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(54) Benævnelse: **FLYDECELLE MED INTEGRERET TØRSTOF**

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Flow cell with integrated dry substance

Description

The invention relates to a flow cell having a dry substance which is arranged within the flow cell in a cavity, wherein the flow cell has the features of Claim 1.

Microfluidic flow cells, as are increasingly being used as "mini-laboratories" for analysis and/or synthesis of fluids, in particular in diagnostics, contain reaction substances in liquid and/or solid form that are to be introduced into the flow cells during the production of the flow cells.

For the purpose of introducing a dry reagent, in one assembly step, in which the region, for example of a channel or a chamber, which is intended for receiving the dry reagent within the flow cell is still accessible, a reagent liquid to be dried, that is to say a carrier liquid with reagent dissolved or suspended therein, is brought to the respective location. Afterwards, the entire flow cell component, which has been wetted only partially with the reagent liquid, is to be subjected, prior to the further assembly thereof, to a drying process which, for the purpose of speeding it up, is commonly associated with a heat treatment or which, for the purpose of protecting the reagents and in consideration of stability and re-suspension properties, is realized as a freeze-drying process. Disadvantageously, the component, whose dimensions normally far exceed those of the region which alone is to be dried, takes up a lot of space in a drying chamber. Furthermore, the drying treatment can impair this flow cell component itself, in particular sensitive components fitted thereto. The dry substance which has formed can especially during the full assembly of the flow cell be subject to impairments by way of air contact, in particular air humidity, and welding heat or to the influence of adhesives used during the assembly, by way of which adhesives corresponding channel regions of a microfluidic flow cell are to be sealed off hermetically in many cases. A method for introducing a dry substance into a flow cell as has been described above is presented for example in EP 2 198 964 B1.

DE 10 2008 021 364 A1 presents a flow cell in which recesses in a substrate are closed off by a film cover. The film cover carries on the inner side dry reagents which face towards the cavities formed by the covered recesses.

A flow cell known from WO 2012/154306 A1 has a cavity which is accessible after a hatch or flap has been opened and which has in a base a depression for receiving a cuboidal support element. When the flow cell is operated, the hatch or flap is to be closed. A flow cell having the features of the preamble of Claim 1 is presented in each case in WO 2011/051735 A2 and WO 2008/134462 A1.

The invention is based on the object of providing a new microfluidic flow cell of the type mentioned in the introduction that has an integrated dry substance and that can be produced more easily than according to the prior art without impairments by the production surroundings of the dry substance or of other constituent parts of the flow cell.

Said object is achieved by the features of Claim 1.

It is advantageously possible for the formation of the dry substance through drying of a reagent liquid to be realized on a support element separated from the entirety of the rest of the flow cell, which support element serves only for receiving the dry substance and allows the introduction of the dry substance into the flow cell in a concluding assembly step. Impairments of flow cell components by way of the drying process and impairments of introduced dry reagent by way of further assembly steps at the flow cell are eliminated. The support element may have significantly smaller dimensions than the flow cell, wherein the dimensions of the support element are based on the size of the region supporting the dry reagent. Coatings promoting the adhesion of the dry substance to the support surface thereof may advantageously remain limited to the support surface of the support element, so that impairments of welded or adhesive connections by such coatings can be ruled out.

It goes without saying that the cavity may form a channel network for the transport, analysis and/or synthesis of a fluid. Multiple support elements, if appropriate with different dry substances, may be insertable into the flow cell.

In one embodiment of the invention, the cavity is delimited by a recess in a plate-shaped substrate and by a cover of film-type form that closes off the recess. The passage is formed in the substrate, which is thicker in comparison with the film-type cover.

It goes without saying that the passage is expediently led to an outer surface of the flow cell, so that the introduction of the dry substance into the flow cell can be realized during the production thereof in a last assembly step.

The support element is preferably designed in such a way that it can be connected to the flow cell in a detachable manner, and/or in a non-detachable manner, such that the cavity is closed off. Preferably, in terms of its shape, the passage is matched to the shape of the support element. Fluid tightness can be achieved in particular by welding-in and/or adhesive bonding in place, if appropriate also just by mechanical pushing of the support element into the passage.

