIG. 20

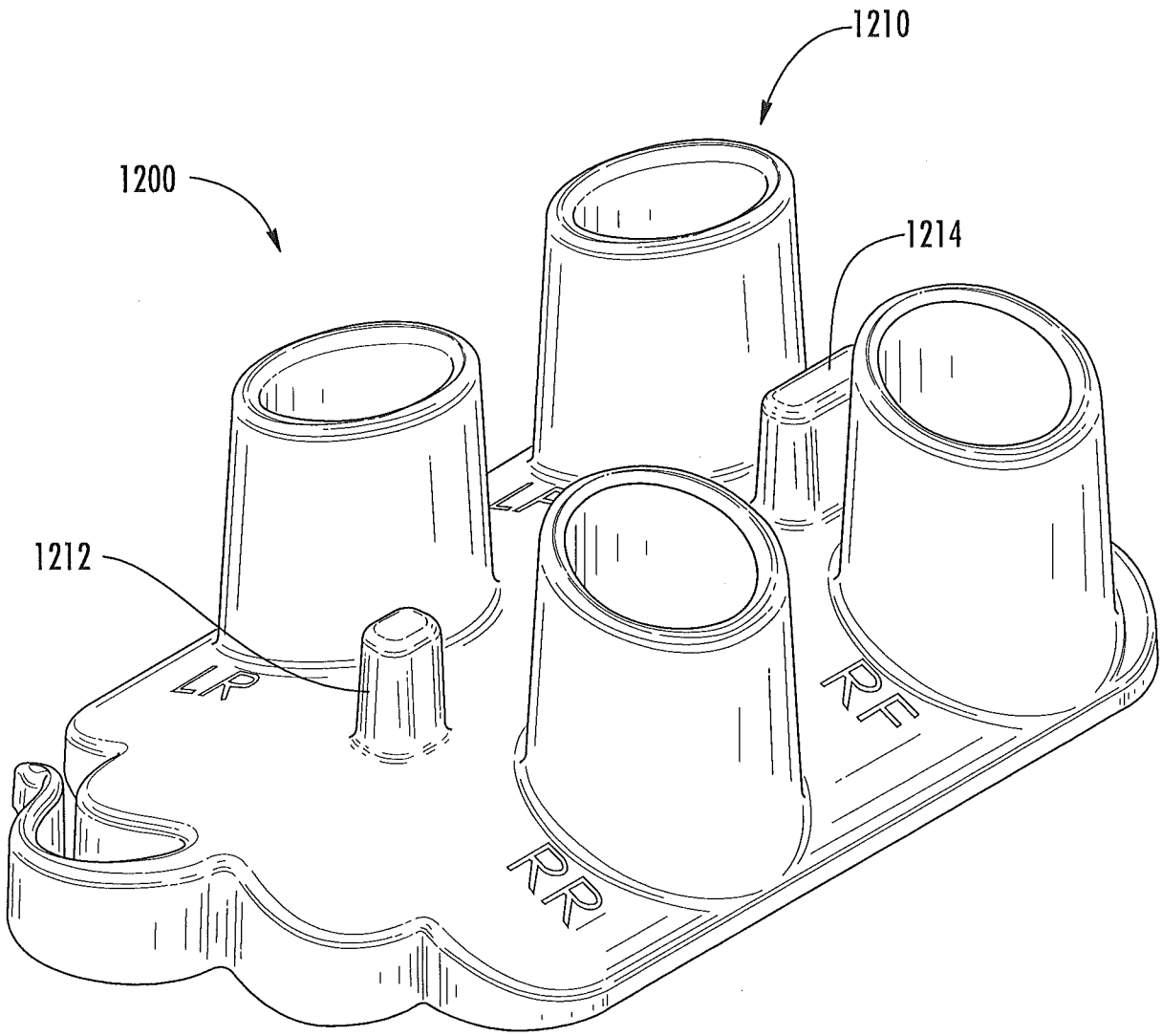

FIG. 21

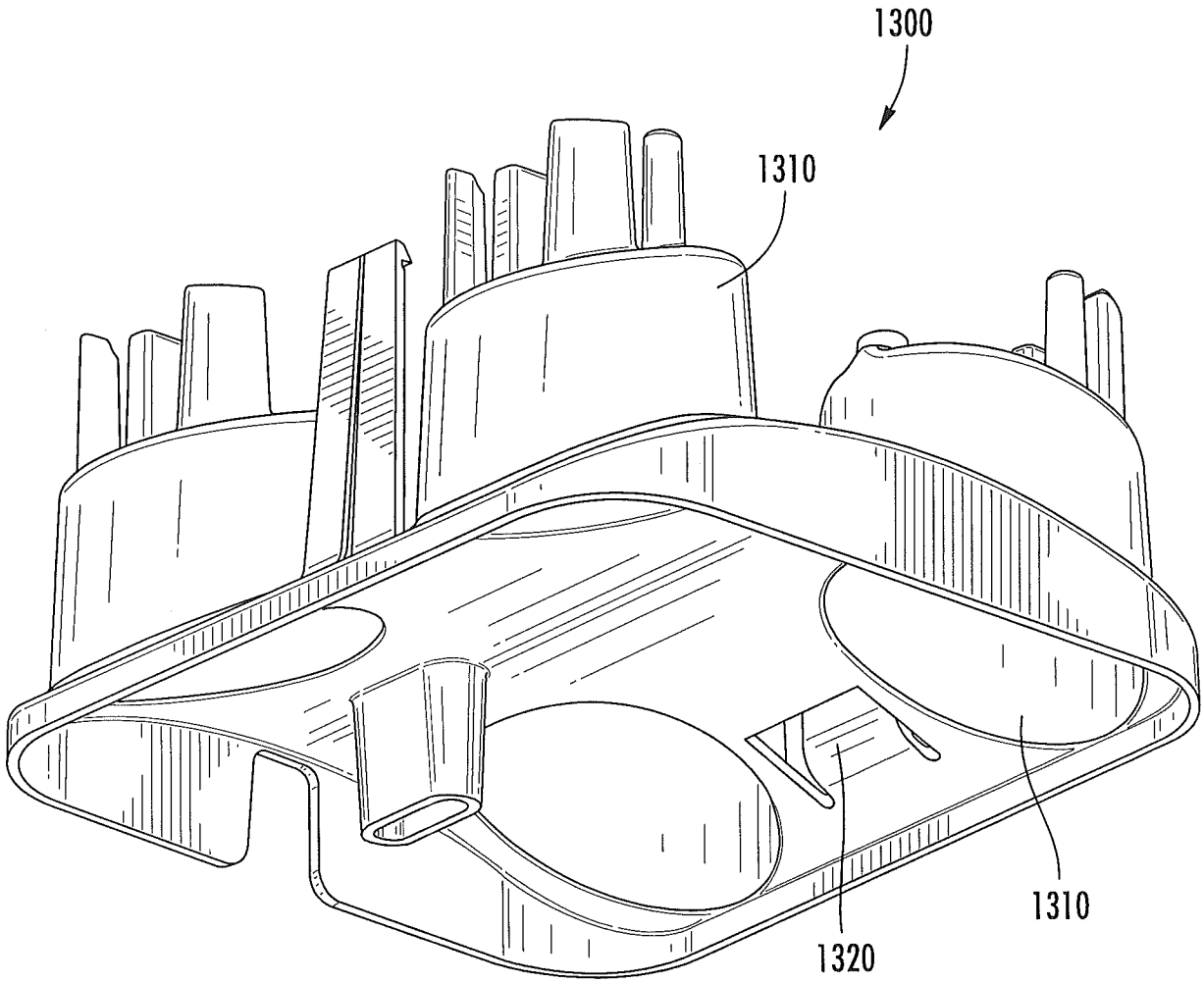

FIG. 22

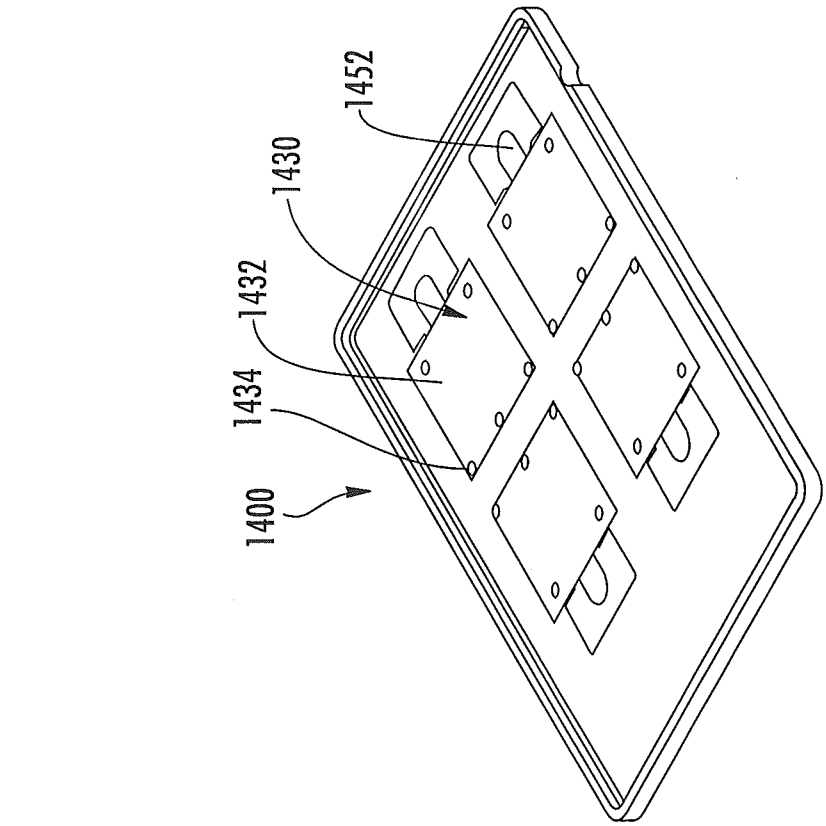

FIG. 24

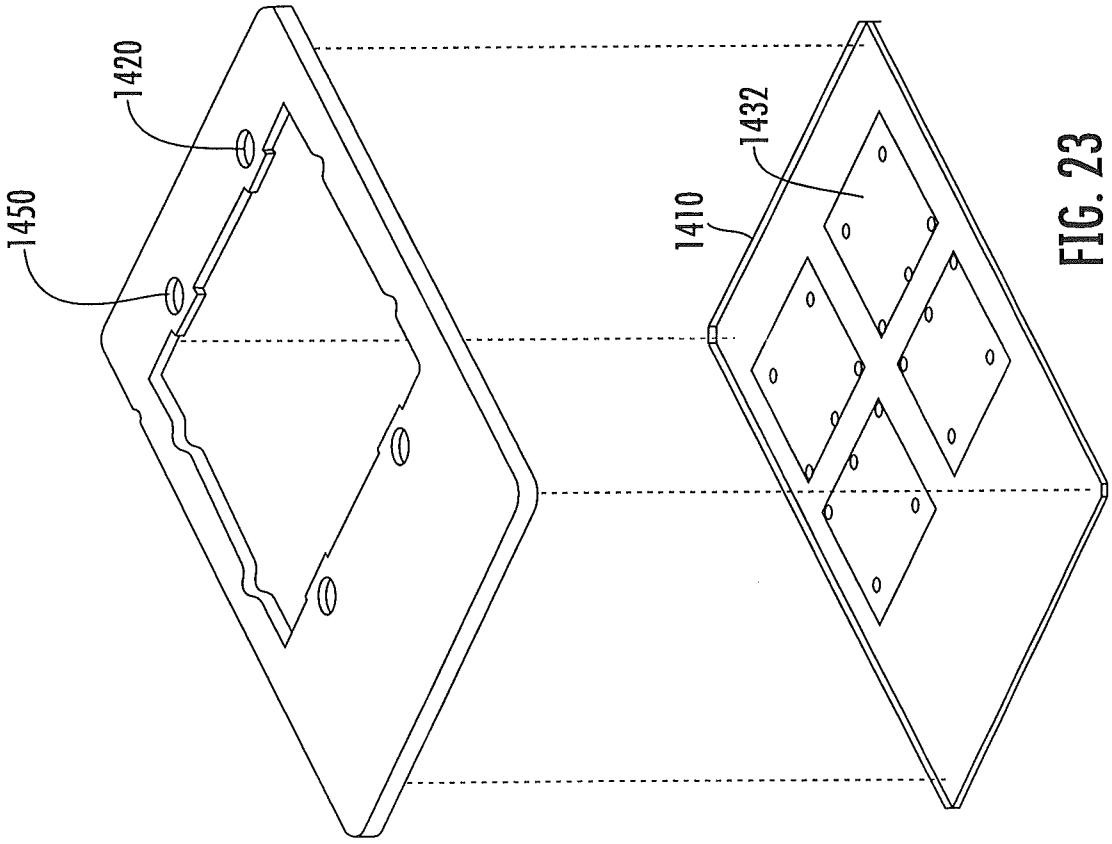

FIG. 23

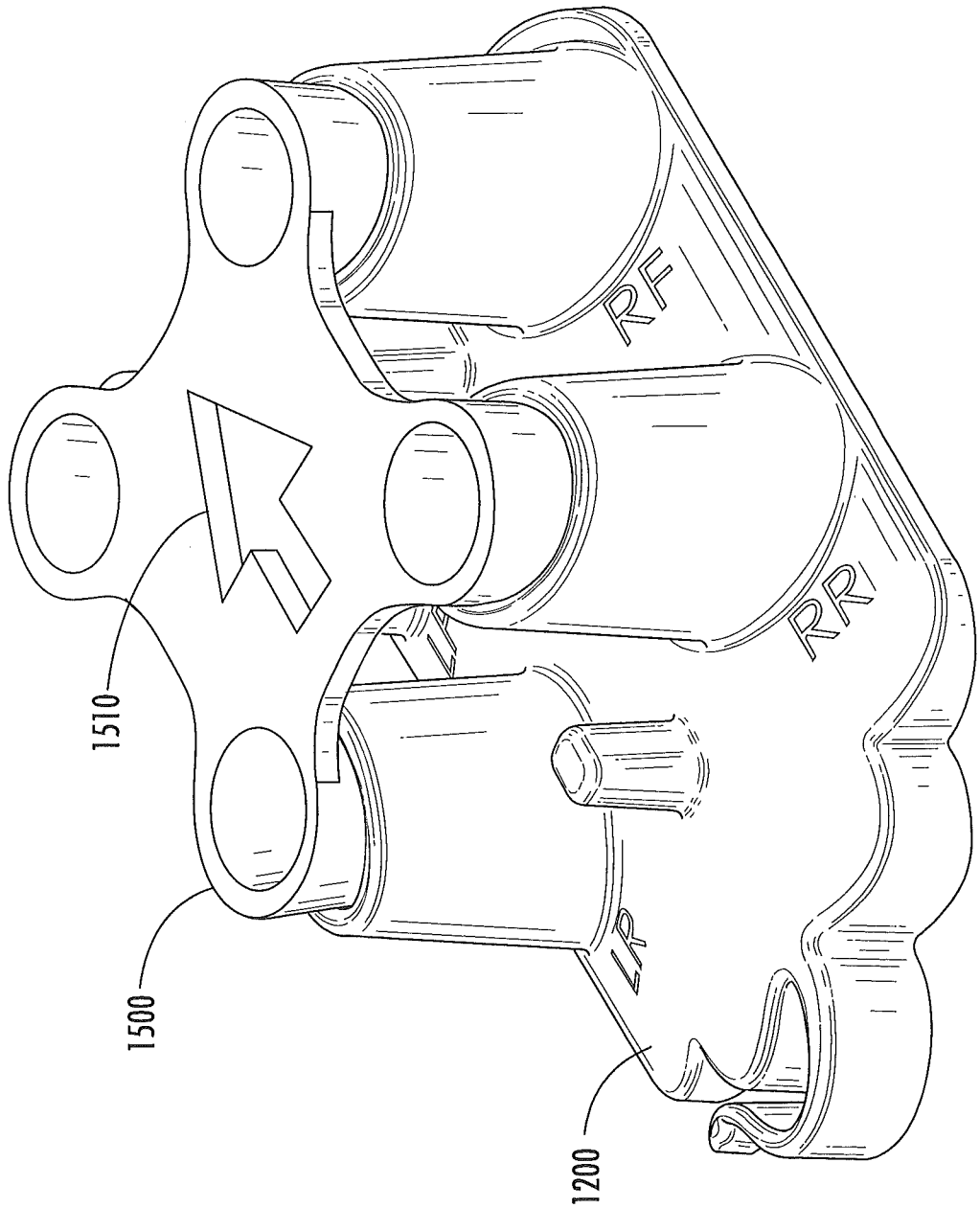

FIG. 25

INTERNATIONAL SEARCH REPORT

International application No
PCT/US2013/046760

A. CLASSIFICATION OF SUBJECT MATTER
INV. A01J5/013 G01N1/28
ADD. B01L3/00

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)
B01L G01N G02B A01J

