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(54) **MACHINE AND METHOD FOR LYSING A MICRO-ORGANISM**

(57) The present invention relates to a machine (2) for lysing a micro-organism with a chip (10) which features a fluidic network comprising ports, and a lysing chamber (25) for the lysing of the sample;

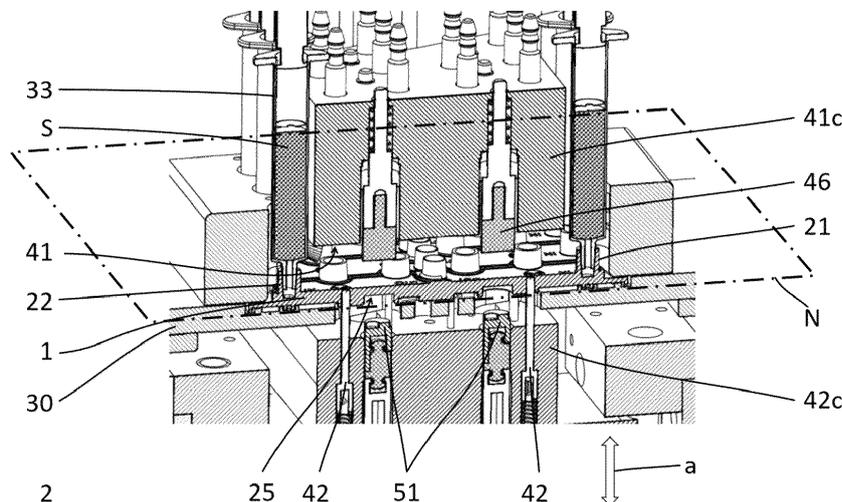
- the chip (10) being planar, extending in a chip plane (P);
- the machine comprising:
 - a nesting platform (30) mounted on a frame (60), and comprising a nesting plate (37) defining a nesting plane (N), so that the nesting platform (30) so that the chip plane

(P) is parallel to the nesting plane (N),

- fluidic actuators linked to the frame (60), and configured to actuate the fluidic network;
- a pestle (51) mounted on the frame (60), for applying pressure to the lysing chamber (25).

The pestle (51) or one of the fluidic actuators faces a first side of the nesting plane (N), and another one of the fluidic actuators is oriented differently.

[Fig. 9]



EP 4 541 460 A1

Description**FIELD OF INVENTION**

[0001] The present invention relates to a machine and a method for lysing a micro-organism.

BACKGROUND OF INVENTION

[0002] It is known from the document WO2023131714, in the name of the Applicant, a chip for micro-organism lysis, a mechanical system operating with such chip and methods of micro-organism lysing. The chip for micro-organism mechanical lysis comprises a lysing chamber, and is used in order to prepare a sample and collect the Deoxyribonucleic acid (DNA) of micro-organisms contained in said sample.

[0003] The lysing of a sample comprises the following steps:

- introducing the sample into the lysing chamber, through an inlet sample port, the sample, e.g., food diluted in a solution, containing micro-organisms;
- rinse the lysing chamber in order to remove unwanted elements of the sample, but keep the micro-organisms, thanks to a dedicated filter;
- apply pressure on the lysing chamber, e.g., with a pestle, in order to lyse the micro-organisms so they release their DNA;
- elude the released DNA towards a sample outlet port.

[0004] Once the DNA of the micro-organisms is ready to be collected in the sample outlet port, it is available for being collected with a pipette and sent to a polymerase chain reaction (PCR) process in order to detect, e.g., whether the micro-organisms contain infectious agents.

[0005] Nevertheless, the systems for operating known chips can be cumbersome, whereas the equipment for laboratory use are expected to be compact and space-saving.

[0006] In an embodiment of WO 2023/131714 (illustrated figure 8a), a plate comprises several chips arranged in an array, in order to improve the efficiency of a laboratory when there is a need to conduct several tests. This embodiment allows a laboratory technician to collect all the samples simultaneously, with a standard multi-pipette.

[0007] It is further difficult to design a machine for operating a plate comprising several chips, because the number of couplers needed for actuating each chip is by consequence multiplied by the number of chips.

[0008] In the application EP22306591.3 of October 20, 2022, also in the name of the Applicant, an improved chip for lysing comprises several ports in order to avoid cross contamination of the ports during the different steps of the process. On each duct linking a port to the lysing chamber, a valve is activable in order to open or to close said

duct, according to the step of the process. E.g., during the elution phase, only the duct linking the elution port to the lysing chamber and the duct linking the lysing chamber to the sample outlet port are open.

[0009] The aforementioned drawback is exacerbated in the case of chips with valves, because:

- it needs supplementary pipe connectors to plug on the extra ports, so the layout of a chip is further obstructed; and
- the machine for operating the plate of chips needs further actuators, as pins, configured to actuate the valves, so the machine is more complex to design and to operate.

[0010] Thus, there is a need to improve the systems for operating a chip for micro-organism lysis.

SUMMARY

[0011] The aim of the invention is to overcome the drawbacks of the prior art, in particular by providing a machine for operating a chip for micro-organism lysis, which is compact and well suited for a laboratory use.

[0012] Another aim of the invention is to provide a machine further suitable for operating chips with valves, and preferably plates of chips with valves.

[0013] To this end, it has been developed a machine for micro-organism lysing with a chip,

the chip featuring:

- a fluidic network comprising ports including a sample inlet port for injecting the sample in the chip, and a sample outlet port for collecting a lysed sample; and
- a lysing chamber for the lysing of the sample;
- the chip being globally planar and extending in a chip plane;

the machine comprising:

- a frame;
- a nesting platform mounted on the frame, and comprising a generally planar support defining a nesting plane, the nesting platform being configured to maintain the chip so that the chip plane is parallel to the nesting plane,
- fluidic actuators linked to the frame, and configured to actuate the fluidic network of the chip
- a pestle mounted on the frame, and configured to apply pressure to the lysing chamber.

[0014] According to the invention:

- the pestle or one of the fluidic actuators is oriented in face of a first side of the nesting plane, and
- another one of the fluidic actuators is oriented differ-

ently.

[0015] This way, the pestle and/or the fluidic actuators of the machine can be distributed around the chip, both above and under it, and even on its side. This new degree of freedom allows the design of more compact machines, presenting a reduced layout on a laboratory bench.

[0016] Preferably, the nesting platform comprises several bays each configured for receiving one chip, the planar support of each bay is coplanar with the nesting plane, and the bays are arranged aligned with an axis and regularly spaced apart so that:

- the sample outlet port of each chip coincides with the axis, and
- the sample outlet ports of two adjacent chips are spaced apart with a pitch; and

the machine comprises fluidic actuators and a pestle for each bay.

[0017] This way, the machine is compatible with plates comprising several chips, which are sought when there is a need to improve the productivity of the machine or of the laboratory.

[0018] The pitch is preferably the pitch of a standard multi-pipette, and is generally comprised between 8mm to 14mm, e.g., 9mm.

[0019] In order to simplify the kinematics of the machine, a fluidic actuator is axially mobile according to a moving direction between:

- a passive position where the fluidic actuator is distant from the chip and does not interact with the fluidic network, and
- an active position where the fluidic actuator is contact with the chip and interacts with the fluidic network.

[0020] Advantageously, the fluidic actuator is linked to the frame with an elastic compensation in the moving direction. This compensation prevents jams, and protects the chip, during an automatic processing of the chip. This compensation also allows to calibrate the force which is applied on the chip by the fluidic actuator.

[0021] If the chip is of the type with at least one valve, it comprises a valve configured to selectively open and close a duct of the fluidic network, and

- the fluidic actuators comprise a pin mobile between
 - a passive position wherein the pin is distant from the valve, which is in flowing position, and
 - an active position wherein the pin is actuating the valve, which is in closed position.

[0022] Preferably, the pin is mechanically driven between its passive position and its active position by a driver, through a coupler as a cable or a camshaft. The

coupler converts the direction of movement of the pin into another direction, preferably orthogonal to the direction of movement of the pin. The use of a coupler allows to place a driver of the pin, as a motor, remotely from the pin.

5 This provides a supplementary degree of freedom for designing the machine. Moreover, it allows to use an electrical actuator and avoid pneumatics or hydraulics actuators, which is preferable for a laboratory use.

10 **[0023]** In order to improve the compacity of a machine designed to operate a plate of chips, a collector links the driver to several couplers of the pins, so that a single driver enables the simultaneous processing of several chips.

15 **[0024]** In order to improve the efficiency of the lysing, the pestle is linked to the frame by a ball and socket joint, as a cardan joint.

[0025] Preferably, the chip comprises a valve configured to selectively open and close a duct of the fluidic network and the fluidic actuators comprise pipe connectors configured to plug with the ports of the chip. A pipe connectors carrier comprises all the pipe connectors, faces an upper side of the nesting plane; and is axially mobile between:

- 20
- 25 - a connected position wherein the pipe connectors plug with the ports, and
 - a disconnected position wherein the pipe connectors are distant from the chip.

30 **[0026]** This way, a single actuator or motor is needed to actuate all the pipe connectors.

[0027] When the pipe connectors face an upper side of the nesting plane, a pestles carrier comprises the pestle, faces a lower side of the nesting plane and is mobile between:

- 35
- a grinding position wherein the pestle can apply pressure to the lysing chamber, and
 - 40 - a retracted position wherein the pestle is distant from the chip.

[0028] This way, a single actuator or motor is needed to actuate all the pipe connectors This allows to ease the design of the machine, by grouping together the fluidic devices and the piping of the pipe connectors. Moreover, the pressure applied by the pestle during the lysing will tend to reinforce the plugging of the pipe connectors onto the ports, as well as counterbalance the forces applied to the chip, thus limiting its deformation.

50 **[0029]** In that case, a pins carrier which comprises the pin, faces a lower side of the nesting plane and is mobile between:

- 55
- a selecting position wherein the pin can actuate the valve, and
 - a distant position wherein the pins carrier is distant from the chip.

[0030] The distant position allows the movability of the nesting platform, between:

- a processing position wherein the chip can be processed by the fluidic actuators and the pestle, and
- a loading position wherein the nesting platform is accessible to an operator placing a chip on the nesting platform. The loading position facilitates the loading and unloading of the chip into the machine.

[0031] A convenient way to prepare the sample and load them into the chips without polluting the machine or risking any cross contamination is to use syringes. In that case, the machine comprises:

- a gantry holding a syringe pusher, configured to push a syringe containing the sample and plugged to the sample inlet port, and mobile between at least:
 - a waiting position wherein the gantry is away from the nesting platform so as the syringe pusher is distant from the syringe, and
 - a discharge position wherein the gantry is close from the nesting platform, so as the syringe pusher has pushed the syringe in order to load the sample into the chip.

[0032] The connector carrier further comprises an ejector configured to apply a pressure on the chip towards the nesting platform when the connector carrier moves from the connected position towards the disconnected position, in order to help the unplugging of the pipe connectors from the ports.

[0033] Preferably, the ejector and the pestle are face to face, in order to limit the deformation of the plate under the pressure applied by the pestle during the lysing.

[0034] Advantageously, the machine comprises a four-way fluidic distributor comprising:

- a first inlet intended to be linked to a rinsing solution tank,
- a second inlet intended to be linked to an elution solution tank,
- a third inlet intended to be linked to a compressed air source, and
- an outlet intended to be linked to an elution port of the chip through a tubing,

wherein:

- during the rinsing step:
 - a rinsing pump is configured to deliver a predefined rinsing solution quantity from the first inlet to the outlet, and
 - a compressor is configured to deliver compressed air at a first pressure from the third inlet

to outlet,

so that the predefined rinsing solution quantity can be pushed through the tubing by the predefined compressed air quantity, and

- during the elution step:
 - an elution pump is configured to deliver a predefined elution solution quantity from the second inlet to the outlet, and
 - the compressor is configured to deliver compressed air at a second pressure from the third inlet to outlet,

so that the predefined elution solution quantity can be pushed through the tubing by the second predefined compressed air quantity.

[0035] This specific design allows to manage precisely the amounts of solutions delivered to the chip during the different phases of the cycle, as well as using simple and standard pumps for delivering said solutions.

[0036] The invention also relates to a chip designed for receiving a sample to be lysed and to be processed by the afore-mentioned machine, and featuring:

- a fluidic network comprising at least a first fluidic interface and a second fluidic interface, as ports or valves,
- a lysing chamber access hole, configured for the pestle can apply pressure on the lysing chamber.

[0037] According to the invention, the lysing chamber access hole or the first fluidic interface is placed on a lower side of the chip plane, and the second fluidic interface is placed differently.

[0038] This disposition of the pestle and fluidic interfaces allows the specific disposition of the pestle and fluidic actuators of the machine, thus providing a compact machine.

[0039] The invention also relates to a method of lysing micro-organisms by operating the afore-mentioned machine, comprising the steps of:

- displacing the nesting platform:
 - from a loading position wherein the nesting platform is accessible to an operator placing a chip on the nesting platform;
 - towards a processing position wherein the fluidic actuators face the chip;
- plugging the pipe connectors to their respective ports;
- loading the sample into the lysing chamber of the chip;
- rinsing the sample;
- lysing the sample with the pestle;
- eluding the sample;

- disconnecting the pipe connectors;
- displacing the nesting platform from the processing position towards the loading position.

[0040] These steps provide a user-friendly and efficient method. 5

[0041] Preferably, the step of bringing the gantry from the discharge position to the waiting position is conducted after disconnecting the pipe connectors. This way, the gantry helps maintaining the chip during the disconnection of the pipe connectors, thus preventing any deformation of the chip, or spillage. 10

BRIEF DESCRIPTION OF THE DRAWINGS

[0042]

[Fig. 1] is a schematic illustrating different phases of the lysing of micro-organisms, with the use of a chip according to the invention. Figure 1 reminds the main components and main phases of a mechanical lysing of a sample using a chip. 20

[Fig.2] is a top view of a first embodiment of a plate comprising chips of figure 1 aligned with an axis and regularly spaced apart with a first pitch. 25

[Fig.3] is a top view of a preferred embodiment of a plate comprising chips of figure 1, comprising chips aligned with an axis, disposed in a staggered manner, and regularly spaced apart with a second pitch. 30

[Fig.4] is a view in perspective, from above, of the embodiment of figure 3 illustrating frangible lines between the chips. 35

[Fig.5] is a view in perspective, from below, of the embodiment of figure 3, illustrating locating elements of the chips and access holes for actuating valves of the chips. 40

[Fig.6] is a top view of a chip separated from the plate of the preferred embodiment.