Accordingly, the support element, at least in cross section, expediently completely fills the passage, wherein the support element and the passage, in cross section, preferably have a circular shape, which is advantageous in terms of manufacture.

In a further configuration of the invention, the support element tapers towards the cavity, while the passage narrows towards the cavity. In particular, it is thus possible for a sealed closure of the cavity in the form of a press fit to be achieved solely by mechanical pushing of the support element into the passage.

Preferably, the support element has a portion which projects outwardly from the flow cell, said portion being able to serve as a grip part for simplifying manual handling or automated fitting.

The projecting portion may engage over the flow cell on the outside in the manner of a collar, wherein, by way of the collar, additional sealing of the cavity can be achieved.

In a further embodiment, the support element can be screwed into the passage.

The support surface of the support element may be arranged so as to be flush with, or so as to be set back from, an adjacent wall surface of the cavity. Alternatively, the support element projects into the cavity from the adjacent wall surface.

The support surface expediently has a structuring, coating and/or surface modification which promotes the adhesion of the dry substance.

The support element and the support surface which supports the dry reagent preferably consists of plastic. Alternatively, the support surface may be formed by a separate surface component composed of glass, silicon, ceramic or metal that is connected to the rest of the support element, and be attached by means of welding or adhesive bonding. This is advantageous if, for the application of the dry reagent, surface properties which are realizable other than by a plastic surface or coating are necessary.

Possible dry reagents include salts, buffers, for example for cell lysis, magnetic and non-magnetic beads, enzymes, antibodies, DNA fragments, proteins and PCR reagents, or alternatively also cells.

The invention will be explained in more detail below on the basis of exemplary embodiments and the appended drawings, which relate to said exemplary embodiments and in which:

Figure 1 shows an illustration which explains the production of flow cells with an integrated dry substance according to the prior art,

Figure 2 shows an illustration which explains the production of a flow cell,

Figure 3 shows a detail illustration of the flow cell shown in Figure 2,

Figure 4 shows exemplary embodiments for the arrangement of a support surface of a support element within a cavity of a flow cell,

Figure 5 shows further exemplary embodiments for support elements,

Figure 6 shows exemplary embodiments for support surface areas of support elements,

Figure 7 shows an illustration explaining the application of a dry substance to support elements, and

Figure 8 shows an exemplary embodiment for a support element according to the invention.

A flow cell shown in part in Figure 1 comprises a plate-shaped substrate 1 with a recess 2 which is covered, so as to form a cavity 3, by a film 4 which is adhesively bonded and/or welded to the substrate. The cavity 3 is part of a channel network (otherwise not illustrated in Figure 1) of the flow cell, it in particular forming a channel region in which a for example antibody-containing dry reagent 5 adheres to a channel wall 6.

The dry reagent 5 originates from a reagent liquid 7 which, prior to the recess 2 being covered by the film 4, was introduced by dispensing into the recess 2, said recess forming a channel region or chamber region of the flow cell. For forming the dry reagent 5 from the reagent liquid 7, the entire substrate 1 was subjected to heat treatment and/or freeze-drying.

Figure 2 indicates a method for introducing a dry substance, in particular a dry reagent 5, into a flow cell, in which method the dry reagent 5 is applied to a separate support part 8. A cavity 3 in a flow cell, which cavity may for example be a region

of a channel 9 shown in Figure 3, has a passage opening 10 into which the support element 8 can be inserted, such that the cavity 3 is closed off in a fluid-tight manner, by way of a conical portion 11 having a support surface 13 for the dry reagent 5. After the fitting, the support surface 13 forms a part of the wall surface of the cavity 3. A fluid processed or transported in the cavity 3 can thus interact with the dry reagent, it being possible in particular for the dry reagent to be dissolved by the fluid and re-suspended. Furthermore, constituents of the fluid such as cells or analytes can interact with the dry reagent, and/or bond thereto, in that the fluid flows over the support surface, possibly multiple times with changing of the transport direction.

