"DEVICES, APPARATUS, KIT AND METHOD FOR TREATING A BIOLOGICAL SAMPLE"

TECHNICAL SECTOR

The present invention concerns a hollow device, a covering device, an apparatus, a kit and a method for treating a sample.

BACKGROUND TO THE INVENTION

It is known that biological samples are treated in different ways in order to obtain the isolation of particular types of particles (normally, cells).

Examples in this regard are the devices and methods described in the patent applications PCT/IB2010/000615 PCT/IB2010/000580 (relative to the DEPArray™ system).

Usually, at the end of the above-mentioned treatments, samples are obtained in which the particles are inserted at low concentrations in a liquid component. In this regard, it should be noted that the liquid component is normally a buffer, which cannot be used in subsequent analysis steps, and the volume of the samples is usually too high. For example, the samples obtained following use of the DEPArray™ system have volumes of approximately 38µL, whereas subsequent steps (like WGA - Whole Genome Amplification) require volumes lower than 1µL.

The samples therefore have to be treated by centrifugation at high speed and an operator has to very carefully withdraw the excess liquid manually using a pipette (and slanting the test tube containing the sample). There are many problems connected with this procedure, including:

- the success of the operations depends largely on the ability of the operator; there is a risk, which can be high if the operator does not operate correctly, of

removing the particle together with the excess liquid. The success rate of the procedure is not reliable and cannot always be reproduced, and depends on the type of buffer used;

- the operations are relatively slow;
- the procedure requires particular care, such as the use of dedicated pipettes and contamination-free tips with dual filter to reduce the risk of the sample becoming contaminated during handling by the operator;
- there is a relatively high risk of the particle/s being damaged due to the centrifugation which, as mentioned, is performed at relatively high speeds(therefore imparting a relatively high stress to the particle/s).

The object of the present invention is to provide a hollow device, a covering device, an apparatus, a kit and a method which overcome, at least partially, the drawbacks of the known art and if possible are, at the same time, easy and inexpensive to produce.

SUMMARY

According to the present invention, a hollow device, a covering device, an apparatus, a kit and a method are provided as described in the following independent claims and, preferably, in any one of the claims depending directly or indirectly on the independent claims.

BRIEF DESCRIPTION OF THE DRAWINGS

The invention is described below with reference to the accompanying drawings, which illustrate some non-limiting embodiment examples thereof, in which:

- figure 1 is a lateral view of an apparatus produced according to the present invention;
- figure 2 is a lateral section of the apparatus of figure 1;
- figure 3 is a view of the apparatus of figure 1 in a

different operating configuration;
- figure 4 is a perspective view of the apparatus of figure 1;
- figure 5 is a lateral section of the view of figure 3;
- figure 6 is a lateral view of a further embodiment of an apparatus produced according to the present invention;
- figure 7 is a lateral section of the apparatus of figure 6;
- figure 8 is a view of the apparatus of figure 6 in a different operating configuration;
- figure 9 is a perspective view of the apparatus of figure 6;
- figures 10 to 14 schematically show different steps of use of a device, part of the apparatus of figure 1, according to the present invention; and
- figure 15 is an enlarged scale view of a detail of figure 14.

EMBODIMENTS OF THE INVENTION

In figures 1 to 5 and 10 to 14, the number 1 indicates as a whole an apparatus for the treatment of a sample A (biological) (see, in particular, figures 13 and 14).

The apparatus 1 is used to amplify (for example by means of PCR/RT-PCR) the (part of the) DNA/RNA contained in the sample A, for example by means of a PCR machine (known per se and not illustrated), inside which the apparatus 1 is inserted.

According to one aspect of the present invention, a hollow device 2 is provided. The hollow device 2 is suited to the treatment of a sample B (biological) and/or of the above-mentioned sample A.

In particular, the sample B (figures 10 and 11) comprises at least one particle (advantageously, at least one cell) C and a liquid component D (more precisely, a buffer, in which the cell is immersed). The sample A (in particular, figures 13 and 14) is obtained by concentration of the sample B; also the sample A therefore comprises at least the particle C.

In the present text, by particle we mean a corpuscle having the largest dimension of less than 1000 μm (advantageously less than 100 μm). Non-limiting examples of particles are: cells, cell debris (in particular, cell fragments), cell aggregates (for example small clusters of cells deriving from stem cells such as neurospheres or mammospheres), bacteria, lipospheres, microspheres (in polystyrene and/or magnetic) and microspheres linked to cells. Advantageously, the particles are cells.