[Fig.7] is a cross section of a side view of the chip of figure 6, illustrating conic shapes of fluidic interfaces and access holes of the chip. 45

[Fig. 8] is a partial view, in perspective, of a machine according to the invention, displaying the main sub-assemblies of the machine. 50

[Fig. 9] is a detailed view of a cross section of the machine, illustrating actuators disposed on each side of a plate of chips, during the lysing. 55

[Fig. 10] is a schematic profile view of the machine, illustrating the relative positions of the nesting plat-

form, a gantry and a connector carrier.

[Fig. 11] is a perspective view, from below, of the connector carrier.

[Fig. 12] is a schematic top view illustrating the fluidic connection of an elution port of the chip.

[Fig. 13] is a partial cross section illustrating the fluidic connection of an elution port of the chip, using capillary tubes for the delivery of a rinsing solution and an elution solution.

[Fig. 14] is a detailed cross section, in perspective, of a pin carrier and a pestle carrier. 15

[Fig. 15] is a schematic based on figure 16, illustrating the kinematics of the pin carrier and the pestle carrier.

[Fig. 16] is a detailed cross section, in perspective, of the driving of pins, from a fixed driver to the movable pin carrier.

[Fig. 17] is a detailed cross section, in perspective, of the pin carrier which is traversed by pestles from the pestle carrier. 25

[Fig. 18] is a detailed view, in perspective, of a pestle and its mounting. 30

[Fig. 19] is a perspective view of a nesting platform, ready to receive the plate of figures 5 to 7.

[Fig. 20] is a perspective view of the nesting platform, holding the plate in position. 35

[Fig. 21] is a perspective view of the nesting platform, syringes containing samples to be tested being plugged onto the plate. 40

DETAILED DESCRIPTION

[0043] The invention relates to a machine for processing a chip (10) for the mechanical lysing of a sample, the chip (10) being globally planar and defining a chip plane (P). The machine comprises actuators (40), configured to process the chip (10), and which are disposed in various direction around the chip (10), e.g., above and below the chip plane (P). 50

[0044] The structure and functioning of such a chip (10) are described below.

[0045] Figure 1 illustrates a single chip (10) during different steps of a mechanical lysing of a sample, so that a terminology of the following description will be better understood. 55

[0046] A chip (10) comprises at least a sample inlet port (21), a sample outlet port (22), and a lysing chamber (25).

A loading duct (d1) links the sample inlet port (21) to the lysing chamber (25), and an elution duct (d2) links the lysing chamber (25) to the sample outlet port (22).

[0047] Preferably, the chip (10) further comprises an elution port (23) and a discharge port (24). A supplementary duct (d3) links the elution port (23) to the lysing chamber (25) and a discharge duct (d4) links the lysing chamber (25) to the discharge port (24).

[0048] When the chip (10) comprises four ports (21, 22, 23, 24), each duct (d1, d2, d3, d4) comprises a valve (26, 27, 28, 29) configured to selectively open and close its respective duct (d1, d2, d3, d4), thus allowing to manage the fluid distribution among the ducts (d1, d2, d3, d4) of the fluidic network.

[0049] The first panel (I) illustrates the loading of the sample into the lysing chamber (25), as illustrated by the first arrow (i). A sample container (e.g., a syringe) is connected to the sample inlet port (21), and loads the sample into the lysing chamber (25).

[0050] Any extra fluid can be evacuated by the discharge port (24) as illustrated by a second arrow (o), while the solid material remains in the lysing chamber (25), which contains a filter for separating macroscopic materials as food remains, from microscopic materials as the sought micro-organisms. Further details of this filtration can be found in document WO 2023/131714, e.g., pages 8 to 10, or page 15.

[0051] During this step of loading, a second valve (27) of the elution duct (d2) and a third valve (28) of the supplementary duct (d3) are closed, as illustrated by the "X" symbol, to prevent the extra fluid to contaminate the sample outlet port (22) and the elution port (23).

[0052] The second panel (II) illustrates the rising of the sample, during which the macroscopical material of the sample are discharged from the lysing chamber (25) in order to keep only the sought micro-organisms for the next step of lysing.

[0053] During the rinsing step, a rinsing solution is loaded through the elution port (23), as illustrated by a first arrow (i), the solution drags the macroscopic material, then leaves the chip (10) through the discharge port (24), as illustrated by the second arrow (o).

[0054] During the rinsing step, a first valve (26) of the loading duct (d1) and the second valve (27) are closed, as illustrated by the "X" symbol, thus guiding the elution solution to the discharge port (24).

[0055] The third panel (III) illustrates the lysing step, during which pressure is applied on the lysing chamber (25), as illustrated by a third arrow (p). Given the pressure can lead to a slight oozing from the sample, a valve remains open so that any exuded fluid can go in direction of a duct. Preferably, the third valve (28) remains open so that the exuded fluid goes in direction of the elution duct (d3), while the three other valves (26, 27, 29) are closed, as illustrated by the "X" symbol. Any exuded fluid contained in the elution duct (d3) will be re-collected by the elution solution during the elution step, and will be carried towards the sample outlet port (22). This allows to not

lose any sample material.

[0056] The pressure applied on the micro-organisms breaks their cells, thus releasing the DNA of the micro-organisms.

[0057] The fourth panel (IV) illustrates the elution step, during which the released DNA is brought to the sample outlet port (22), by an elution solution coming from the elution port (23).

[0058] During this step, the first valve (26) and the fourth valve (29) are closed, as illustrated by the "X" symbol.

[0059] The elution is a critical step of the whole process, as the quantity of elution solution sent into the chip (10) is of a few microliters only, and because the DNA tends to stick to the internal walls of the elution duct (d2).

[0060] Even if it not illustrated, the chip (10) can be of the type without valves (26, 27, 28, 29), as disclosed in WO 2023/131714. In this case:

- the sample inlet port (21) is also used as the discharge port (24) during the rinsing step; and as the elution port (23) during the elution step;
- the sample outlet port (22) is also used as the discharge port (24) during the loading step, and as the elution port (23) during the rinsing step.

[0061] Because managing pressures, quantities and fluids delivery are very delicate in the field of micro-fluidics, the fluidic network is the shortest possible. This is mostly important for the elution duct (d2), used when the DNA is already released from the micro-organisms and must be conveyed to the elution port (23).

[0062] The length of the elution duct (d2) is less than 25mm, preferably less than 20mm. Best results of elution were obtained with elution ducts of length from 10mm to 15mm included.

[0063] In reference to figure 2, a plate (1) of chips (10) comprises an arrangement of the chips (10) is an array, so that each sample outlet port (22) is disposed on an axis (10a). The sample outlet ports (22) are distant the one from the other by a first pitch (P1) corresponding to the pitch of a standard multi-pipette, e.g., 9mm, the same pitch separating each chip (10). Given the elution ducts (d2) are the same for each chip (10) of the plate (1), the behavior of each chip (10) is equivalent from the one to the other, and each chip (10) provides result of equivalent level. False negative results and erratic behavior, between chips (10) of a same plate (1), are avoided.

[0064] To do so, functional parts of a chip (10) overlap the rectangle area corresponding of an adjacent chip (10), in a non-functional area said adjacent chip (10). To that end, the chip (10) comprises, on a first lateral side (10 11), a protrusion (13) resulting from the projection of a functional part, as a port, and a second lateral side (10 12), opposite to the first lateral side (10 11), comprises a concavity (14) matching said protrusion (13).

[0065] As a result, two adjacent chips (10) engage together in the manner of a tile pavement, or as puzzle

parts. This leads to a decrease of the first pitch (P1), which can be as small as e.g., 14mm: this value corresponds to the widest pitch of common multi-pipettes.

[0066] On the illustrated embodiment, the chips (10) comprise four ports (21, 22, 23, 24): given the high number of ports to place on the layout of the chip (10), each chip (10) thus comprises two protrusions (13) and two concavities (14), placed on its lateral sides.

[0067] The first lateral protrusion (13a) and the second lateral protrusion (13b) can be on the same lateral side. Preferably, the first lateral protrusion (13a) is on a first lateral side (10 11) and the second lateral protrusion (13b) is on a second lateral side (10 12), opposite the first lateral side (10 11). This allows a better balance of the chip (10) when pipe connectors (41) are plugged into the ports (21, 22, 23, 24) of the chip (10), especially if done by an automatic machine:

[0068] the two ports are distributed on each lateral side, so the forces applied by the pipe connectors on said ports tend to compensate themselves relating to a longitudinal direction (L). The chip (10) is better balanced, avoiding its flipping around a transversal direction (T).

[0069] Figure 3 illustrates a preferred embodiment of the invention, wherein the plate (1) comprises two rows of chips (10), each aligned with the axis (10a), but with a first row (R1) disposed on a first side in relation to the axis (10a), and a second row (R2) disposed on a second side in relation to the axis (10a), the chips (10) of the two rows (R1, R2) being disposed head-to-head, in a staggered manner.

[0070] The staggered disposition of the chips (10) allows to obtain a plate (1) wherein a second pitch (P2) is further reduced in comparison to the first embodiment: the second pitch (P2) equals the first pitch (P1) divided by two.

[0071] The pitch is measured between two consecutive chips (10) in the direction of the axis (10a):

- in the first embodiment, the first pitch (P1) is measured between two adjacent chips (10);
- in the preferred embodiment, the second pitch (P2) is measured between chips (10) placed alternatively on the first row (R1) and on the second row (R2).

[0072] In this embodiment, the chips (10) comprise a transversal protrusion (13c), on a transversal side (10 t), and resulting from the projection, in a top view, of the sample outlet port (22). The transversal side (10 t) also comprises a transversal concavity (14c) allowing space for the transversal protrusion (13c) of a facing chip (10).

[0073] It must be understood that the engagement of a protrusion (13) and a concavity (14) does not need to be adjusted or to be tight: the main feature is to make the most of the non-functional area of a chip (10). A concavity (14) may leave a gap around its matching protrusion (13).

[0074] Preferably, all the chips (10) of the plate (1) are identical. That leads to facing chips (10) being symme-

trical, according to a central symmetry around a middle point between their respective sample outlet ports (22).

[0075] Figure 4 illustrates the preferred embodiment from an above perspective point of view. This figure shows frangible lines (S) linking the chips (10) of the plate (1). A frangible line (S) allows to separate manually the chips (10) from the plate (1). This may be needed if a test does not require to use all the chips (10) of the plate (1). The laboratory technician can take only the chips (10) he needs from the plate (1), and keep the remaining chips (10) for a later use.

[0076] The frangible line (S) can be obtained by any appropriate mean. It can be a score or a notch made for example in a connecting panel (15) of the plate (1). It can be a dotted line obtained by perforation. In the case of a multi-material connecting panel (15), the connecting panel (15) can be mostly made of a first material whereas the frangible line (S) can be made of a second material, weaker than the first material.

[0077] When the chips (10) of a plate (1) are all identical and frangible, this allows to use any separated chip (10) indifferently, regardless of the position it had on the plate (1). These interchangeable chips (10) are more convenient to use.

[0078] Figure 4 and 5 show that the plate (1) globally extends in a plane (P) which defines an upper face (10u) of the chip (10) and a lower face (10l) opposite to the upper face (10u).

[0079] Fluidic interfaces (11) of the chips (10) are preferably disposed on each side of the plane (P). This way, fluidic actuators configured to actuate the fluidic interfaces (11) are lesser a constraint when designing the layout of the chips (10): both sides of the chip (10) can be used. The layout of the chip (10), thus the plate (1), can be further reduced.

[0080] Preferably, all the ports (21, 22, 23, 24) of the chip (10) are disposed on the upper face (10u), wherein valves access holes (12) are all disposed on the lower face (10l). The valves access holes (12) allow an actuator, as a pin, to apply pressure on the valve which is of the type of a deformable membrane.

[0081] Having the ports (21, 22, 23, 24) on the upper face (10u) allows:

- a better access to the sample outlet port (22) during pipetting, and avoids spillage;
- to freely place syringes containing the samples on the sample inlet ports (21).

[0082] Having all the valve access holes (12) on the lower face (10l) of the plate (1) allows to better allocate space for fluidic actuators in an automated machine processing the plate (1), with:

- the pipe connectors on the same side of the plane (P), and
- valve actuators on the same side of the plane (P).

[0083] Grouping the fluidic actuators by their category, i.e., pipe connectors and valve actuators, allows to simplify the conception of the automated machine.

[0084] Figure 5 also illustrates locating elements (16) allowing to correctly position each chip (10) in a nesting platform of the machine configured for an automated lysing. More precisely, the nesting platform is configured to receive the plate (1), and the nesting platform comprises a bay for each of the chip (10). Each bay comprises locators configured to match with the locating elements (16) of the chip (10), and correctly position the chip (10) for the automated process.

[0085] This way:

- a complete plate (1) is correctly positioned in the nesting platform; and
- each chip (10) which has been separated from the plate (1), thanks to the frangible line (S), is also correctly positioned in the nesting platform;

provided that each chip (10) of the plate (1) is held by the locators of each bay, through the locating elements (16).

[0086] In order to ease the positioning of the chips (10) or the plate (1) when laying them on the nesting platform, the locating elements (16) are disposed on the lower face (101) of the chip (10) plane (P). Gravity maintains the chips (10) or the plate (1) in a correct positioning.

[0087] In the preferred illustrated embodiment, the locating elements (16) comprise a centering pin (16c), a planar support (16p) and an anti-rotation device (16ar). This combination provides an isostatic positioning, which avoids a deformation of the chip (10) or of the plate (1).

[0088] Other types of locating elements (16) can be used. An isostatic positioning is preferred.