The support element 8 fitted into the passage opening 10 may be adhesively bonded or welded to the substrate. A portion 12 of the support element 8 that projects beyond the passage opening 10 on that side of the substrate 1 which faces away from the cavity 3 serves as a grip part which facilitates the fitting of the support element 8.

By contrast to the example relating to the prior art in Figure 1, for forming the dry reagent 5 from a reagent liquid 7, not the entire substrate 1, as in the example in figure 1, but merely the support element 8 needs to be subjected to drying, which saves space in a drying chamber. The main components of the flow cell, the substrate 1 and the film 4, are not subjected to any stress resulting from the drying process, and the dry substance 5 subsequently introduced into the flow cell is not subjected to any impairment due to finishing of the flow cell, with welding of substrate 1 and film 4, that occurs at a later stage.

As Figure 3 shows, multiple openings for receiving support elements 8, with possibly different dry reagents 5 arranged thereon, may be present in the channel 9. In the example in Figure 3, the meandering channel 9 serves for redissolution of the dry reagents 5 introduced by the support elements 8, through being washed over in an alternating manner.

The substrate 1 and the film 4 of the flow cell preferably consist of a plastic, in particular both of the same plastic, wherein for example PMMA, PC, PS, PEEK, PP, PE, COC and COP come into consideration for this purpose. The support element 8, too, is preferably a plastic part, which consists in particular of the same plastic as the substrate. The production of the substrate and the support elements from plastic is expediently realized by way of injection moulding.

As can be seen in Figure 4, the support surface 13, receiving the dry reagent 5, of the support element 8 may be flush with the adjacent wall surface 14 of the cavity 3 or set back from said wall surface. As per figure 4b, the support element 8 may also project, with the support surface 13, into the cavity 3. This may be advantageous for locally generating in a laminar flow, which is normally present in micro-channels, turbulence by way of abrupt changing of the channel cross section, and/or, for example for the purpose of speeding up and controlling a redissolution of the dry reagent, for achieving, through reduction of the channel cross section, an increase in the flow speed of the fluid in the channel region into which the support element 8 is introduced. Furthermore, the advantage that fitting and/or component tolerances can be compensated in the case of automated fitting of the support element 8 is obtained.

Figure 5 shows further embodiments for support elements 8, which may be formed to be cylindrical, as per figure 5a, and formed to be cylindrical with a collar 15 engaging behind the substrate 1, as per figure 5b.

Figure 5c shows an embodiment of a cylindrical support element 8 which has a collar 13 and which has an outer thread 16 which engages into an inner thread in the respective passage opening. Advantageously, in the latter embodiment, the support element 8 can be detached from the flow cell provided that no adhesive bonding or welding to the substrate 1 is realized in addition to the screw connection. The detachability may be advantageous if the dry reagent, after interacting with the fluid, is to be separated from the flow cell again and subjected to further analysis.

Figure 5e shows a support element 8 which can be detached from the flow cell and which has an extended grip part 17. The support element 8 can be pushed into the respective passage opening in the substrate 1 such that the cavity 3 is closed off in a fluid-tight manner.

The guidance of the support element 8 can be improved by way of an elevated edge 25, as per Figure 5f, on the substrate 1, the thickness of which is typically between 0.5 and 3 mm.

Figure 5d shows a support element 8 with a conical portion and with a collar 15 which projects from a passage opening and which is sealed off with respect to the substrate 1 by a ring seal 18.

The rotationally symmetrical support elements may have a marking, which makes it possible to introduce the support elements into the passage in a desired rotational position.