According to some embodiments, the particles have the smallest dimension greater than 1μm.

In the present text, by dimensions of a particle we mean the length, width and thickness of the particle.

The apparatus 1 comprises (see, in particular, figures 5 and 13-15) the hollow device 2. The hollow device 2 in turn has an inner chamber 3; a wall 4, which delimits the inner chamber 3; and an opening 5, which establishes a communication between the inner chamber 3 and the external environment. The inner chamber 3 is suited to house the above-mentioned sample A (and/or B).

It should be noted that the hollow device 2, in addition to being used as part of the apparatus 1, is also used to separate the particle C and the liquid component D from each other (figures 10-12). In this way, sample A is obtained (concentrated with respect to sample B - figure 12) which is the one actually treated by means of the apparatus 1 (figures 13 and 14).

In particular, the hollow device 2 has an elongated (cylindrical) shape; an end 6, in the area of which the opening 5 is arranged; and a closed end 7 (which is opposite

the end 6). More precisely, the hollow device 2 has a substantially tubular shape (with one closed end). The inner chamber 3 has substantially circular transverse sections.

Advantageously, the hollow device 2 comprises at least a part 8 (which is also hollow) tapered (towards the end 7). More precisely, the part 8 has substantially the shape of a truncated cone. Analogously, the inner chamber 3 comprises at least one tapered part (towards the end 7). More precisely, the inner chamber 3 has substantially the shape of a truncated cone (in the area of the part 8). The end 7 is also an end of the part 8.

According to some embodiments, the hollow device 2 comprises a part 8', which is integral with the (more precisely is in one piece with the) part 8, is substantially tubular (cylindrical) and has a substantially constant transverse section (or with a lower degree of tapering than the degree of tapering of the part 8). The end 6 is also an end of the part 8'.

According to alternative non-illustrated embodiments, the hollow device 2 does not comprise the part 8' (in other words, the hollow device 2 consists of the part 8). In these cases, the part 8' is a separate device which can be added to the hollow device 2 for certain steps of the treatment of the sample A (and/or B). For example, the part 8' can be added during a step of genetic amplification (for example by means of PCR) when the hollow device 2 is part of the apparatus 1 in order to maintain the hollow device 2 in a substantially fixed position (as illustrated in figure 14, for example).

The inner chamber 3 has substantially circular transverse sections.

The inner chamber 3 has transverse sections with area from $10mm^2$ to $80mm^2$ (in particular, from $20mm^2$ to $35mm^2$).

The hollow device 2 also comprises a filter 9 which is arranged in the area of the end 7 and separates the inner chamber 3 from the external environment. The filter 9 has a porosity via which, in use, at least part of the liquid component D of the sample B can pass through the filter 9 (coming out of the inner chamber 3), with the particle C not being allowed to pass through the filter 9. Advantageously, the filter 9 substantially does not allow the passage of DNA/RNA fragments.

The filter 9 has pores with diameters up to 1μm; specifically, the filter 9 does not have pores with diameters greater than 1μm. Advantageously, the filter 9 has pores with diameters up to 600nm; specifically, the filter 9 does not have pores with diameters greater than 600nm. Advantageously, the filter 9 has pores with diameters up to 500nm; specifically, the filter 9 does not have pores with diameters greater than 500nm. Advantageously, the filter 9 has pores with diameters up to 450nm; specifically, the filter 9 does not have pores with diameters greater than 450nm.

In particular, the filter 9 has pores with diameters of at least 2nm (more precisely, at least 15nm); specifically, the filter 9 has the majority of pores with diameters of at least 2nm (more precisely, at least 15nm). Advantageously, the filter 9 has pores with diameters of at least 100nm (more precisely, at least 250nm); specifically, the filter 9 has the majority of pores with diameters of at least 100nm (more precisely, at least 250nm). Advantageously, the filter 9 has pores with diameters of at least 300nm; specifically, the filter 9 has the majority of pores with diameters of at least 300mm. More advantageously, the filter 9 has pores with diameters of at least 350nm; specifically, the filter 9 has the majority of pores with diameters of at least 350mm.

According to some embodiments, the filter 9 does not have pores with diameters smaller than 2nm (more precisely, smaller than 15nm; advantageously, smaller than 100nm; in particular, smaller than 250nm; more advantageously, smaller than 300nm; in particular, smaller than 350nm).