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)
EPO-Internal, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 02/48681 A2 (MOLECULAR DIAGNOSTICS INC [US]) 20 June 2002 (2002-06-20) figure 1 page 7, line 6 - line 14 page 10, line 26 - line 30 page 8, line 12 - line 19 page 12, line 21 - line 26 -----	1-37
X	WO 03/009776 A2 (APPLIED BIOTECH INC [US]; WEYKER DANIEL C [US]; FUJII ALAN [US]; ZEIS) 6 February 2003 (2003-02-06) page 4, line 1 - line 26; figure 1 -----	1-37
X	US 2012/106811 A1 (CHEN STEPHEN L [US] ET AL) 3 May 2012 (2012-05-03) figures 1a,1b paragraph [0036] -----	1-37
	-/--	

[X] Further documents are listed in the continuation of Box C.

[X] See patent family annex.

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Date of the actual completion of the international search	Date of mailing of the international search report
11 October 2013	18/10/2013

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Mauritz, Jakob

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INTERNATIONAL SEARCH REPORT

International application No
PCT/US2013/046760

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	EP 2 264 424 A1 (SWITCH BVBA [BE]) 22 December 2010 (2010-12-22) figure 5a paragraph [0008] - paragraph [0009] -----	1-37
A	EP 1 380 345 A2 (KALTEK S R L [IT]) 14 January 2004 (2004-01-14) figure 1 -----	1-37

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INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/US2013/046760

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