[0089] In particular, a ridge defines both a structural reinforcement of the chip (10) and the planar support (16p). In this way, the design of the chip (10) is simpler, and the manufacturing of the chip (10) is cheaper. The ridge may be peripheral.

[0090] Preferably, the locating elements (16) are disposed with a dissymmetric pattern, in order to prevent the mounting of a chip (10) in an inverted position. This provides "mistake-proofing" locating elements (16), because a chip (10) cannot be mounted in the nesting platform in a different way than the one expected.

[0091] In the illustrated example:

- the centering pin (16c) is offset in a longitudinal direction (L) of the chip (10),
- the anti-rotation device (16ar) is offset both in the longitudinal direction and a transversal direction (T) of the chip (10).

[0092] Figure 6 illustrates in details the components of the preferred chip (10) from the plate (1), and summarizes the afore-mentioned features.

[0093] In order to avoid cross contamination during the lysing of the sample, the chip (10) comprises the following

separate ports: a sample inlet port (21), an elution port (23), a rinsing port, and sample outlet port (22).

[0094] The fluidic network comprises the following ducts:

- a loading duct (d1), linking the sample inlet port (21) to the lysing chamber (25);
- a elution duct (d2), linking the lysing chamber (25) to the sample outlet port (22);
- a supplementary duct (d3), linking the elution port (23) to the lysing chamber (25);
- a discharge duct (d4), linking the lysing chamber (25) to the rinsing port.

[0095] In addition to the ports, the fluidic interfaces (11) comprise the following valves:

- a first valve (26), on the loading duct (d1);
- a second valve (27), on the elution duct (d2);
- a third valve (28), on the supplementary duct (d3);
- a fourth valve (29), on the discharge duct (d4),

each valve (26, 27, 28, 29) being configured to be actuated between an open state allowing the flow of fluid through its duct (d1, d2, d3, d4), and a closed state preventing any flowing through its duct (d1, d2, d3, d4).

[0096] According to the different phases of the mechanical lysing, and the configuration to adopt for the fluidic network, each valve (26, 27, 28, 29) can be actuated by a pin (42) through the valve access hole (12).

[0097] On the top view of figure 6, the shapes of the protrusions (13) and the concavities (14) are clearly visible.

[0098] The elution port (23) provides the first lateral protrusion (13a), on the first lateral side (1011) of the chip (10). The first lateral concavity (14a), antagonist of the first lateral protrusion (13a), is disposed on the second lateral side (10 12) of the chip (10). The first lateral concavity (14a) is configured to receive the first lateral protrusion (13a) of an adjacent chip (10), placed alongside with the illustrated chip (10), on the second lateral side (10 12).

[0099] The discharge port (24) provides the second lateral protrusion (13b), on the second lateral side (1012) of the chip (10). The second lateral concavity (14b), antagonist of the second lateral protrusion (13b), is disposed on the first lateral side (10 11) of the chip (10). The second lateral concavity (14b) is configured to receive the second lateral protrusion (13b) of another adjacent chip (10), placed alongside with the illustrated chip (10), on the first lateral side (10 11).

[0100] The sample outlet port (22) provides the transversal protrusion (13c), on a transversal side (10 t) of the chip (10). The transversal concavity (14c), antagonist of the transversal protrusion (13c), is disposed on the same transversal side (10 t). The transversal concavity (14c) is conformed to receive the transversal protrusion (13c) of a facing chip (10), placed in a staggered manner with the

illustrated chip (10), on the other side of the axis (10a) which is aligned with the transversal side (10 t).

[0101] Figure 7 illustrates a cross section view of the chip (10), illustrating a conicity (α) of fluidic interfaces (11). The conicity (α) allows a better self-positioning of a fluidic actuator actuating the chips (10) of the plate (1), notably during the automated process of the plate (1). The conicity (α) helps guiding the actuator.

[0102] On figure 7, not all conicities (α) are illustrated in order to prevent overfilling figure 7.

[0103] Preferably:

- the conicity (α) of a pestle access hole (25a) for a pestle eases the introduction of said pestle for the mechanical lysing of the sample;
- the conicity (α) of the valve access hole (12) eases the introduction of actuators, as pins, for actuating its valve (26, 27, 28, 29).

[0104] As for the ports (21, 22, 23, 24), the conicity (α) provides two advantageous effects: it eases the introduction of the pipe connector, or syringe, intended to plugged on its port, but it also ensures the sealing of this connection through a radial tightness obtained through the diameter increase of the conicity (α).

[0105] The illustrated ports (21, 22, 23, 24) present an internal conicity (α), because their interfaces are configured to plug into said ports (21, 22, 23, 24). The conicity (α) can also be external, for interfaces capping said ports (21, 22, 23, 24), instead of plugging into said ports (21, 22, 23, 24).

[0106] The structure and functioning of the machine are described below. Only the best mode wherein the machine is configured to process a plate (1) of chips (10) is described. It must be understood the machine can also be configured to process a single chip (10), or several chips (10) detached from a plate (1).

[0107] In reference to figures 8 and 9, the machine comprises several subassemblies:

- a nesting platform (30) configured to position and to hold the plate (1);
- a pin carrier (42c) configured to position pins (42) in relation with the valve access holes (12);
- a pestle carrier (51c) configured to bring pestles (51) in contact with the lysing chambers (25) during the mechanical lysing;
- a connector carrier (41c) configured to plug the pipe connectors (41) onto their respective ports;
- a fluidic system (70) linking the pipe connectors (41) to supply fluids used during the process, e.g., an elution solution tank.
- a gantry (47c) configured to load the samples to be tested in their respective chips (10);
- a frame (60) defining a structure of the machine;
- an electrical cabinet (90) containing a power supply for the machine and a computer configured to automatically process the plate (1). A human machine

interface is connected to the computer.

[0108] The plate (1) defines a chip plane (P), and the nesting platform (30) defines a nesting plane (N) so that the chip (10) is positioned and held by the nesting platform (30), the chip plane (P) being parallel to the nesting plane (N).

[0109] In the illustrated embodiment, the nesting plane (N) is horizontal because the elution port (22) of the chip (10) is protruding orthogonally in relation to the chip plane (P), and it is preferred that the elution port (22) is vertical and oriented upwards, in order to prevent spillage of the eluded sample. Other designs of the machine can be sought, according to the design of the chip (10).

[0110] We can see on figure 8 that actuators (40) of the machine are disposed on both sides of the nesting plane (N), some actuators (40) being above the nesting plane (N), and other actuators (40) being below the nesting plane (N). In the preferred embodiment:

- the pipe connectors (41) are mounted on the connector carrier (41c), above the nesting plane (N), and
- below the nesting plane (N), there are:

- the pestles (51) mounted on the pestle carrier (51c), and
- the pins (42) mounted on the pin carrier (42c).

[0111] Distributing the actuators (40) in different orientations regarding the chip (10) allows to better occupy the volume surrounding the chip (10) with some components of the machine. By consequence, the layout of the machine can be reduced, and occupy less space on a laboratory bench.

[0112] In particular reference to figure 9, we can see in better details that:

- above the nesting plane (N) are disposed:

- syringes (33) containing the samples, each syringe (33) being plugged onto the sample inlet port (21) of its chip (10);
- pipe connectors (41), configured to link each chip (10) to the fluidic system of the machine, and placed on the connector carrier (41c);
- ejectors (46), configured to help in holding the plate (1) during the process, as well as unplug the pipe connectors (41) from their ports at the end of the lysing process; and

- below the nesting plane (N) are disposed:

- pestles (51) configured to apply pressure on the lysing chambers (25) of the chips (10) during the mechanical lysing;
- pins (42) configured to actuate the valves of the chip (10).

[0113] Syringes (33) are a preferred embodiment to load the samples into the chips (10): being disposable, syringes (33) avoid any cross-contamination between successive lysing on the machine.

[0114] Syringes (33) are plugged onto the sample inlet ports (21) before the lysing process. Loading the sample (S) into the chip (10) requires to push on a piston of the syringe (33). In order to reduce the number of actuators (40) needed on the machine, a syringe pusher (47) simultaneously pushes the pistons of several syringes (33).

[0115] The syringe pusher (47) is mounted on a gantry (47c), which is linked to the frame (60) through a sliding connection, comprising an upper rail (61) mounted on the frame (60), and bushes (62) mounted on the gantry (47c). The gantry (47c) is actuated by a first motor (M1), through a screw-and-nut connection.

[0116] According to the plate (1) layout, two rows of syringes (33) need to be pushed, so the gantry (47c) comprises two syringe pushers (47), and the first motor (M1) alone can actuate the whole pistons at once.

[0117] As shown in figure 10, the gantry (47c) is mobile between at least:

- a waiting position (47c-w) wherein the gantry (47c) is away from the nesting platform (30) so that the syringe pusher (47) is distant from the syringe (33), and
- a pushed position (47c-d) wherein the gantry (47c) is close to the nesting platform (30), so as to push the piston of the syringe (33) in order to load the sample (S) into the chip (10);
- and preferably an intermediate position (not shown), between the waiting position (47c-w) and the pushed position (47c-d), wherein the syringe pusher (47) comes in contact with a piston, but has not begun to load the sample (S) into the chip (10).

[0118] The connector carrier (41c) is mobile between a disconnected position (41c-d), wherein the pipe connectors (41) are distant from the chip (10), and a connected position (41c-c), wherein the pipe connectors (41) are plugged onto the ports.

[0119] The tubing linking the pipe connectors (41) to the fluidic system is not shown in order to avoid overloading the figures. Nevertheless, it will be understood that the shape of the gantry (47c) leaves space between the two syringes pushers (47), which leaves room for the passage of said tubing.

[0120] The connector carrier (41c) is linked to the frame (60) through a sliding connection comprising a rail mounted on the frame (60), and bushes (63) mounted on the connector carrier (41c). In order to improve the capacity of the machine, the connector carrier (41c) and the gantry (47c) are linked to the frame (60) by the upper rail (61).

[0121] As for the gantry (47c), the connector carrier (41c) is actuated by a screw-and-nut connection, driven

by a second motor (M2). Here again, the second motor (M2) alone allows to actuate the plugging and the unplugging of the whole pipe connectors (41) of the machine.

[0122] When the connector carrier (41c) also comprises ejectors (46), they play two distinct roles:

- the ejectors (46) are configured to help in maintaining the plate (1) in the nesting platform (30) during the overall process; this maintaining avoids deformations of the plate (1), which could lead to leakage or spillage,
- the ejectors (46) are also configured to help in unplugging the pipe connectors (41) from their ports, when the pipe connectors carrier (41c) moves back from the connected position (41c-c) to the disconnected position (41c-d), at the end of the lysing process.

[0123] In order to improve the maintaining of the plate (1) during the mechanical lysing, the ejector (46) of a chip (10) is coaxial to the pestle (51) of that chip (10).

[0124] The ejectors (46) are mounted slidable in relation with the connector carrier (41c) according to a direction (a), illustrated by a double arrow on figure 9, and are actuated by compensation means, as a spring. The compensation means is a returning member, elastically deformable, that urges the ejector (46) towards a protruding position wherein the ejector (46) protrudes from the connectors carrier (41c). This way, the actuation of the ejectors (46) does not require a further actuator, as a motor, for unplugging the pipe connectors (41) from their ports.

[0125] If possible, each fluidic actuator (40) is axially mobile according to the direction (a) which is orthogonal to the nesting plane (N), between:

- a passive position where the fluidic actuator (40) is distant from the chip (10) and does not actuate its fluidic interface of the fluidic network, and
- an active position where the fluidic actuator (40) is in contact with the chip (10) and actuates its fluidic interface of the fluidic network,

as this allows an easy driving of the fluidic actuators.

[0126] The nesting plane (N) being horizontal in the illustrated embodiment, the direction (a) is vertical.

[0127] Moreover, preferably each fluidic actuator (40) is linked to the frame (60) with an elastic compensation in the direction (a). The elastic compensation is a returning member, elastically deformable, that urges the fluidic actuator (40) towards the chip (10). The elastic compensation allows to set the force applied to the chip (10) by each fluidic actuator (40).

[0128] Furthermore, in order to help the unplugging of the pipe connectors (41) from their ports without deforming the plate (1), at the end of the lysing process, the gantry (47c) is brought from the pushed position (47c-d)

to the waiting position (47c-w) after the connector carrier (41c) is brought from the connected position (41c-c) to the disconnected position (41c-d).

[0129] This way, the syringe pushers (47) help in maintaining the plate (1) by applying a force through the syringes (33) plugged into the sample inlet ports (21).

[0130] The ejectors (46) comprise an elastomeric head configured to not damage the chips (10) when applying pressure on them. Preferably, the surface of the elastomeric head is greater the diameter of the lysing chamber (25).

[0131] Figure 10 also shows a displacement of the nesting platform (30) parallelly to the nesting plane (N), and between:

- a loading position (301) wherein the nesting platform (30) is accessible to an operator placing a chip (10) on the nesting platform (30); and
- a processing position (30p) wherein the fluidic actuators (40) face the chip (10); so that the chip (10) can be processed by said fluidic actuators (40).

[0132] Figure 11 illustrates the connector carrier (41c) with its bushes (63), a nut (41c-n) for the driving, the ejectors (46), and the pipe connectors (41).

[0133] The pipe connectors (41) may be in plastic material and may be fragile. In order to shield them from unwanted collisions, they are disposed in recesses (41n) machined in the pipe connectors carrier (41c), which is made of a material harder than the material of the pipe connectors (41).

[0134] As for the ejectors (46), the pipe connectors (41) are mounted slidable in relation with the pipe connectors carrier (41c), according to the direction (a), between:

- a deployed position wherein the pipe connectors (41) protrude from the connector carrier (41c) in direction of the nesting platform (30), and
- a retracted position wherein the pipe connectors (41) are drawn back in the recesses of the pipe connectors carrier (41c).

[0135] Each connector (41) is pushed in its deployed position by a compensation part, as a spring.