It has been experimentally observed that pores with dimensions smaller than those indicated cause an excessive reduction in the passage of liquid. To obtain outflow of the liquid in a reasonable time, it is necessary to apply a pressure which would damage the particle/s C.

It has also been observed that pores with dimensions greater than those indicated involve the risk of the particle/s C passing through the filter 9 and dispersing. Therefore, the dimension of the pores must be preferably smaller than that of the particles to be retained in the cavity. Furthermore, pores that are too large may cause, in subsequent analysis steps (for example by PCR), the loss of biological material of interest (for example genetic material).

Unless specified to the contrary, in the present text, by diameter of a pore we mean the limiting diameter, i.e. the diameter of a circle having the same area as the smallest (transverse) section of the pore.

In particular, the limiting diameter is determined by means of the method described in ASTM F316 - 03(2011) (Standard Test Methods for Pore Size Characteristics of Membrane Filters by Bubble Point and Mean Flow Pore Test).

Advantageously, the filter 9 has a thickness of less than 500μm. According to some embodiments, the filter 9 has a thickness of less than 250 μm (advantageously, less than 100μm; in particular, less than 50μm; more precisely, less than 40μm). In this way, among other things, the passage

through the filter 9 is relatively easy and relatively little material can accumulate inside the filter 9.

Advantageously, the filter 9 has a thickness greater than 1μm (in particular, greater than 10μm; more precisely, greater than 15μm). In this way, among other things, the filter 9 has sufficient mechanical strength and is able to operate correctly.

Advantageously, the filter 9 has a hold-up volume, i.e. the volume of liquid that can be contained inside the filter 9, of less than 2μL (advantageously, less than 1μL).

In this regard, it should be noted that for a high hold-up volume, higher than the starting volume specified for the desired nucleic acid analysis procedure (e.g. DNA or RNA), the reaction volumes would have to be reduced to maintain in specification the concentration of the active reagents, at least in the first steps of the procedure. This can make the reaction inefficient.

In particular, the hold-up volume is measured as follows: an initial known volume of solution is placed in the hollow device 2; the hollow device 2 is arranged in a discharge element 22 (described and illustrated below in figures 10 to 12); it is centrifuged (approximately 2000rpm for two minutes) so that there is no more solution in the inner chamber 3; the volume of liquid collected is measured and the volume measured is subtracted from the initial volume.

The filter 9 comprises (in particular, is composed of) a material chosen from the group consisting of: PC (polycarbonate), PP (polypropylene), polyethersulfones (PES), polyethylene (PE), PVDF (polyvinylidene fluoride), nylon, silicon, SiO_2, silicon nitride, fibreglass or a combination thereof.

According to some embodiments, the filter 9 comprises (in particular, is composed of) a material chosen from the group consisting of: PC (polycarbonate), PP (polypropylene), polyethersulfones (PES), PVDF (polyvinylidene fluoride), nylon, silicon, SiO_2, silicon nitride, fibreglass or a combination thereof.

According to some embodiments, the filter 9 comprises (in particular, is composed of) an organic polymer.

Advantageously, the filter 9 is made of a material that does not allow the DNA (or RNA) to become attached. In particular, the filter 9 does not contain aluminium oxide.

Advantageously, the filter 9 comprises (in particular, is composed of) a material chosen from the group consisting of: PC, (PES). In particular, the filter 9 comprises (in particular, is composed of) PC.

According to some specific embodiments, the filter 9 comprises (in particular, is composed of) Nuclepore™ (a polycarbonate membrane) and/or Supor® (a PES membrane).

The inner chamber 3 has a volume up to 2mL (advantageously up to 1 mL; in particular, up to 0.5mL; more precisely, up to 0.2mL). Advantageously, the inner chamber 3 has a volume of at least 40µL (in particular, at least 50µL; more precisely, at least 60µL).

The opening 5 has an area of at least $9mm^2$ (advantageously, at least $12mm^2$; in particular, at least $18mm^2$). According to some embodiments, the opening 5 has an area up to 80 mm^2 (advantageously, up to $70mm^2$; in particular, up to $18mm^2$).

The wall 4 has a further opening 10, which is arranged

corresponding to the end 7. In particular, the opening 10 is opposite the opening 5. The opening 10 has an area of at least $0.2mm^2$ (advantageously, at least $0.7mm^2$; in particular, at least $2.5mm^2$). The opening 10 has an area up to $13mm^2$ (in particular up to $7mm^2$).