Figure 6 shows exemplary embodiments of support elements 8 with differently formed support surfaces 13, wherein Figure 6a has a support element with a depression 19 for receiving a dry reagent 5. In the exemplary embodiment of Figure 6b, a support surface 13 having a multiplicity of receiving depressions in the form of grooves 20 which are arranged in the manner of a cross and which have typical cross-sectional dimensions of 0.01 x 0.01 mm^2 to 1 x 1 mm^2 is formed for a dry reagent. This advantageously allows the surface area of the support surface 13 to be increased in a simple manner such that a larger quantity of dry reagent 5 can be received for the same dimensions of the support element 8 and/or the dry substance can dry in a more homogeneous manner than a large drop on a smooth support surface and/or the microstructure of the support surface 13 that is formed by the receiving depressions 20 can generate turbulence when flowed over by the fluid, which turbulence positively influences the redissolution behaviour. The grooves could alternatively also have the form of concentric circles. Figure 6c shows a receiving surface having a porous element 21 which is attached to the support surface by way of clamping, adhesive bonding or welding and in which a dry

substance can settle. Advantageously, the porous element 21 forms an increased surface area for receiving the dry reagent 5.

Figure 6d shows a support element with a treated support surface, wherein the treatment may for example be a wet-chemical treatment, a plasma treatment or a corona treatment. Alternatively, the treatment may lead, for example by means of plasma polymerization or by means of a PVD process, to a coating 22, for example a glass or metal coating.

A support component shown in Figure 6e is formed to have two parts, with a separate surface area component 26. The surface area component 26 forming the support surface area consists for example not, like preferably the rest of the support component, of a plastic, but of glass, silicon, metal or ceramic. If the functionalization, that is to say the application of the dry reagent to the support surface, requires such materials, as is the case for example with protein-based (for example antibody-based) or nucleic-acid based analysis technologies, then the use of said materials, which are often significantly more expensive than plastic, is advantageously limited merely to a surface area region, wherein dimensions of 0.5 x 0.5 mm to 5 x 5 mm and thicknesses between 0.1 and 1 mm are possible. The surface area component 26 may be fastened to the rest of the support component by means of clamping or by way of adhesive bonding or welding.

For application of the dry substance 5, a multiplicity of support elements 8 may be processed simultaneously in that the support elements 8, in step 7a, are arranged on a support tablet 24 which has a row of holes 23. In the following method step 7b, a layer 22 which improves the adhesion of a substance is produced on all the support surfaces 13 of the support elements 8 simultaneously. Here, the coating may also cover other surface area regions of the support element 8, which are not intended for the application of the dry reagent 5. In method steps 7c and 7d, a drying treatment is realized after application of a reagent liquid 7 to the layers 22 such that, on the layers 22, there settles the dry substance 5, which adheres thereto. Finally, in step 7e, the finished support elements 8, provided with a dry substance 5, may be removed for the purpose of processing.

Reference is now made to Figure 8, where a further exemplary embodiment for a support element 8 is shown.

The support element 8 has a support surface for a dry substance 5 that is formed by a membrane 27. Said membrane may be connected in one piece to the rest of the support element 8 or may be formed by a separate component which is connected to the rest of the support element, said separate component preferably consisting of the same plastic as the rest of the support element.

In the case of transparency of the membrane 27, which closes off at one end a passage opening 28 formed in the support element 8, it is possible to monitor the interaction of the fluid with the dry substance 5, as per Figure 8b, by optical detection. Furthermore, as per Figure 8c, it is possible to convexly or concavely shape the membrane 27 through pneumatic or mechanical application of pressure. In particular through alternating outward bulging and inward bulging of the membrane 27, the interaction between the dry substance and the fluid can be stimulated and both improved re-suspension of dry substances and improved depositing of constituents of the fluid onto dry substances, for example in the case of antibodies, can be achieved.