[0136] This design allows:

- to precisely set the force applied by each connector (41) to its mating port, in order to:
 - ensure airtightness of the plugging (if a sufficient force is applied), and
 - prevent jamming of a connector (41) (if a too great force is applied);
- to correct an eventual misalignment of ports and pipe connectors (41): the sliding between one connector (41) and the pipe connectors carrier (41c) also allows a slight rotation of the connector (41), which can self-

align with its port. This self-alignment is further helped if the port features a conical shape.

[0137] Figure 12 illustrates schematically the fluidic connections. For a simplification purpose, only a single chip (10) is illustrated.

[0138] The sample inlet port (21) receives the syringe (33) (not shown), with a rather simple configuration.

[0139] The discharge port (24) is linked through a discharge tube (77) to a discharge tank (78) configured to collect all discharge material from the lysing process, notably a rising solution evacuating macroscopic materials from the sample (S).

[0140] In order to help the evacuation of the rinsing solution, the discharge tank (78) is a vacuum tank, maintained to a pressure inferior to the ambient pressure.

[0141] The elution port (23) is linked by an elution tube (76) to a four-way fluidic distributor (75) comprising:

- a first inlet (71) towards a rinsing solution tank,
- a second inlet (72) towards an elution solution tank,
- a third inlet (73) linked to a compressed air source by an air tube (73t), and
- an outlet (74) linked to the elution tube (76).

[0142] Having distinct first inlet (71) and second inlet (72) allows the use of different solutions during the rinsing step and during the elution step. The nature of each solution can be suited for each step (e.g., Ph, solvent, etc.). By consequence, each of the first inlet (71) and the second inlet (72) is linked to its respective tank through its own tubing.

[0143] Moreover, the quantities of solution delivered during the rinsing step and during the elution step are not of the same order of magnitude: 1mL for the rinsing step, and only 20 μ L to 50 μ L for the elution step.

[0144] Having separate tubing for the rinsing solution and for the elution solution also allows to dispose distinct pumps for delivering said solutions to the four-way fluidic distributor (75). An elution pump needs to be more precise than the rinsing pump. Having two separate pumps, each one tailored for its step of the lysing process, is cheaper than having a single pump designed for delivering such different volumes with the required precision.

[0145] Pushing the rinsing solution or the elution solution through the fluidic network requires a rather high pressure, approximately 4 bars. However, it is difficult to design a pump which is both precise for the small volumes to deliver, and able to deliver a high pressure. Very specific equipment is also very expensive.

[0146] In order to combine the precision of the volume delivery, and the pressure to push the solutions through the chip (10), the fluidic system (70) comprises separate pumps for a precise delivery, and a compressed air source intended to apply the required pressure on the solution once it has been correctly dosed.: once a pump has measured and delivered the desired volume of solution to the four-way fluidic distributor (75), the desired

volume of solution is pushed through the fluidic network thanks to compressed air. To this end, a third inlet (73) is linked to a compressed air source.

[0147] Preferably, the machine comprises an air compressor so that the machine only needs an electric supply.

[0148] The pressure of the delivered compressed air is monitored: if the pressure decreases, it means the solution which is pushed is circulating through the chip (10).

[0149] After the rinsing step, it may be desirable to dry the lysing chamber (25). In that case, compressed air can be delivered after the rinsing solution has fully traversed the chip (10).

[0150] During the elution step, in order to prevent the spillage of the elution solution carrying the lysed sample, the compressed air delivery is stop as soon as the elution solution has fully traversed the chip (10). As illustrated on figure 13, in order to prevent the loss of elution solution inside the tubes, in the form of droplets of elution solution adhering to internal surface of the tubing as the elution tube (76), the link between the elution solution tank and the chip (10) comprises an elution capillary tube (72c) which conveys the elution solution:

- through the four-way fluidic distributor (75),
- through the elution tube (76),
- through the connector (41),

so that it delivers the elution solution as close as possible to the chip (10), if possible inside the elution port (23) itself.

[0151] The elution capillary tube (72c) being of a very small diameter, the loss of elution solution is greatly reduced.

[0152] Once the desired quantity of elution solution is delivered, it is then pushed through the chip (10) by the compressed air which is delivered through the air tube (73t), through the elution tube (76) which surrounds the elution capillary tube (74t), and through the connector (41).

[0153] For the same purpose of controlling the delivered volume of rinsing solution, the same design can be applied with a rinsing solution capillary tube (71c) travelling from the rinsing solution tank towards the elution port (23).

[0154] In order to reduce the costs, only one elution pump and one rinsing pump are installed on the machine. The tubing between the pumps and the chips is divided by the number of chips (10) on the plate (1).

[0155] But to guarantee each chip (10) receives the correct amount of solution, the distribution between each chip (10) is sequential:

- a first volume is delivered to a first chip (10),
- then a second volume is delivered to a second chip (10),
- and so on until each chip (10) has received its solution.

[0156] The compressed air can be delivered to every chip (10) in a single step, as controlling and monitoring its delivery is less difficult compared to the solutions.

[0157] Now referring to figures 14 to 18, a part of the machine disposed below the nesting plane (N) will be described.

[0158] Figure 14 and 15 illustrates the pin carrier (42c) and the pestle carrier (51c).

[0159] The pestle carrier (51c) is slidable according to the direction (a) between:

- a grinding position (51c-g) wherein the pestles (51) can apply pressure to the lysing chamber (25), and
- a retracted position (51c-r) wherein the pestles (51) are distant from the chip (10).

[0160] The pestle carrier (51c) is linked to the frame (60) through a sliding connection comprising a lower rail (64) mounted on the frame (60), and bushes (65) mounted on the pestle carrier (51c). The pestle carrier (51c) is driven by a screw-and-nut liaison, actuated by a third motor (not shown).

[0161] The movement of the pestle carrier (51c) towards the grinding position (41c-g) brings the pestles (51) in contact with the chips (10), for the mechanical lysing. There is a need for the pestles (51) to contact the chips (10) only during the lysing step.

[0162] The pin carrier (42c) is mobile according to the direction (a) between:

- a selecting position (42c-s) wherein the pin carrier (42c) is close to the chip (10) so that the pins (42) can actuate their mating valves by moving from its passive position to its active position, and
- a distant position (42c-d) wherein the pin carrier (42c) is distant from the chip (10).

[0163] The pin carrier (42c) carries the pins (42) configured to actuate the valves of the chip (10). Each pin (42) is mobile according to the direction (a) between:

- a passive position wherein the pin (42) is retracted in the pin carrier (42c),
- a deployed position wherein the pin (42) fully protrudes from the pin carrier (42c), the deployable position being reachable by the pin (42) only if its movement is not obstructed by its mating valve, when the pin carrier (42c) is in its distant position (42c-d), and
- an active position, between the passive position and the deployed position, and wherein the pin (42) is in contact with the valve, thus actuating the valve in its closed position. The active position is reached when the pin carrier (42c) is in its selecting position (42c-s).

[0164] The pin carrier (42c) is linked to the frame (60) through a sliding connection comprising a rail mounted on the frame (60), preferably the lower rail (64), and

bushes (66) mounted on the pin carrier (42c). The pin carrier (42c) is driven by a screw-and-nut liaison, actuated by a fourth motor (not shown).

[0165] Here again, using the same lower rail (64) for two sliding connections helps improve the compacity of the machine.

[0166] The mobility of the pin carrier (42c) and of the pestle carrier (51c) allows to give room for the horizontal displacement of the nesting platform (30). Once the nesting platform (30) is in its processing position (30p), the pin carrier (42c) and the pestle carrier (51c) can move up, close to the nesting platform (30).

[0167] The pestle (51) is linked to the pestle carrier (51c) by a pestle shaft, which comprises:

- a mast (56), linked by a pivot joint to the pestle carrier (51c), and driven in rotation by a pestle pinion (55),
- a rod (57), linked to the mast (56) with a sliding joint,
- a pestle compensating member (58) as a spring, which pushes the rod (57) in direction of the nesting plane (N),
- and a ball and socket joint (52) which links the rod (57) to the pestle (51).

[0168] The ball and socket joint (52) allows a balancing of the pestle (51) during the mechanical lysing. The ball and socket joint (52) can be a cardan joint, preferably made of an elastomeric material.

[0169] Here again, the pestle compensating member (58) allows to set the force applied by the pestle (51) on the lysing chamber (25), when the pestles carrier (51c) is in its grinding position (51c-g).

[0170] A single pestle motor (not shown) can drive the rotation of all the pestles (51), thanks to meshing of the pestle pinions (55), the one with the other.

[0171] Moreover, in order to reduce the carried mass of the pestle carrier (51c), said pestle motor is fixed on the frame (60), drives a shaft (53), and a master pinion (54) is mounted:

- with a pivot joint on the pestle carrier (51c), and
- with a sliding rotation on the shaft (53).

[0172] This way, the pestle carrier (51c) does not need to carry the pestle motor, the master pinion (54) slides on the shaft (53) when the pestle carrier (51c) moves from its retracted position (51c-r) to its grinding position (51c-g).

[0173] Now referring to figure 16, the pin (42) is preferably pushed back in its active position by a pin compensation part (48), as a spring. This allows to set precisely the force the pin (42) applies the valve, based on the stiffness of the pin compensation part (48), and on the stroke between the active position and the deployed position. A too great force may damage or even pierce the valve, whereas a too low force will not close the valve.

[0174] Preferably, the pin (42) is mechanically driven between its passive position and its active position by a driver (43), through a coupler (44) which may be a cable.

[0175] To open one valve, the driver (43) pulls the cable (44) of its mating pin (42), compresses the pin compensation part (48), thus opening said valve.

[0176] Using cables (44) to drive the pins (42) allows to dispose the driver (43) remotely in the machine, as long as the cable (44) is conveyed from the pin (42) to the driver (43).

[0177] The conveying can be made with pulleys or deflections, but preferably the conveying is made through a duct (44g), as illustrated.

[0178] In order to avoid using as many as drivers (43) as there are pins (42), corresponding pins (42) of several chips (10), e.g., the pins (42) configured to actuate the first valve of the chip (10), are driven by the same driver (43).

[0179] To that end, the cables (44) of said corresponding pins (42) are linked to the driver (43) by a harness (45). The harness (45) allows to have a single cable (44m) linked to the driver (43). The actuation of a single cable (44m) avoids the tangling which may occur if each cable (44) was directly linked to the driver (43).

[0180] In another embodiment (not illustrated), the coupler is a camshaft driven by the driver (43). The camshaft extends below the process position (30p) of the nesting platform (30), and is driven by the driver (43) which is at the back of the machine (opposite to the loading position (301)).

[0181] The camshaft comprises cams pulling the pin (42) either through a crank, either through a cable. If the camshaft comprises cams for several chips (10), the camshaft can combine the functions of the coupler (44) and the collector (45).

[0182] In reference to figure 17, we can see:

- the pestle carrier (51c) is in its retracted position (51c-r), because the pestles (51) are away from the plate (1);
- the pin carrier (42c) is in its selecting position (42c-s), because pins (42) can switch between:
 - their active position (as is illustrated a first pin (42) on the left), and
 - their passive position (as is illustrated a second pin (42) on the right).

[0183] Because of the requested compacity of the machine, the pestles (51) may go through the pin carrier (42c), which presents holes configured to allow the passage of the pestles (51) according to the movements of the pestle carrier (51c) and the pin carrier (42c). Preferably, the holes are configured to guide the pestles (51) and/or the rods (57), according to an adjustment of the dimensions of the hole in relation to the dimensions of the pestles (51) and/or the rods (57). This guiding also prevents to have a cantilever mounting for the pestles (51), with a too great length unguided between the pestle carrier (51c) and the lysing chamber (25).

[0184] Figure 18 illustrates in details the mounting of

the pestle (51), made of a rigid material, with the ball-and-socket joint (52), which is here a cardan made of an elastomeric material.

[0185] Figures 19 to 21 illustrate the nesting platform (30).

[0186] Figure 19 shows the nesting platform (30) empty, ready to receive a plate (1). It comprises:

- a nesting plate (37) featuring bay (31), each bay (31) being configured to receive a single chip (10), and
- holding components, as arms (34).

[0187] Each bay (31) comprises locators (36) configured to match with the locating elements (16) of the chip (10), and correctly position the chip (10) for the automated process.

[0188] In order to make way for the pestle (51) and the pins (42) when the nesting platform (30) is in its processing position (30p), each bay (31) comprises a passage hole (38).

[0189] The arms (34) are configured to maintain the plate (1) during the process. They can be locked in position by an interlock (35) manipulable by hand.

[0190] The nesting platform (30) preferably comprises presence sensors (32) configured to detect, for each bay (31), if a chip (10) is present or not. If there is no chip (10), the computer will not send fluids (rinsing solution, etc.) to the pipe connectors (41) of this bay (31), in order to prevent spillage. This allows to use either a whole plate (1), or an incomplete plate (1) or individual chips (10).

[0191] Figure 20 illustrates the nesting platform (30) once the plate (1) has been placed, and the arms (34) are in place.

[0192] Figure 21 illustrates the plate (1) once the syringes (33) have been plugged into the chips (10).

[0193] In the illustrated example, the presence sensors (32) are configured to detect the presence of the syringe (33). Given that a syringe (33) can be mounted only if its chip (10) is present, this method is equivalent as verifying the presence of a chip (10) itself.

[0194] The alignment of the syringes (33) forming two rows is clearly visible. Each row will be pushed by a syringe pusher (47) of the gantry (47c).

[0195] Furthermore, the machine can be shaped differently from the examples given without departing from the scope of the invention, which is defined by the claims.

[0196] In particular, it is reminded that the machine can be designed for a single chip (10) instead of a plate (1), and/or the chip (10) may be of the simple type, with valves.

[0197] The pipe connectors (41) can be distributed in different direction on relation to the nesting plane (N), instead of all being on the same side. The same applies to the pins (42) and to the pestles (51). The main feature is that the fluidic actuators are distributed in different direction on relation to the nesting plane (N).

[0198] The kinematics of the modules of the machine can be different, e.g., the nesting platform (30) is fixed

and the displacement of another of the module allows to place the plate (1).

[0199] In addition, the technical features of the various embodiments and variants mentioned above can be combined with one another, either in whole or in part. In this way, the machine can be adapted in terms of cost, functionality and performance.