The filter 9 is arranged in order to substantially completely cover the opening 10. Advantageously, the filter 9 has an area S (figure 15) facing the inner chamber 3 of up to $12.6mm^2$ (in particular, up to $10mm^2$). This is particularly advantageous when the hollow device 2 is used for steps of PCR. In these cases, the volume of solution is relatively small and, therefore, a large area S of the filter 9 would lead to an excessive distribution of the solution on the surface S. This would reduce the efficiency of the PCR.

In some cases, the area S is at least $0.1mm^2$ (advantageously, at least $0.7mm^2$; in particular, at least $1.2mm^2$).

The dimensions indicated in this text can be measured with profilometers.

Advantageously, the filter 9 is securely connected to the wall 4. In particular, the connection between filter 9 and wall 4 is provided by one of the following techniques: thermal bonding, solvent bonding, ultrasound bonding), laser bonding, gluing, mechanical interlocking or a combination thereof.

Advantageously, the wall 4 has a thermal conductivity of at least 0.08W/mK (in particular, at least 0.12W/mK). Conductivities above these limits are useful for allowing correct performance of treatment steps of the sample A which entail the production of heat (for example the PCR).

According to some embodiments, the wall 4 has a thermal conductivity up to 0.7W/mK (in particular, up to 0.2W/mK).

The thermal conductivity is measured according to the common technical standards. In particular, the thermal conductivity is measured according to the methodology established by ISO 22007. It should be noted that the measurements performed according to this standard are substantially compatible with those performed in accordance with ASTM 1225-09.

Advantageously, the wall 4 has a thickness up to 700μm (in particular, up to 600μm). More precisely, the wall 4 has a thickness up to 520μm (in particular, up to 450μm). Thicknesses below these limits are useful for allowing correct performance of treatment steps of the sample A which entail the production of heat (for example the PCR).

According to some embodiments, the wall 4 has a thickness of at least 40pm (in particular, at least 170μm). More precisely, the wall 4 has a thickness of at least 200μm (in particular, at least 250μm).

It is important to underline that, advantageously, the hollow device 2 is (dimensionally and structurally) suited to be used in standard PCR machines.

In this regard, we underline that, advantageously, the materials and the roughness are chosen so as to avoid the absorption of DNA/RNA fragments. The materials are chosen so as not to prevent WGA processes. The materials are chosen so as to withstand high temperatures (greater than 100°C), such as those reached during the PCR thermal cycles.

In particular, it should be noted that the hollow device 2 can be obtained by cutting (and removing) the upper part of a test tube for PCR and the tip of the test tube. The hole (opening 10) obtained corresponding to the tip is closed by a membrane (which therefore acts as a filter 9) connected to the test

tube by means of thermal bonding.

Figures 6 to 9 illustrate an alternative embodiment of the apparatus 1 and the hollow device 2. More precisely, the hollow device 2 (of the figures 6-9) is substantially identical to the hollow device 2 described above with reference to figures 1 to 4 and 10 to 14 and differs from it only in the shape of the part 8. The part 8 has an end portion 8a (arranged corresponding to the end 7) with reduced section.

According to one aspect of the present invention, a covering device 11 is provided.

With particular reference to figures 1 to 4, the apparatus 1 furthermore comprises the covering device 11. The covering device 11 is suited to preventing the outflow of liquid from the hollow device 2. In particular, the covering device 11 is suited to preventing the passage of liquid through the opening 10. More precisely, the covering device 11 is suited to fluid-tight coupling with the end 7.

The covering device 11 is (therefore) suited to coupling with the hollow device 2 so as to substantially obstruct the filter 9. In this way, the passage of material (in particular, biological material such as DNA/RNA fragments) from the inner chamber 3 to the outside through the filter 9 is substantially prevented.

As can be noted from the figures, the covering device 11 can be combined with the hollow device 2 so as to obtain the apparatus 1 (figures 1, 2 and 4) or be separated from the hollow device 2 (figures 3 and 5).

More precisely, the covering device 11 has a cavity 12 provided with an opening 13 (in particular, an open end). The cavity 12 is shaped so that it is suited to housing at least

part (in some cases, all) of the hollow device 2. In particular, the cavity 12 has a shape which is substantially complementary to the outer shape of the hollow device 2.