PATENTKRAV

1. Mikroflydende flydecelle omfattende et tørstof (5), der er arrangeret indeni flydecellen i et hulrum (3) og som sørger for integration med en væske placeret i hulrummet (3), hvor hulrummet (3) er afgrænset af en udsparing (2) i stoffet (1) og et dække (4), der lukker af for udsparingen (2), og hvor flydecellen omfatter et separat støtteelement (8) med en støtteflade (13) for tørstoffet (5), hvilken flade er tilvejebragt for et arrangement nær ved hulrummet (3), hvor en udadtil åben passage (10), der leder til en ydre flade af flydecellen åbner op indtil udsparingen (3), og at det separate støtteelement (8) er i stand til at blive indført i passagen (10) fra ydersiden, således at hulrummet (3) er aflukket, og således at støttefladen (13) er arrangeret nær ved hulrummet (3), **kendetegnet ved, at** støttefladen (13) for tørstoffet (5) er dannet af en membran (27), der aflukker en passageåbning (28) i støtteelementet (8) imod hulrummet (3).

2. Flydecelle ifølge krav 1, **kendetegnet ved**, at hulrummet (3) danner et kanalnetværk (9) for transport, analyse og/eller syntese af en væske, og ved at hvis hensigtsmæssigt flere af sådanne støtteelementer (8) er i stand til at blive indført i en flerhed af passager (10).

3. Flydecelle ifølge krav 1 eller 2, **kendetegnet ved**, at udsparingen (2) er udformet i et pladeformet stof (1) og er aflukket af en filmtype dække (4), og ved at passagen (10) er udformet i det pladeformede stof.

4. Flydecelle ifølge krav 3, **kendetegnet ved**, at passagen (10) ledes til en pladeoverflade af det pladeformede stof (1).

5. Flydecelle ifølge ethvert af kravene 1-4, **kendetegnet ved**, at støtteelementet (8) er i stand til at blive forbundet til flydecellen på en aftagelig måde, og/eller på en ikke-aftagelig måde, således at hulrummet (3) afdækkes på væsketæt måde.

6. Flydecelle ifølge ethvert af kravene 1-5, **kendetegnet ved**, at støtteelementet (8) i det mindste i tværsnit komplet fylder passagen (10), hvor fortrinsvis støtteelementet

(8) og passagen (10) har cirkulært tværsnit og støtteelementet (8) snævrer sig ind imod hulrummet (3) og passagen (10) bliver snæver imod hulrummet.

7. Flydecelle ifølge ethvert af kravene 1-6, **kendetegnet ved**, at støtteelementet (8) har en del (12; 15; 17) der rager udad fra flydecellen, hvor hvis hensigtsmæssigt delen griber ind bagved flydecellen på ydersiden på en måde som en krave (15).

8. Flydecelle ifølge ethvert af kravene 1-7, **kendetegnet ved**, at støttefladen (13) af støtteelementet (8) er arrangeret således at det flugter med eller således at det rager bagud fra en nærliggende vægflade (14) af hulrummet (3) eller ved at støtteelementet (8) rager ind i hulrummet (3) fra den nærliggende vægflade (14) af hulrummet.

9. Flydecelle ifølge ethvert af kravene 1-8, **kendetegnet ved**, at det tørre stof klæber til støttefladen (13), og at støttefladen (13) fortrinsvis har en strukturering (19-21), belægning (22) og/eller overflademodifikationer der fremmer vedhæftningen.

10. Flydecelle ifølge ethvert af kravene 1-9, **kendetegnet ved**, at støttefladen (13) for tørstoffet (5) er dannet af en separat komponent (21; 26). der er forbundet til resten af støtteelementet (8), og som materialemæssigt adskiller sig fra resten af støtteelementet (8).

11. Flydecelle ifølge ethvert af kravene 1-10, **kendetegnet ved**, membranen (27) er transparent.

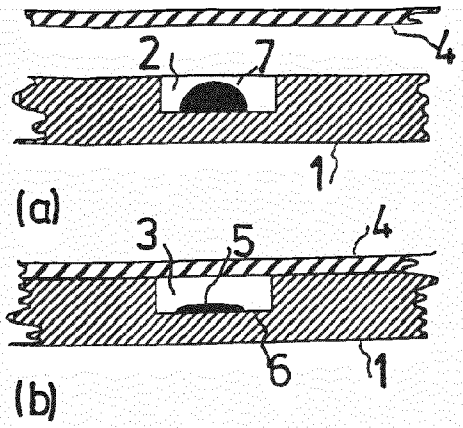

FIG.1
(PRIOR ART)