10 Claims

1. Machine (2) for lysing a micro-organism of a sample contained in a chip (10),

the chip (10) featuring:

- a fluidic network comprising ports, including a sample inlet port (21) for injecting the sample in the chip (10) and a sample outlet port (22) for collecting the lysed micro-organism; and
- a lysing chamber (25) for the lysing of the sample;
- the chip (10) being globally planar and extending in a chip plane (P);

the machine comprising:

- a frame (60);
- a nesting platform (30) mounted on the frame (60), and comprising a generally planar nesting plate (37) defining a nesting plane (N), the nesting plate (37) being configured to maintain the chip (10) so that the chip plane (P) is parallel to the nesting plane (N),
- fluidic actuators linked to the frame (60), and configured to actuate the fluidic network of the chip (10);
- a pestle (51) mounted on the frame (60), and configured to apply pressure to the lysing chamber (25);

characterised in that:

- the pestle (51) or one of the fluidic actuators faces a first side of the nesting plane (N), and
- another one of the fluidic actuators is oriented differently.

2. The machine (2) of claim 1, wherein the nesting platform (30) comprises several bays (31) each configured for receiving one chip (10), the nesting plate (37) of each bay (31) is coplanar with the nesting plane (N), and the bays (31) are arranged aligned with an axis (10a) and regularly spaced apart so that:

- the sample outlet port (22) of each chip (10) coincides with the axis (10a), and
 - the sample outlet ports (22) of two adjacent chips (10) are regularly spaced apart with a pitch; and
- the machine (2) comprises fluidic actuators (40) and one pestle (51) for each bay (31).
- 3.** The machine (2) of any of the preceding claims, wherein one fluidic actuator (40) is axially mobile according to a moving direction (a) between:
- a passive position where the fluidic actuator (40) is distant from the chip (10) and does not actuate the fluidic network, and
 - an active position where the fluidic actuator (40) is contact with the chip (10) and actuates the fluidic network.
- 4.** The machine (2) of claim 3, wherein the fluidic actuator (40) is linked to the frame with an elastic compensation (46) along the moving direction (a).
- 5.** The machine (2) of any claims 3 or 4, wherein:
- the chip (10) comprises a valve configured to selectively open and close a duct of the fluidic network, and
 - the fluidic actuators (40) comprise a pin (42) mobile between
 - the passive position wherein the pin (42) is distant from the valve so the duct is open, and
 - the active position wherein the pin (42) is actuating the valve so the duct is closed.
- 6.** The machine (2) of claim 5 wherein the pin (42) is mechanically driven between its passive position and its active position by a driver (43), through a coupler (44) which converts the direction of movement of the pin (42), according to the direction (a), into another direction, preferably orthogonal to the direction (a).
- 7.** The machine (2) of claim 6 when combined with claim 2, wherein a collector (45) links the driver (43) to several couplers (44) of the pins (42), so that the driver (43) enables the simultaneous actuating of the valves of several chips (10).
- 8.** The machine (2) of any preceding claims, wherein:
- the fluidic actuators (40) comprise pipe connectors (41) configured to plug with the ports (23, 24) of the chip (10)
 - a connector carrier (41c) comprises all the pipe connectors (41), faces an upper side of the nesting plane (N); and is axially mobile according to a direction orthogonal to the nesting plane (N) between:
 - a connected position (41c-c) wherein the pipe connectors (41) plug with the ports (23, 24), and
 - a disconnected position (41c-d) wherein the pipe connectors (41) are distant from the chip (10);
 - a pestle carrier (51c) comprises the pestle (51), faces the lower side of the nesting plane (N) and is mobile between:
 - a grinding position (51c-g) wherein the pestle can apply pressure to the lysing chamber, and
 - a retracted position (51c-r) wherein the pestle is distant from the chip (10).
- 9.** The machine (2) of claim 8 taken in combination with claim 5, wherein the machine comprises a pin carrier (42c) which comprises the pin (42), faces a lower side of the nesting plane (N) and is mobile between:
- a selecting position (42c-s) wherein the pin (42) can actuate the valve, and
 - a distant position (42c-d) wherein the pin carrier (42c) is distant from the chip (10).
- 10.** The machine (2) of claims 8 or 9, further comprising
- a gantry (47c) holding a syringe pusher (47), configured to be in contact with a syringe (33) containing the sample and plugged to the sample inlet port (21), and mobile between at least:
 - a waiting position (47c-w) wherein the gantry (47c) is away from the nesting platform (30) so that the syringe pusher (47) is distant from the syringe (33), and
 - a discharge position (47c-d) wherein the gantry (47c) is close to the nesting platform (30), so as to actuate the syringe (33) in order to load the sample (S) into the chip (10).
- 11.** The machine (2) of claim 8 or 9 or 10, wherein the connector carrier (41c) comprises an ejector (46) configured to apply a pressure on the chip (10) towards the nesting platform (30) when the connector carrier (41c) moves from the connected position (41c-c) towards the disconnected position (41c-d).
- 12.** The machine (2) of claim 11, wherein the ejector (46) and the pestle (51) are face to face.

13. The machine (2) of any preceding claims, comprising a four-way fluidic distributor (75) comprising:

- a first inlet (71) towards a rinsing solution tank,
- a second inlet (72) towards an elution solution tank, 5
- a third inlet (73) intended to be linked to a compressed air source, and
- an outlet intended (74) to be linked to an elution port (23) of the chip (10) through a tubing, 10

wherein:

- during a rinsing step: 15
 - a rinsing pump is configured to deliver a predefined rinsing solution quantity from the first inlet (71) to the outlet (74), and
 - a compressor is configured to deliver a first predefined compressed air quantity from the third inlet (73) to outlet (74), 20

so that the predefined rinsing solution quantity can be pushed through the tubing by the predefined compressed air quantity, and 25

- during an elution step:
 - an elution pump is configured to deliver a predefined elution solution quantity from the second inlet (72) to the outlet (74), and 30
 - the compressor is configured to deliver a second predefined compressed air quantity from the third inlet (73) to outlet (74),

so that the predefined elution solution quantity can be pushed through the tubing by the second predefined compressed air quantity. 35

14. Chip (10) designed for receiving a sample to be lysed and to be processed by the machine (2) of claims 1 to 13, and featuring: 40

- a fluidic network comprising at least a first fluidic interface and a second fluidic interface, as ports or valves, 45
- a lysing chamber access hole (25a), configured for the pestle (51) can apply pressure on the lysing chamber (25),

characterised in that the first fluidic interface is placed on a lower side of the chip plane (P), and the second fluidic interface is placed differently. 50

15. Method of processing the chip of claim 14 with the machine (2) of claims 1 to 13, and comprising the steps of: 55

- displacing the nesting platform (30):

- from a loading position (301) wherein the nesting platform (30) is accessible to an operator placing a chip (10) on the nesting platform (30);

- towards a processing position (30p) wherein the chip (10) can be processed by the fluidic actuators and the pestle (51);

- plugging the pipe connectors (41) to their respective ports (23, 24);

- loading the sample into the lysing chamber (25) of the chip (10);

- rinsing the sample (S);

- lysing the sample (S) with the pestle (51);

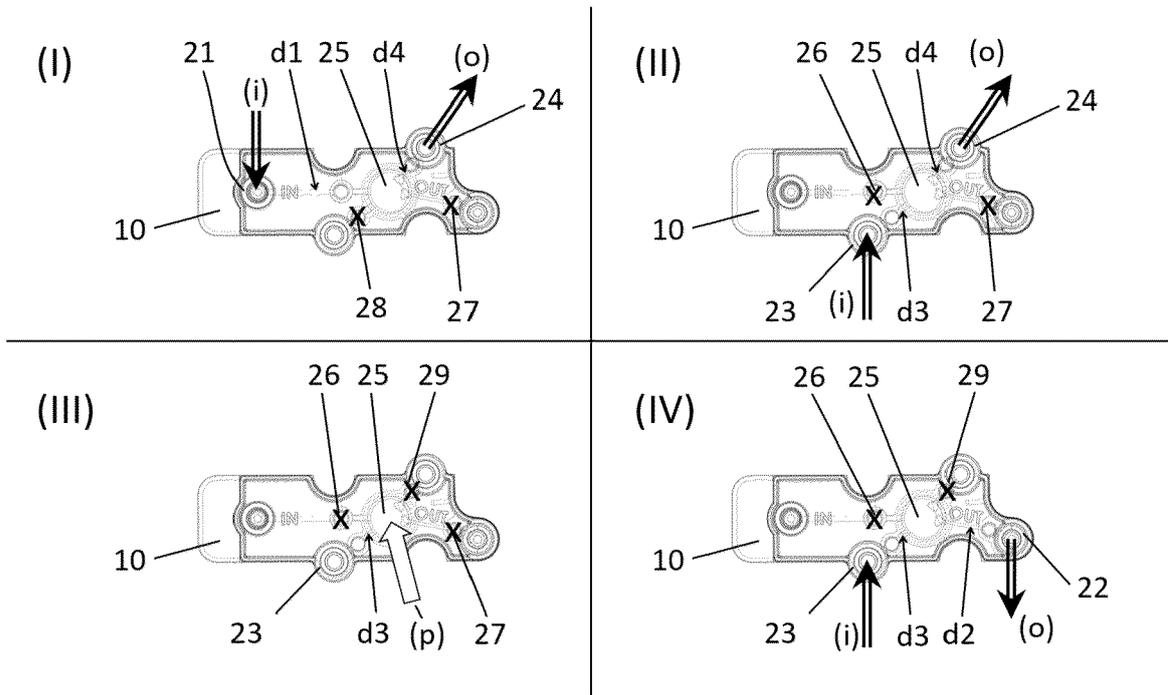
- eluding the sample (S);

- disconnecting the pipe connectors (41);

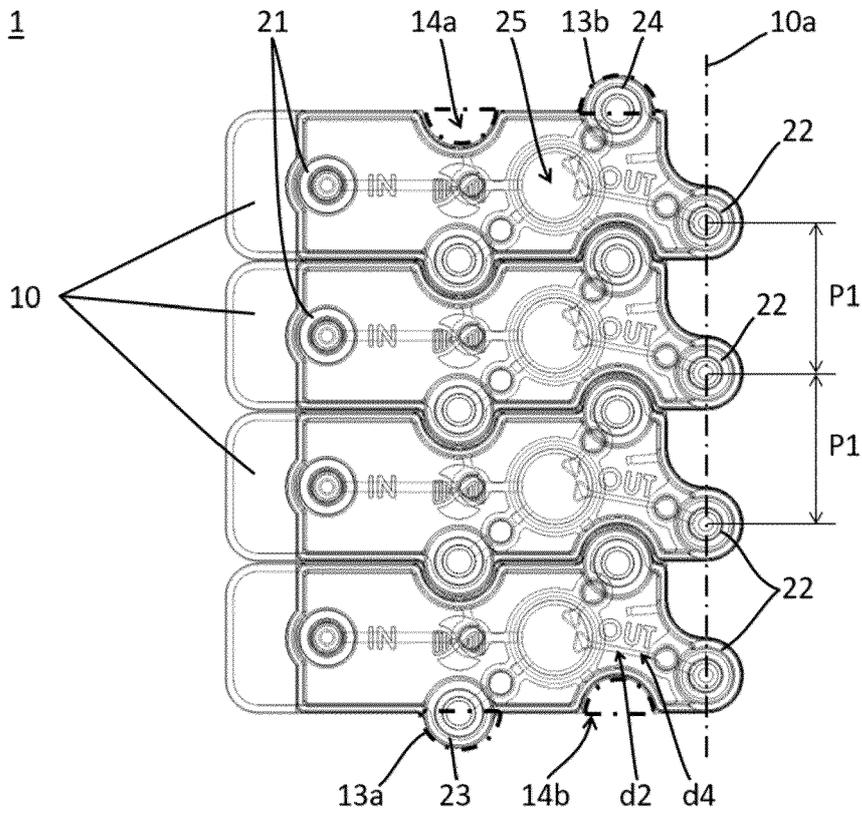
- bringing the gantry (47c) from the discharge position (47c-d) to the waiting position (47c-w);

- displacing the nesting platform (30) from the processing position (30p) towards the loading position (301).