The covering device 11 also comprises an outer wall 14 which delimits the cavity 12.

More precisely, the cavity 12 is shaped so that (when the hollow device 2 is housed in the cavity 12) the wall 14 is (substantially completely) in contact with the hollow device 2 (in particular, with the walls 4 and 8).

Advantageously, the wall 14 has a thermal conductivity of at least 0.08W/mK (in particular, at least 0.12W/mK). Conductivities higher than these limits are useful for allowing the correct performance of treatment steps of the sample A which entail the production of heat (for example the PCR).

According to some embodiments, the wall 14 has a thermal conductivity up to 0.7W/mK (in particular, up to 0.2W/mK).

Advantageously, the wall 14 has a thickness up to 700μm (in particular, up to 600μm). More precisely, the wall 14 has a thickness up to 520μm (in particular, up to 450μm). Thicknesses below these limits are useful for allowing the correct performance of treatment steps of the sample A which entail the production of heat (for example the PCR).

According to some embodiments, the wall 14 has a thickness of at least 40μm (in particular, at least 170μm). More precisely, the wall 14 has a thickness of at least 200μm (in particular, at least 250μm).

It is important to underline that, advantageously, the covering device 11 is (dimensionally and structurally) suited

to use in standard PCR machines.

The cavity 12 has a volume up to 2mL (advantageously up to 1 mL; in particular, up to 0.5mL; more precisely, up to 0.2mL). Advantageously, the cavity 12 has a volume of at least 40µL (in particular, at least 50µL; more precisely, at least 60µL).

In the embodiment of figures 1 to 6, the cavity 12 is suited to housing (figures 3 and 5) and houses (figures 1, 2 and 4) the entire hollow device 2. In this case, the hollow device 2 comprises a gripping device 15 (in particular, a tab, which is suited to facilitating insertion into and/or extraction from the cavity 12).

According to the embodiment illustrated in figures 6 to 9, the cavity 12 is suited to housing (figure 8) and houses (figures 6, 7 and 9) only a portion of the hollow device 2 (in particular, the portion 8a).

Through the opening 13, in use, the hollow device 2 (or a part thereof) is inserted into the cavity 12. The opening 13 is arranged corresponding to one end 16 of the covering device 11.

In particular, the covering device 11 has an elongated (cylindrical) shape and a closed end 17 (which is opposite the end 16). When at least part of the hollow device 2 is inserted in the cavity 12, the end 7 is arranged corresponding to the end 17.

More precisely, the covering device 11 has a substantially tubular shape (with one end closed). The cavity 12 has substantially circular transverse sections.

Advantageously, the covering device 11 comprises at least one part 18 (which is also hollow) tapered (towards the end 17).

More precisely, the part 18 has substantially the shape of a truncated cone. Analogously, the cavity 12 comprises at least one tapered part (towards the end 17). More precisely, the cavity 12 has substantially the shape of a truncated cone (corresponding to the part 18). The end 17 is also an end of the part 18.

Advantageously, the covering device 11 also comprises at least one adjustment element 19.

The adjustment element 19 is arranged in the cavity 12. The adjustment element 19 is (among other things) suited to changing its shape so as to adapt the shape of the cavity 12 to the shape of the hollow device 2 in order to reduce the presence of air between the hollow device and the coupling device. In this way, the transfer of heat from and towards the outside is improved. This is particularly useful when performing genetic amplifications involving cycles that raise the temperature.

In particular, the adjustment element 19 is arranged corresponding to the end 17 (and, therefore, is suited to changing its shape so as to adapt to the shape of the end 7).

The adjustment element 19 is (also) suited to preventing the passage of liquid through the filter 9 (or the opening 10). More precisely, the adjustment element 19 is suited to fluid-tight coupling with the filter 9 (or with the opening 10).

Advantageously, the adjustment element 19 has a thermal conductivity of at least 0.08W/mK (in particular, at least 0.12W/mK). Conductivities higher than these limits are useful for allowing the correct performance of treatment steps of the sample A which entail the production of heat (for example the PCR).

According to some embodiments, the adjustment element 19 has a thermal conductivity up to 0.7W/mK (in particular, up to 0.2W/mK).

Advantageously, the adjustment element 19 comprises (in particular, is composed of) a material chosen from the group consisting of: elastic, elasto-plastic and liquid materials.

