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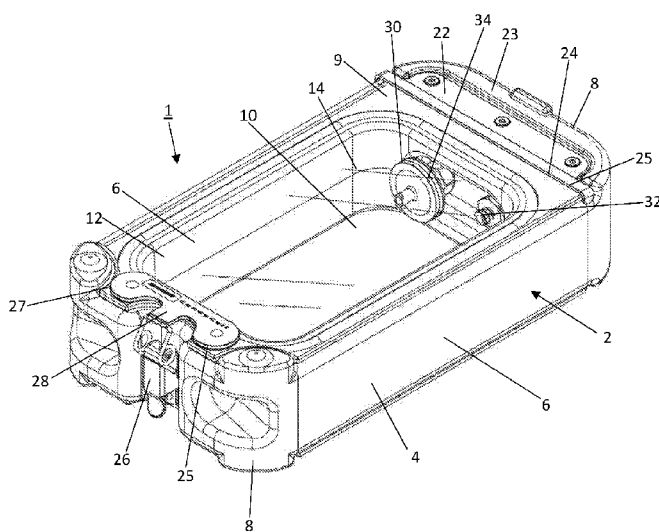


FIG. 1

(57) **Abstract:** A portable storage apparatus for live biological material such as cell cultures or microorganisms comprises a container having an opening for receiving biological material to be stored within the container. A latch is provided to close and seal the opening of the container. An inlet port in the container arranged to allow gas to flow into the container from an external gas source. An outlet port allows gas to flow out of the container. The inlet and outlet ports include valves that open the inlet port and outlet port in a first mode of operation to allow gas to circulate through the container by entering through the inlet and venting through the outlet. In a second mode of operation the inlet port and outlet port are closed by the valves to seal the container and maintain the gaseous environment therein, which enables the container to be safely disconnected from the gas supply for transport.



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## A PORTABLE STORAGE APPARATUS FOR LIVE CULTURES

The present invention relates to a portable storage apparatus for live cultures, and in particular a sealed container for transporting live cultures in a controlled atmosphere.

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In vitro cell culture experiments involve the study of live cultures such as microorganisms, cells or other biological material, in a temperature controlled gaseous environment outside of the biological host. The cells cultures are removed from an in vivo source, and transferred to an in vitro container such as a test tube or dish for analysis. In vitro cell culture experiments were traditionally performed under ambient atmospheric conditions, with oxygen levels of around 21%. However, the in vivo oxygen levels from which the cells are taken may be a factor of 2 to 5 times lower than atmospheric levels. Studies have shown that the higher in vitro oxygen levels can have detrimental effects on the physiology of the cell cultures, which could potentially influence the experimental outcome. Temperature is also important factor in biochemical reactions of cell cultures, and cells can experience thermal shock if the temperature is not properly regulated.

It is therefore known to conduct cell culture experiments at reduced oxygen levels that more closely match in vivo conditions. Such conditions, in which oxygen levels of around 3-7% replicate the physiological in vivo oxygen conditions, are referred to as physiological normoxia, or physoxia. Working under physoxia conditions places restrictions on the experimental process. The two known means of creating a physoxia environment are a tri-gas incubator and a cell culture workstation. Cell culture workstations are limited in size and only permit a single user at any one time. It is possible to operate equipment within the workstation, but the equipment that may be used is limited by the internal size of the workstation. A tri-gas incubator is large enough to store numerous samples, and the gas levels within the incubator may be controlled to the required physoxia levels. However, due to the number of samples from multiple users stored within the incubator, the incubator will be opened regularly throughout the day. Each time the door of the incubator is opened, the oxygen levels within the incubator rise. The range of change of

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gas concentration in an incubator is not dynamic, and it can take several hours to re-stabilise the gas environment and return it to the required oxygen concentration.

It is frequently necessary to remove a sample from the incubator, for numerous reasons.

5    Passaging, in which the culture is divided up into several new culture vessels to allow growth and multiply cell number, requires the controlled atmosphere of a workstation. Analysis of the sample through a microscope to assess parameters such as culture growth rates and general condition requires transfer to the microscopy station as it is not possible to integrate or house such equipment in an incubator. Alternatively, it might  
10    simply be necessary to move the culture to a more convenient incubator, for example where work on that sample is to be undertaken in another laboratory. Upon removal from the incubator the sample is exposed to ambient conditions, and remains in these conditions until it is returned to the incubator.