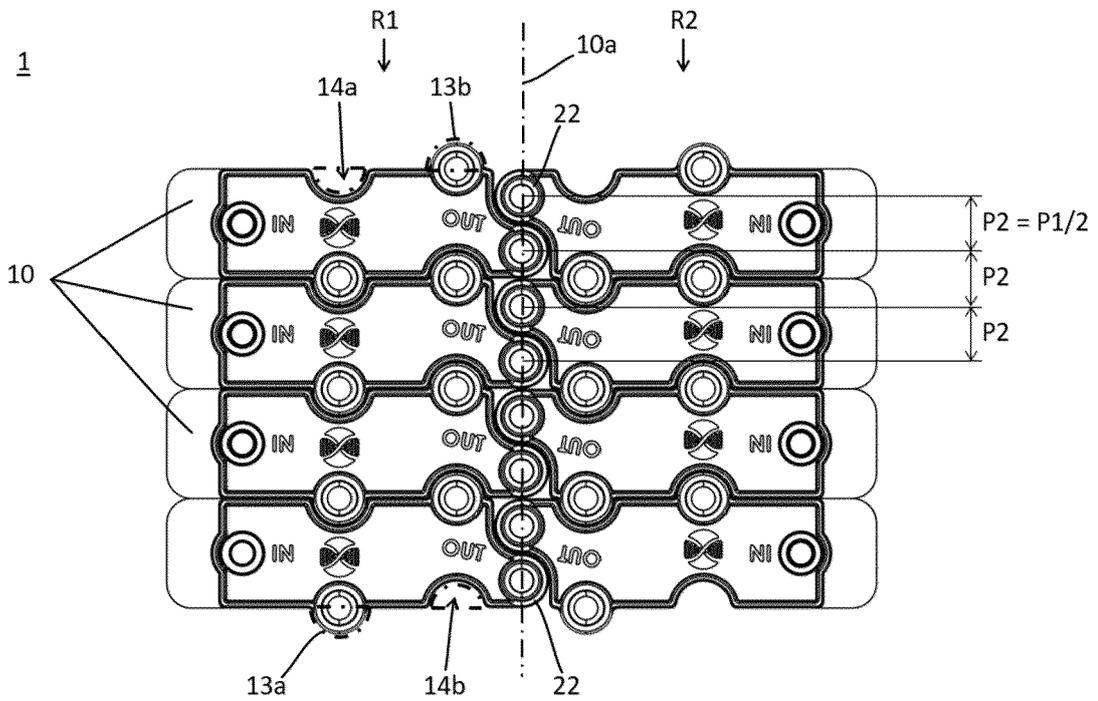
[Fig. 1]



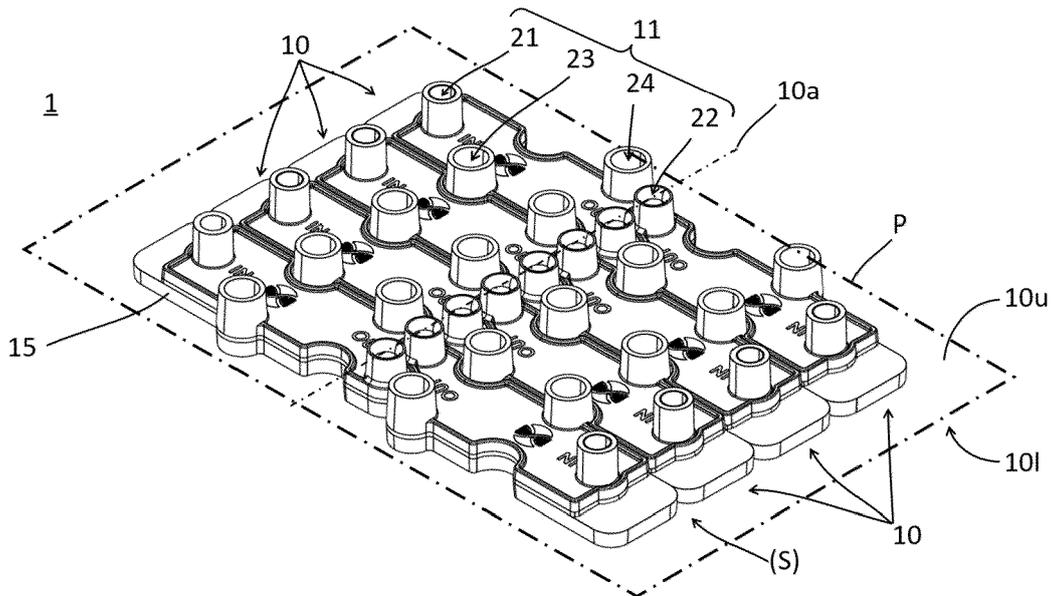
[Fig. 2]



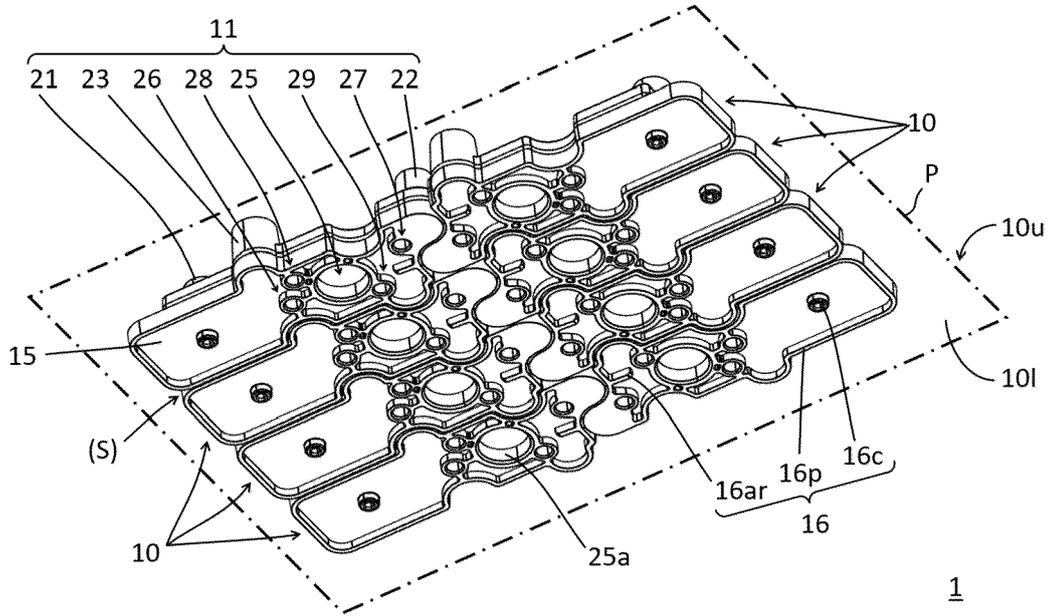
[Fig. 3]



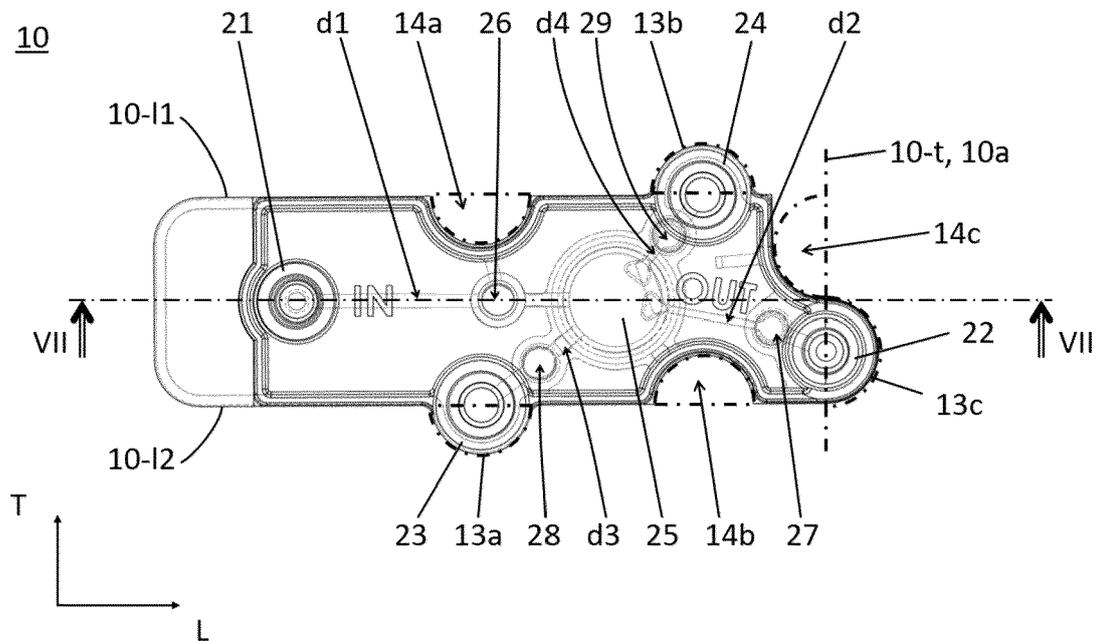
[Fig. 4]



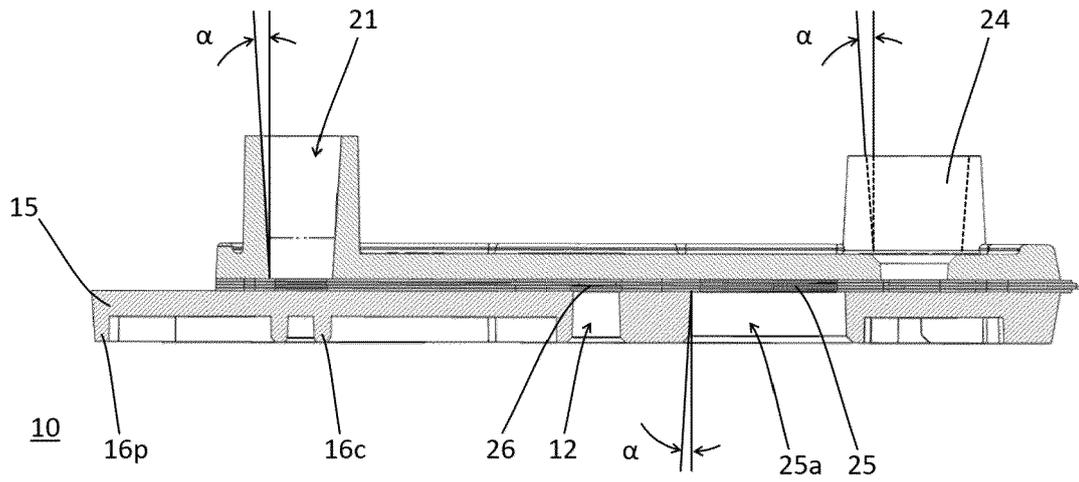
[Fig. 5]



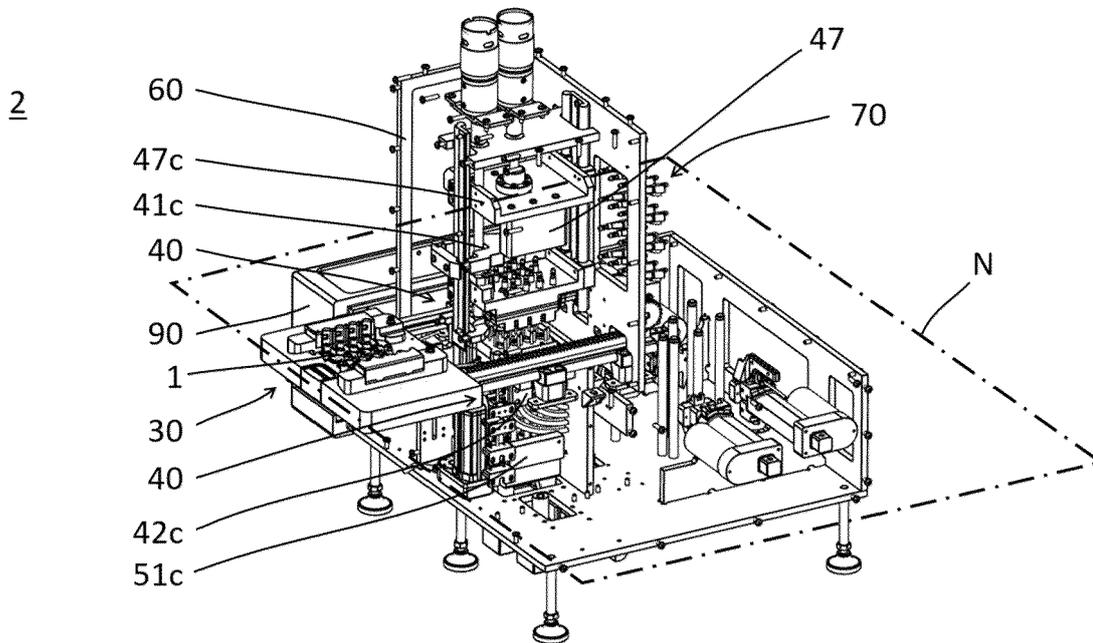
[Fig. 6]



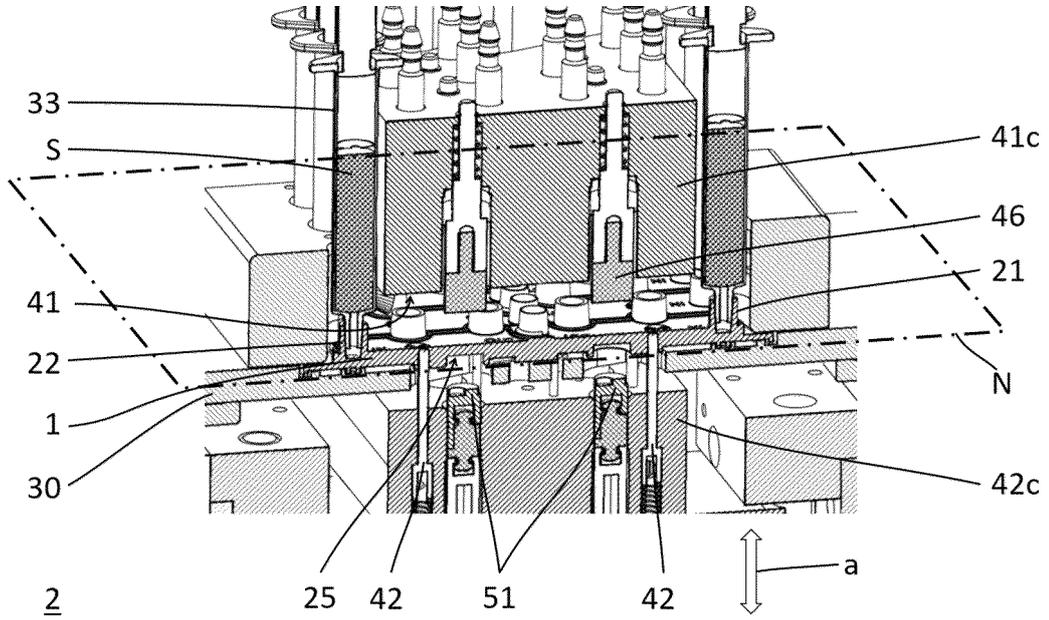
[Fig. 7]