According to some embodiments, the adjustment element 19 comprises (in particular, is composed of) a material chosen from the groups consisting of: silicones, rubbers (natural), polymers (synthetic), lubricants, oils or a combination thereof. In some cases, the adjustment element 19 comprises (in particular, is composed of) a material chosen from the group consisting of silicones and oils.

In particular, the silicones (rubbers and polymers) have a hardness of at least 10 (more precisely, at least 15) Shore A. The silicones (rubbers and polymers) have a hardness up to 80 (more precisely, at least 70). The oils (and lubricants) have a viscosity from 1 mPa·s to 10000 Pa·s.

The hardness is measured according to the common standard techniques. In particular, the hardness is measured according to the method established by ISO 868.

The viscosity is measured according to the common standard techniques. In particular, the viscosity is measured according to the method established by ISO 3104. It should be noted that the measurements performed according to this standard are substantially compatible with those performed according to ASTM D445.

In some cases, the oils (and lubricants) have a density from 0.05g/ml to 10g/ml. More precisely, by oil we mean a mineral oil (in particular, an oil for PCR).

According to the embodiment illustrated in figures 6 to 9, the function of the adjustment element is performed directly by the wall 14 which is shaped so as to adapt to the hollow device 2 (more precisely, to the part 8 and to the relative portion 8a).

With particular reference to figures 13 and 14, it should be noted that the covering device 11 comprises at least one retaining element 20 to maintain the hollow device 2 in position inside the cavity 12.

The retaining element 20 comprises one or more projections which protrude from the wall 14 (towards the inside of the cavity 12) and are suited to coming into contact with the wall 4.

According to some embodiments, the wall 4 has one or more recesses, which are suited to being engaged by the projections of the retaining element 20. Alternatively or in addition, the wall 4 has protrusions (not illustrated) suited to coupling with the wall 14 (in particular, with the retaining element 20). In this way, the hollow device 2 is securely locked inside the cavity 12.

According to a further aspect of the present invention, a kit is provided comprising the hollow device 2 and the covering device 11.

Advantageously, the kit also comprises an adaptor 21 (figures 10 to 12), which is suited to maintaining in position the hollow device 2 inside a discharge element 22 (in particular, a relatively large test tube). The adaptor 21 has a tubular shape (in particular, annular) and an inner aperture 23 suited to receiving part of the hollow device 2.

In particular, the adaptor 21 is suited to coupling with and locking by contact the wall 4. The adaptor is also suited to be inserted in the discharge element 22 and to lock in contact with an inner surface of said element 22.

Advantageously, the kit also comprises the discharge element 22.

The element 22 has a substantially tubular shape with an open end 24, a closed end 25 and a housing 26 (for the hollow device 2 and the adaptor 21).

According to a further aspect of the present invention, a method is provided for treating the sample (biological) B (in particular, comprising at least a particle C and a liquid component D). The sample B is defined according to the above description in relation to the apparatus 1 and the hollow device 2.

The method comprises an insertion step, during which the sample B is inserted into the hollow device 2. According to the illustrations in figures 10 and 11, during the insertion step, the hollow device 2 is arranged inside the discharge element 22.

According to embodiments not illustrated, the hollow device 2 is arranged externally to the element 22, during the insertion step. In this case, after the insertion step, the hollow device 2 is positioned inside the element 22 (as shown in figure 11).

The method also comprises a concentration step (figures 11 and 12), during which a force (in the direction of the arrow F) is applied to the sample B inserted in the hollow device 2 towards the filter 9 so that at least part of the liquid component D passes through the filter 9 and the particle C

remains in the inner chamber 3, thus obtaining the sample A (concentrated). The part of the liquid component D which passes through the filter 9 is deposited in the area corresponding to the end 25.

According to some embodiments, the concentration step is achieved by applying a centrifugal force to the sample B. In said case the hollow device 2 (and the element 22) is rotated about an axis (transverse to the longitudinal axis of the hollow device 2).

It should be noted that the force applied is relatively low. In this way the risks of damaging the particle C are low. In particular, it is sufficient to rotate the hollow device to approximately 300g (2000rpm).

After the concentration step, the hollow device 2 is removed from the element 22 and coupled with (in particular, inserted in) the covering device 11 so as to substantially obstruct said filter 9 and prevent the passage of material (in particular, fragments of DNA/RNA) from the inner chamber 3 to the outside through the filter 9. In this way, the apparatus 1 is obtained.