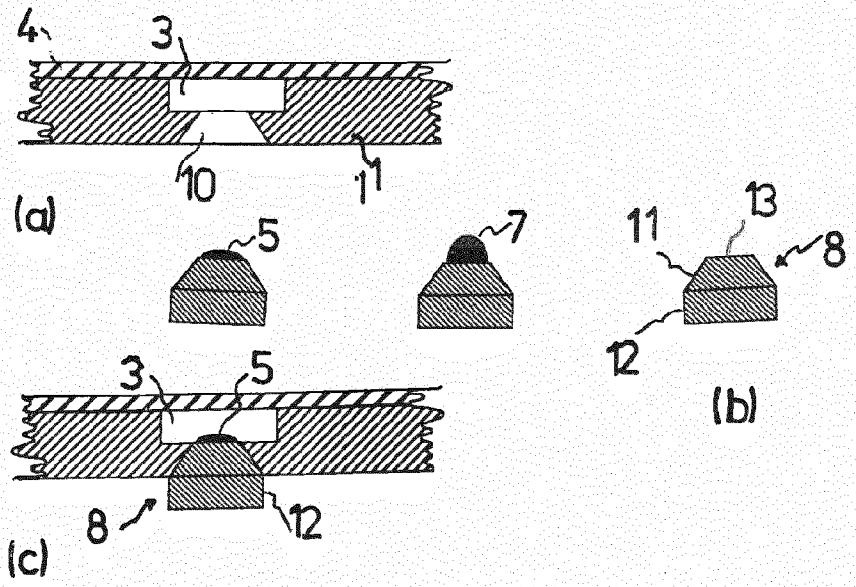

FIG.2

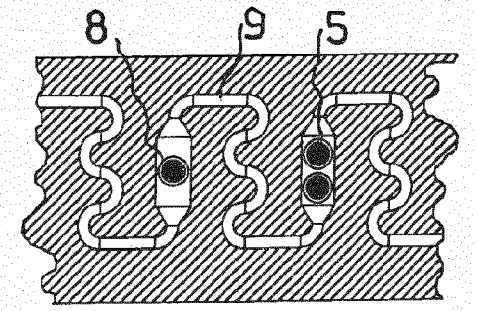

FIG.3