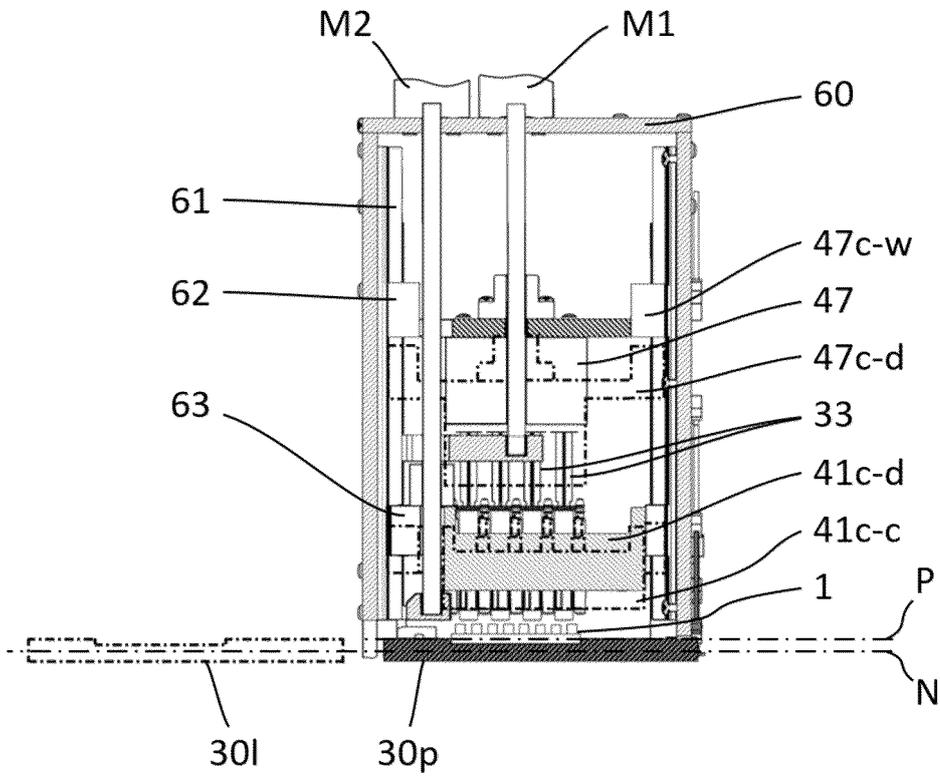
[Fig. 8]



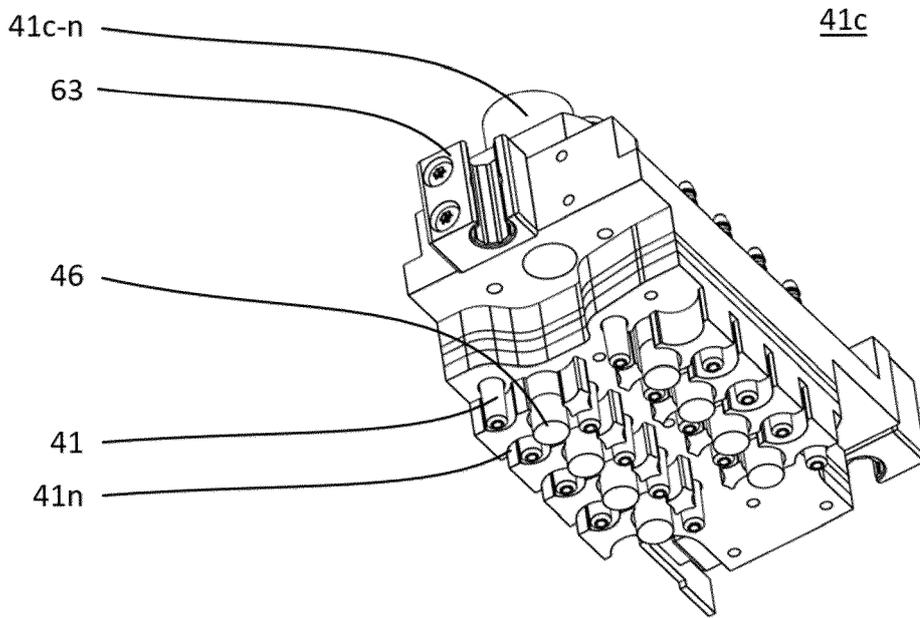
[Fig. 9]



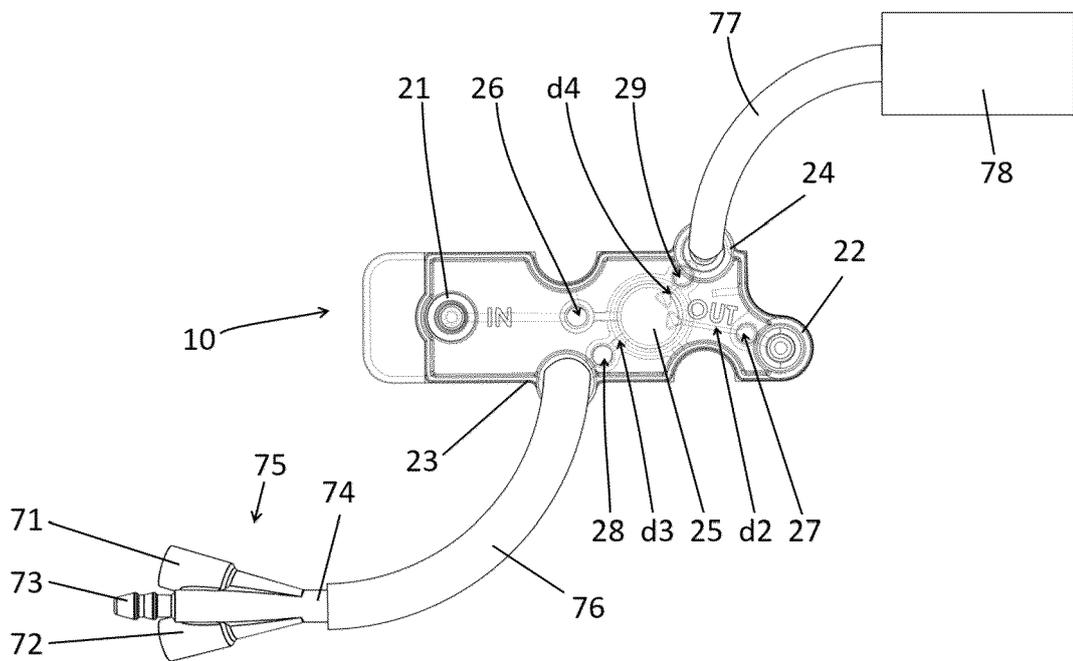
[Fig. 10]



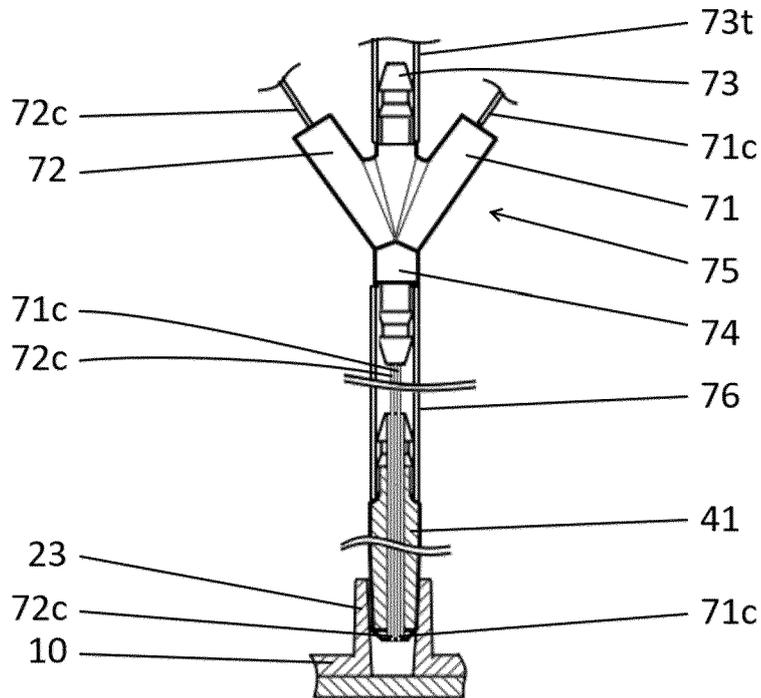
[Fig. 11]



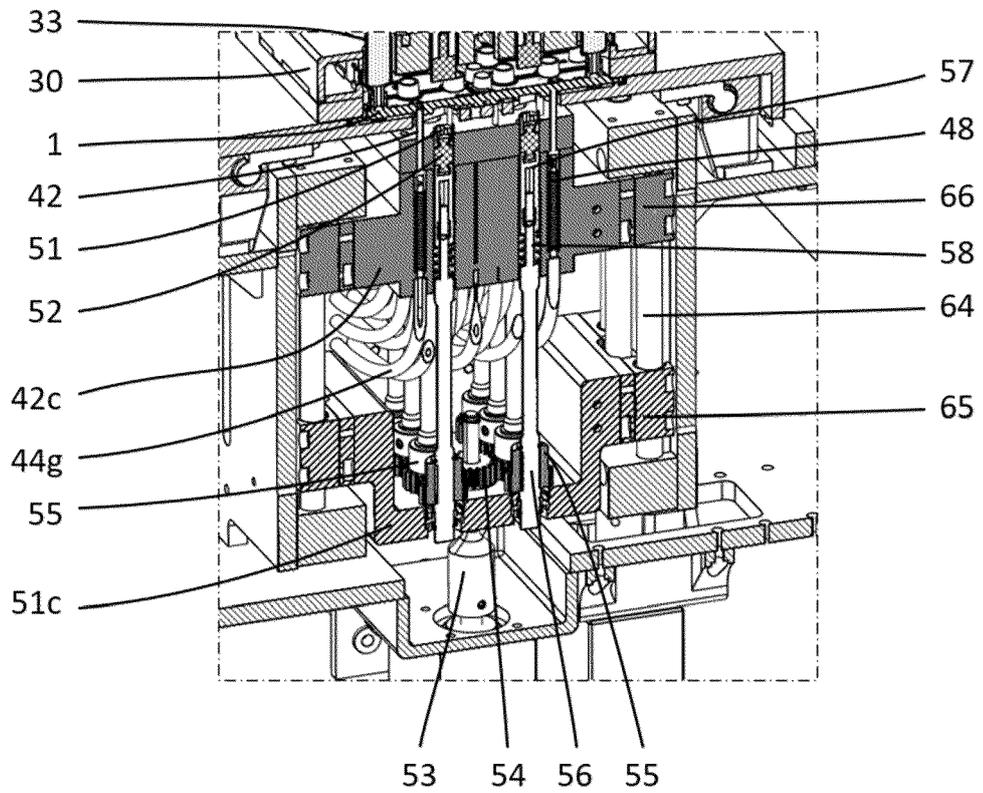
[Fig. 12]



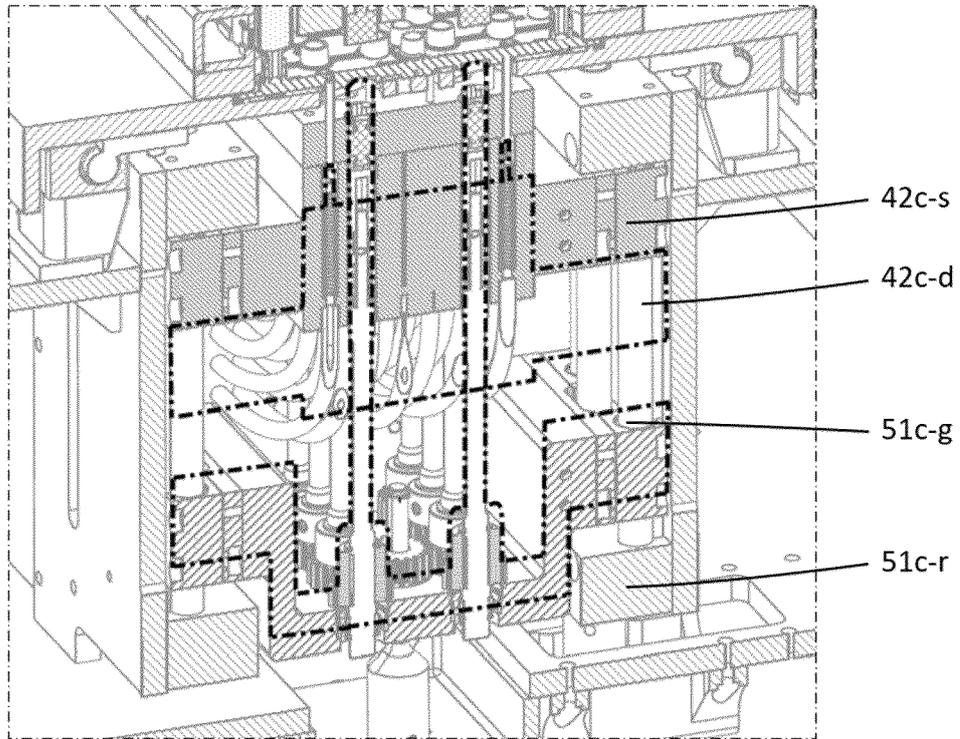
[Fig. 13]



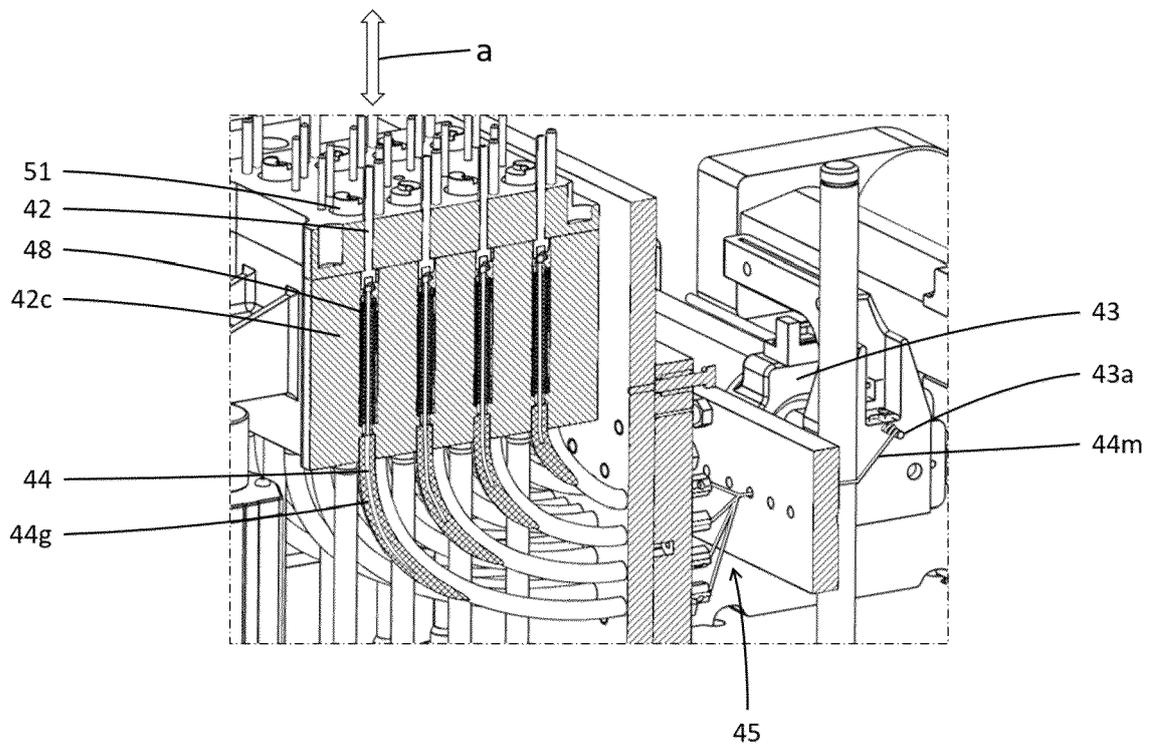
[Fig. 14]