At this point, further treatment steps of the sample A are performed and, more precisely, the particle C undergoes the treatments necessary to obtain a genetic amplification (for example by means of PCR). In particular, as the first further step, a buffer T suited to the purpose is inserted in the hollow device (figure 14).

The information disclosed in the present text can be used downstream of various types of treatment of biological material, for example:

- sorting by DEPArray™;
- other types of sorting processes (micromanipulation,

optical tweezers, laser micro-dissection etc.);

- Fluorescence Activated Cell Sorting - FACS;
- dispensing with pipette;
- dispensing with syringe.

Furthermore, the information disclosed in the present text can be used upstream of various types of treatment, for example:

- whole genome amplification (WGA)
- whole transcriptome amplification - WTA;
- polymerase chain reaction - PCR;
- fixing, permeabilization and staining or a combination thereof.

It is important to underline that the content of the present text offers significant advantages with respect to the state of the art. The advantages include the following:

- the operations are very rapid and simple (this also reduces, among other things, the risk of contamination);
- the hollow device 2 is always used; it is therefore not necessary to carry out risky transfers of samples (due both to the risk of damaging or losing the sample and the risk of contamination);
- the results are reproducible (they do not depend on the ability of the operator);
- the operations are "kind": the sample (and, in particular, the cell) is handled delicately without the need to apply high forces.

Unless explicitly indicated otherwise, the content of the references (articles, books, patent applications etc.) cited in this text is here referred to in full. In particular the above-mentioned references are incorporated here for reference.

Further characteristics of the present invention will be illustrated in the following description of two merely illustrative non-limiting examples.

Example 1

An approximately 150µL hollow device 2 was inserted in a 2mL test tube (Eppendorf) provided with adaptor 21 in order to obtain a structure analogous to the one illustrated in figure 10. A 38µl sample containing a buffer and a cell was inserted in the hollow device 2. The test tube was closed and underwent centrifugation for two minutes at 2000rpm (substantially as illustrated in figures 11 and 12).

The hollow device 2 was extracted and inserted in a covering device 11 (more precisely, a 200µL test tube for PCR) containing a few microlitres of oil for PCR. The 2mL test tube (Eppendorf) was discarded.

At this point, the contents of the device underwent amplification of the whole genome by means of the Ampli1™ WGA kit (Silicon Biosystems) and STR (Short Tandem Repeat) analysis.

The procedure was repeated 10 times and the results with Allele Call Rate were above 90% in all cases.

The known procedure was also performed, under which 10 samples like those described above (38µl samples each containing a buffer and a cell) were treated by centrifugation at high speed and an experienced operator manually withdrew, with great care and attention, the excess liquid using a pipette (and slanting the test tube containing the sample). In this case only 9 of the 10 tests performed produced results with Allele Call Rate above 90%. Also the time taken in these cases was significantly longer than the time taken using the hollow device 2.

CLAIMS

1.- A hollow device for treating a biological sample (A; B), in particular for separating at least a particle (C) and at least part of a liquid (D) from each other; the hollow device (2) comprises an inner chamber (3), which has a volume up to 2mL; a first end (6); a first opening (5), which is arranged in the area of the first end (6), establishes a communication between the outside and the inner chamber (3) and has an area of at least $9mm^2$; and a second end (7);
the hollow device (3) is characterised in that it comprises a filter (9), which is arranged in the area of the second end (7), separates the inner chamber (3) from the outside, has pores with diameters ranging from 2nm to 1µm, an area (S) facing the inner chamber (3) up to $12.6mm^2$, and a thickness up to 500µm.

2.- A hollow device according to claim 1, wherein the filter (9) has a hold-up volume lower than 2µL and an area (S) facing the inner chamber (3) of at least $0.1mm^2$.

3.- A hollow device according to claim 1 or 2, and comprising at least one wall (4), which delimits the inner chamber (3) and has a second opening (10) opposite to the first opening (5), which second opening (10) has an area from $0.2mm^2$ to $13mm^2$; the filter (9) having a thickness from 1µm to 250µm and covering in a substantially complete manner the second opening (10); the wall (4) having a thermal conductivity from 0.08W/mK to 0.7W/mK and a thickness up to 700µm; the inner chamber (3) having transverse sections with areas from $10mm^2$ to $80mm^2$; the filter (9) having pores with diameters ranging from 250nm to 600nm.