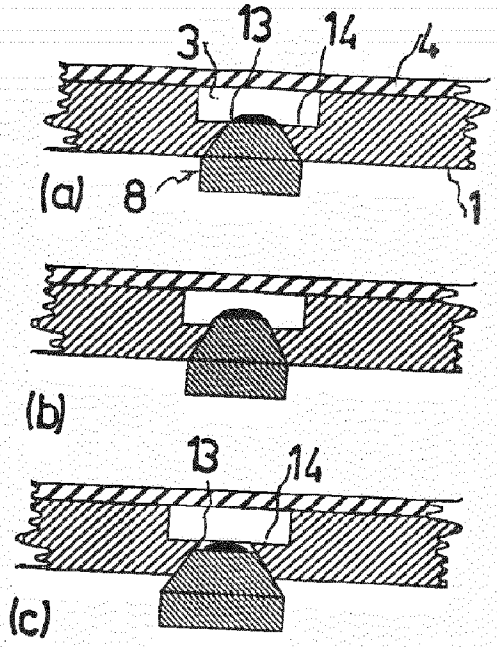

FIG. 4

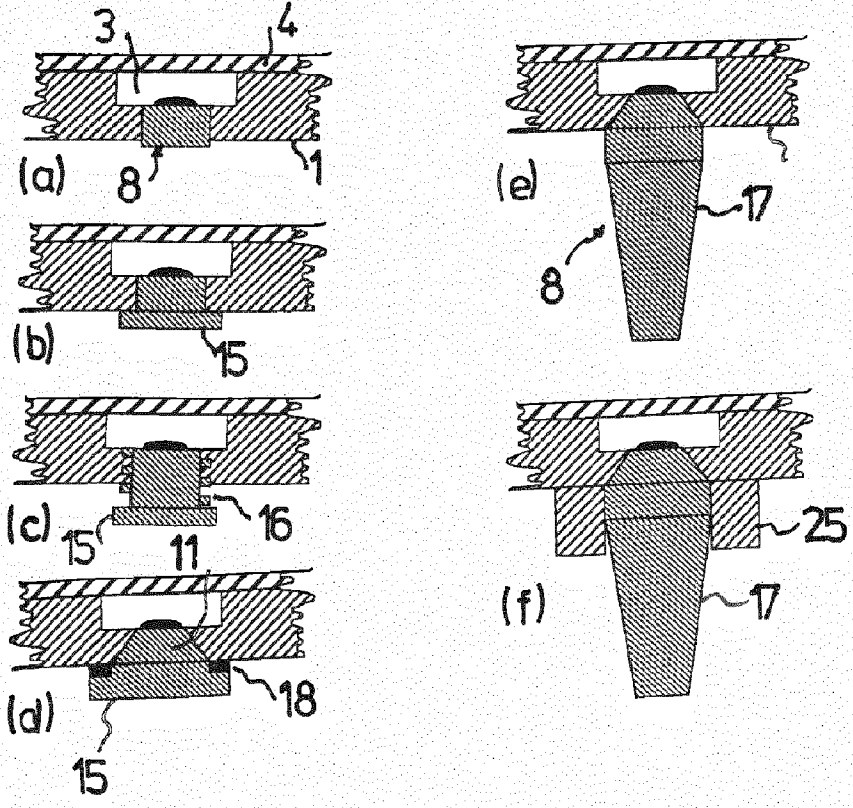

FIG. 5

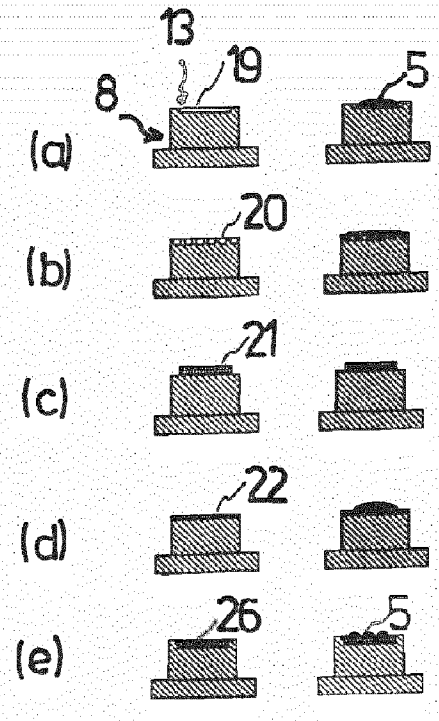