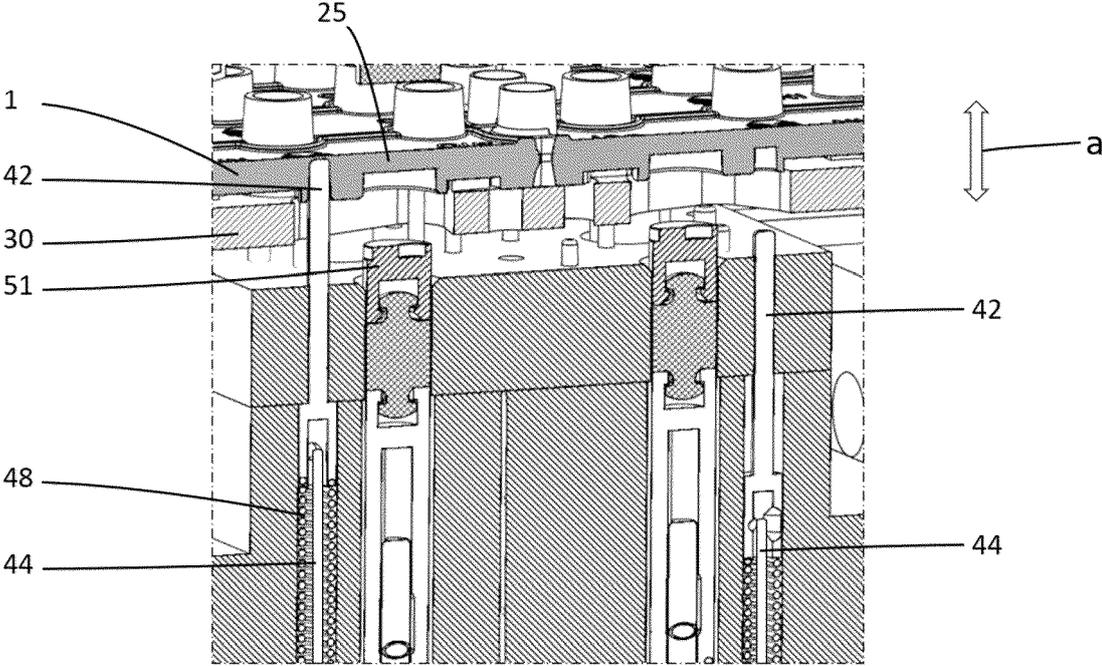
[Fig. 15]



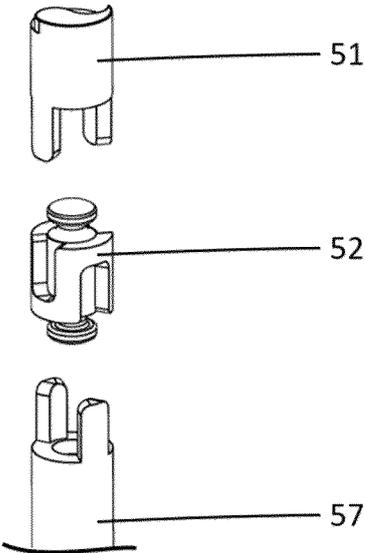
[Fig. 16]



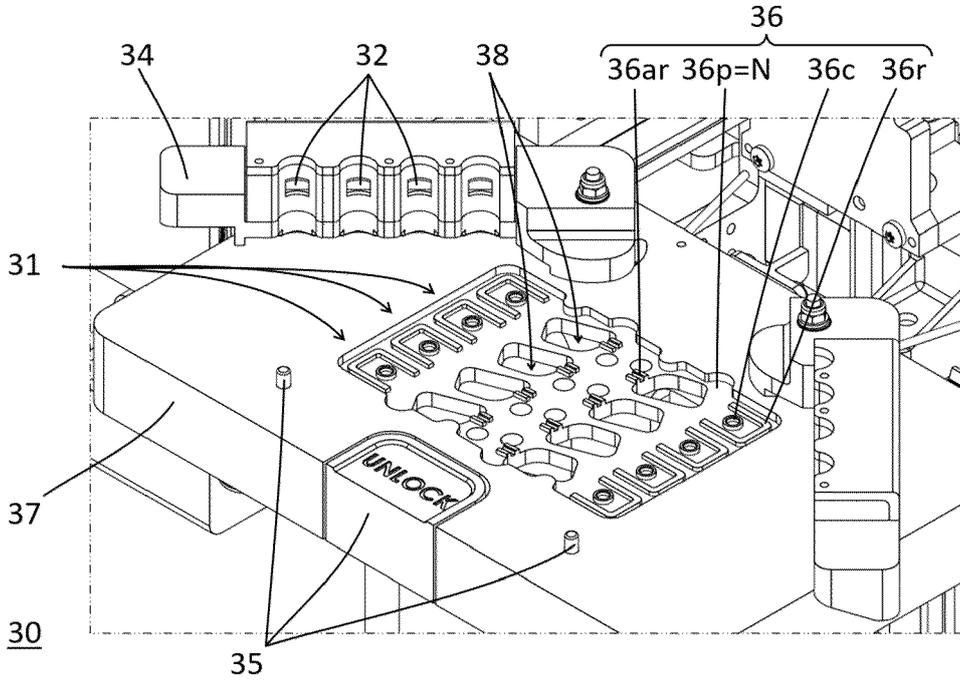
[Fig. 17]



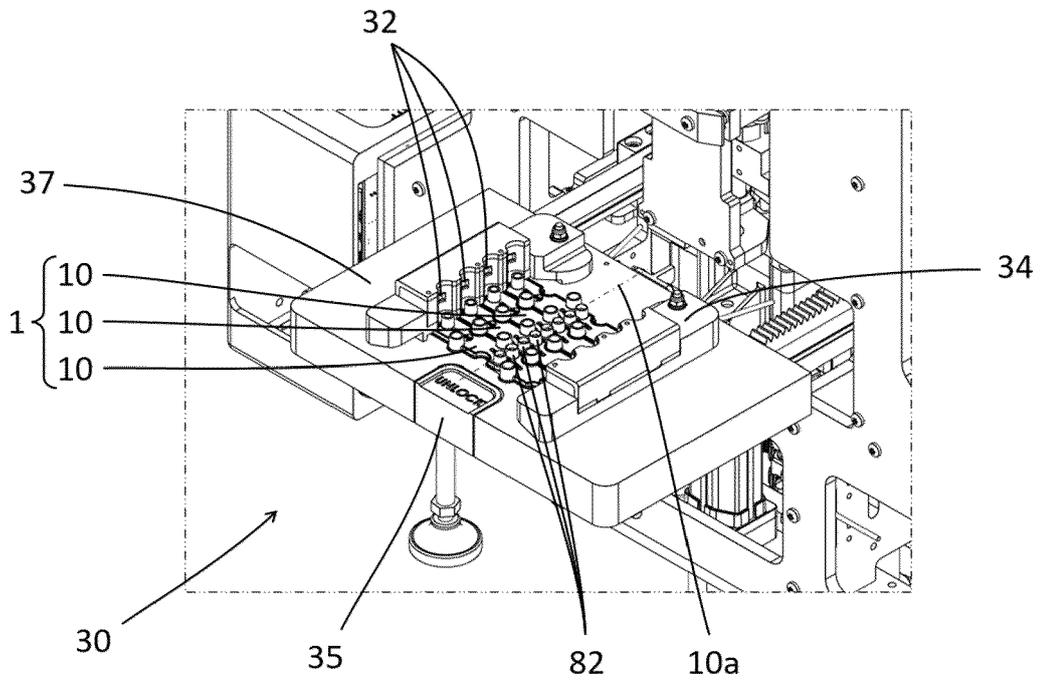
[Fig. 18]



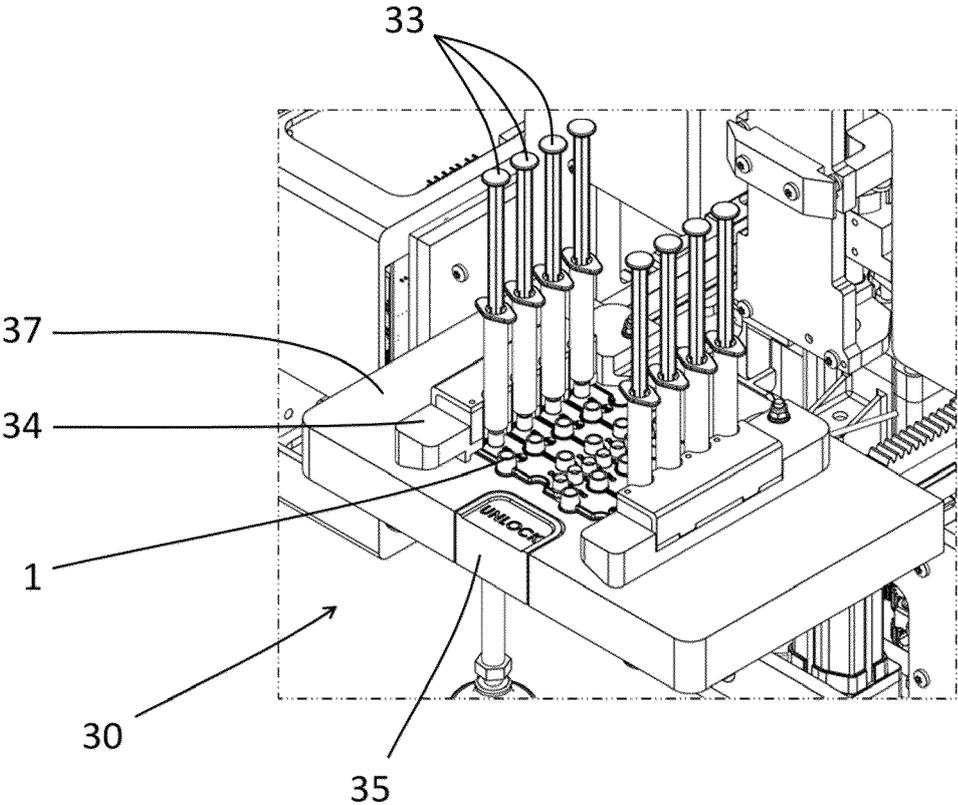
[Fig. 19]



[Fig. 20]



[Fig. 21]





EUROPEAN SEARCH REPORT

Application Number
EP 23 30 6841

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DOCUMENTS CONSIDERED TO BE RELEVANT			
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (IPC)
X	US 2014/057279 A1 (MALIK IMRAN R [US] ET AL) 27 February 2014 (2014-02-27)	1, 3, 10-15	INV. B01L3/00 B01L9/00
Y	* paragraph [0147] - paragraph [0209]; figures 34 - 38 *	2, 4-9	
Y	----- US 2011/124128 A1 (OOSTERBROEK RIJK EDWIN [CH] ET AL) 26 May 2011 (2011-05-26) * paragraph [0127] - paragraph [0130]; figure 2 *	2	
Y	----- US 2015/258544 A1 (STERN SETH [US] ET AL) 17 September 2015 (2015-09-17) * paragraph [0079] - paragraph [0089]; figures 5, 8, 19 *	4-9	
			TECHNICAL FIELDS SEARCHED (IPC)
			B01L
The present search report has been drawn up for all claims			
Place of search The Hague		Date of completion of the search 15 April 2024	Examiner Ueberfeld, Jörn
CATEGORY OF CITED DOCUMENTS X : particularly relevant if taken alone Y : particularly relevant if combined with another document of the same category A : technological background O : non-written disclosure P : intermediate document		T : theory or principle underlying the invention E : earlier patent document, but published on, or after the filing date D : document cited in the application L : document cited for other reasons & : member of the same patent family, corresponding document	

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ANNEX TO THE EUROPEAN SEARCH REPORT
ON EUROPEAN PATENT APPLICATION NO.

EP 23 30 6841

5

This annex lists the patent family members relating to the patent documents cited in the above-mentioned European search report. The members are as contained in the European Patent Office EDP file on
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15-04-2024

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Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 2014057279 A1	27-02-2014	NONE	

US 2011124128 A1	26-05-2011	CN 101893640 A	24-11-2010
		EP 2253958 A1	24-11-2010
		JP 5322996 B2	23-10-2013
		JP 2011007778 A	13-01-2011
		US 2011124128 A1	26-05-2011

US 2015258544 A1	17-09-2015	CN 106461697 A	22-02-2017
		EP 3117221 A1	18-01-2017
		US 2015258544 A1	17-09-2015
		US 2017096705 A1	06-04-2017
		US 2020206736 A1	02-07-2020
		WO 2015138696 A1	17-09-2015

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REFERENCES CITED IN THE DESCRIPTION

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Patent documents cited in the description

- WO 2023131714 A [0002] [0006] [0050] [0060]
- EP 22306591 [0008]