4.- A hollow device according to one of the previous claims; the hollow device (2) has a substantially tubular shape and, at least partially, the shape of a truncated cone; in

particular, the inner chamber (3) has transverse sections that are substantially circular.

5.- A covering device suited to be externally coupled to the hollow device (2) according to one of the previous claims, so as to substantially obstruct said filter (9) and prevent material from passing from the inner chamber (3) to the outside through the filter (9); the covering device (11) has a cavity (12), which is provided with an open end (16), which is suited to allow the hollow device (2) to be inserted into the cavity (12), and with a closed end (17); and adjustment means (19), which are suited to change their shape, thus adjusting themselves to the shape of the hollow device (2).

6.- A covering device according to claim 5, wherein the cavity (12) is shaped so as to be suited to house at least part of the hollow device (2); the adjustment means (19) are arranged in the area of the closed end (17) and are suited to change their shape, so as to reduce the presence of air between the hollow device (2) and the covering device (11).

7.- A covering device according to claim 5 or 6, wherein the adjustment means (19) comprise a material selected within the group consisting of: elastic materials, elasto-plastic materials and liquid materials.

8.- A covering device according to claim 7, wherein the adjustment means (19) comprise a material selected within the group consisting of: silicone, natural rubbers, synthetic polymers, lubricants, oil and combinations thereof; the silicone, the rubbers and the polymers having a hardness ranging from 10 to 80 Shore A; the oil and the lubricants having a density ranging from 0.05g/ml to 5g/ml and a viscosity ranging from 1 mPa s to 10000 Pa s.

9.- A covering device according to one of the claims from 5 to

8, comprising at least an outer wall (14), which at least partially delimits the cavity (12) and has a thermal conductivity from 0.08W/mK to 0.7W/mK and a thickness from 40µm to 700µm.

10.- An apparatus for treating a sample (A), in particular for PCR, comprising a hollow device (2) according to one of the claims from 1 to 4 and a covering device (11) externally coupled to the hollow device (2), so as to substantially obstruct said filter (9) and prevent material from passing from the inner chamber (3) to the outside through the filter (9); the covering device (11) has a cavity (12), which is provided with an open end (16) and a closed end (17); the hollow device (2) is at least partially arranged inside the cavity (12); the second end (7) being arranged in the area of the closed end (17).

11.- An apparatus according to claim 9, wherein the covering device (11) comprises an outer wall (14); the hollow device (2) being arranged in contact with the outer wall (14).

12.- An apparatus according to claim 10 or 11, wherein the covering device (11) is as claimed in one of the claims from 5 to 9.

13.- A method for treating a sample (B) comprising at least one particle (C) and a liquid component (D); the method comprises:

an insertion step, during which the sample (B) is inserted inside a hollow device (2) according to one of the claims from 1 to 4;

a concentration step, during which a force is applied to the sample (B) inserted into the hollow body (2) towards the filter (9), so that at least part of the liquid component (D) passes through the filter (9) and the particle (C) remains in the inner chamber (3).

14.- A method according to claim 13 and comprising a coupling step, which takes place after the concentration step and during which a covering device (11) is coupled to the hollow device (2), so as to substantially obstruct said filter (9) and prevent material from passing from the inner chamber (3) to the outside through the filter (9); the covering device (11) has a cavity (12), which is provided with an open end (16), through which at least part of the hollow device (2) is inserted into the cavity (12), and with a closed end (17); the second end (6) being arranged in the area of the closed end (17).

15.- A method according to claim 14, wherein the covering device (11) is as claimed in one of claims from 5 to 9.

16.- A method according to one of the claims from 13 to 15 and comprising a genetic amplification step, which takes place after the coupling step and during which at least part of the nucleic acids of the particle (C) are amplified.

17.- A kit comprising a hollow device (2) according to one of the claims from 1 to 4, and a covering device (11) suited to be externally coupled to the hollow device (2), so as to substantially obstruct said filter (9) and prevent material from passing from the inner chamber (3) to the outside through the filter (9); the covering device (11) has a cavity (12), which is provided with an open end (16), through which at least part of the hollow device (2) is inserted, in use, into the cavity (12), and with a closed end (17); the cavity (12) is shaped so as to be suited to house at least part of the hollow device (2).

18.- A kit according to claim 17, wherein the covering device (11) is as claimed in one of claims from 5 to 9.