FIG.6

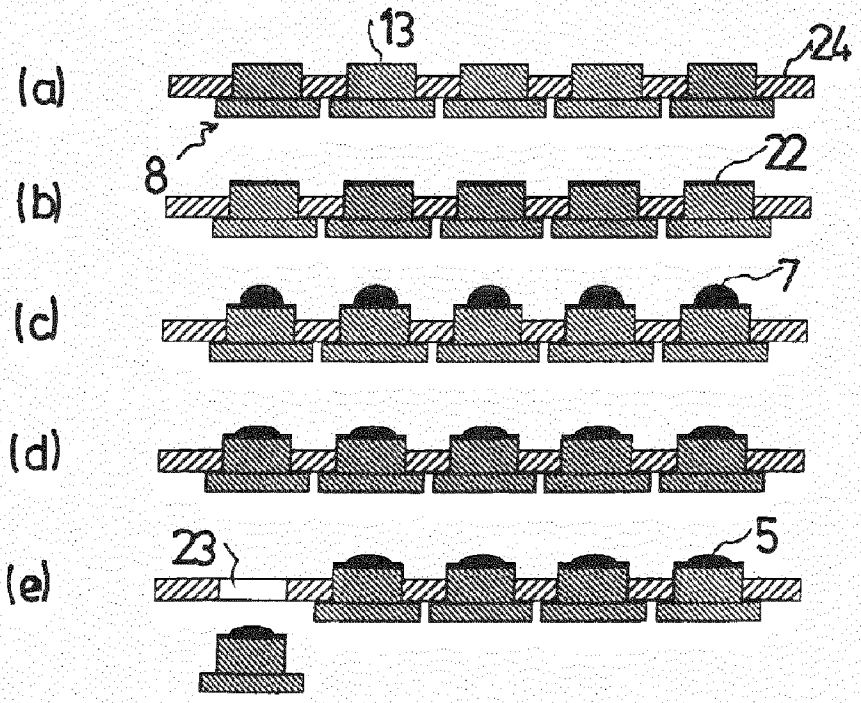

FIG.7

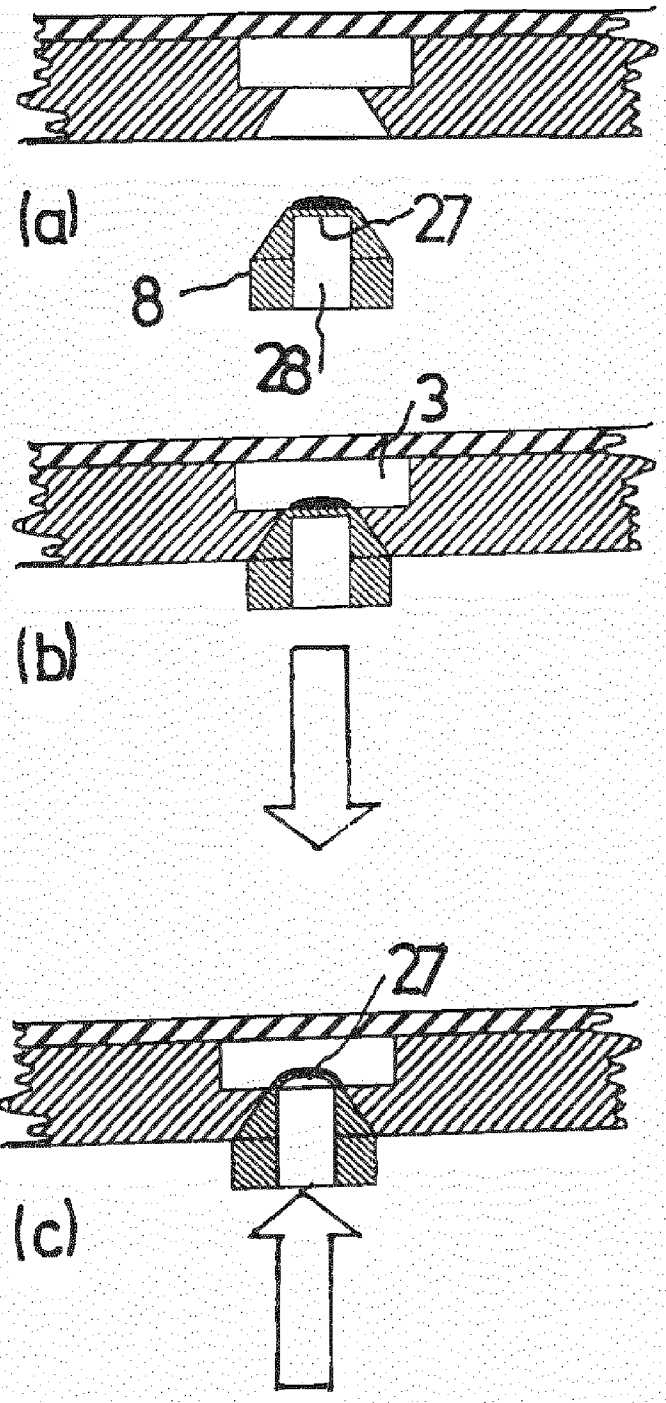

FIG.8