15    The use of open samples within an incubator has also given rise to concerns of cross contamination within the incubator, as the circulation of gas within the incubator carries contaminants between cultures.

It is therefore desirable to provide an improved storage apparatus for biological material  
20    which addresses the above described problems and/or which offers improvements generally.

According to the present invention there is provided a storage apparatus for biological material as described in the accompanying claims. In addition there is provided a method  
25    of storing biological material as described in the accompanying claims

In an embodiment of the invention there is provided a portable storage apparatus for live biological material such as cell cultures or microorganisms. The portable storage apparatus comprises a container having an opening for receiving biological material to be  
30    stored within the container. A closure is arranged to close and seal the opening of the

container. An inlet port is arranged to allow gas to flow into the container from a gas source and an outlet port arranged to allow gas to flow out of the container. In addition, valve means are configured to open the inlet port and outlet port in a first mode of operation to allow gas to circulate through the container by entering through the inlet and venting through the outlet, and to close the inlet port and outlet port in a second mode of operation to seal the container and maintain the gaseous environment therein.

In this way, the gaseous environment within the container may be controlled by circulating gas at the required levels through the container. The gas source is preferably the ambient atmosphere within which the container is located. When it is required to transport the sample, for example to analyse the sample under a microscope or move the sample or another location, the container is disconnected from the gas source. Once disconnected, the valve means, which may be one or more valves associated with the inlet and outlet, automatically close and seal the inlet and outlet so the gaseous environment within the container is sealed and maintained during transit.

Preferably the inlet port is configured for detachable connection to a gas source, and the valve means is configured to automatically close the inlet port when the inlet port is disconnected from the gas source.

The valve means preferably comprises an inlet valve coupling connected to the inlet port and an outlet valve coupling connected to the outlet port. As such, both ports have their own valve connectors and independently close and seal on disconnection.

The inlet valve coupling is preferably configured for detachable connection to a gas source coupling and is configured to open when connected to said gas source coupling in the first mode of operation and to automatically close when detached from said gas source fluid coupling to seal the inlet port in the second mode of operation. The gas source coupling is preferably a push fit female coupling and the inlet valve coupling is a male coupling.

The outlet valve coupling may be configured for detachable connection to a complimentary mating coupling, and configured to open when connected to said fluid coupling in the first mode of operation and to automatically close when detached from said fluid couplings to seal the outlet port in the second mode of operation. The  
5 complimentary mating coupling is any coupling that is able to connect to and open the outlet valve coupling. The complimentary mating coupling preferably is not connected to an outlet tube or conduit and instead vents directly into the surround atmosphere.

The container preferably includes a base, a plurality of walls upstanding from the base,  
10 and an upper surface forming an enclosure, and at least a portion of at least one of the base and the upper surface comprises a transparent material arranged such that the contents of the container are visible through said transparent material. Therefore the culture may be visually inspected without opening the container and exposing the culture to the surrounding atmosphere.

15 The plurality of walls are preferably formed from a thermally insulating material or materials. The walls may be formed from a structural foam. Alternatively, the walls may be formed having a hollow outer shell containing a thermally insulating material.

20 The base and the upper surface both comprise a transparent material arranged to such that the contents of the container are visible from above and below the container to enable the contents of the container to be studied using an optical instrument such as a microscope. The glass panels of the base and the lid align and allow the culture to be viewed from below using a microscope while being illuminated from above.

25 The closure means preferably comprises a lid formed at least in part of a transparent material and movable between an open position and a closed position.

A securing means, such as a latch or clamp is preferably provided for holding the lid in the  
30 closed position.

The portable storage apparatus may further comprise a gas circulation device for circulating gas through the container, the gas circulation device being detachably connectable to the inlet port to supply gas to the container and the valve means being configured to open the inlet port when the gas circulation means is connected and close the inlet port when the gas circulation device is disconnected. The term 'gas' is not limited to a pure gas supply, and includes for example ambient atmosphere comprising liquids and/or particulates in addition to gases. The gas circulation device is any device capable of gas movement and may be a pump, vacuum source, or pressurised gas supply, but is preferably a blower. The gas circulation device preferably comprises a fan and is configured to direct a stream of ambient atmosphere into the container when connected to the inlet port. The term ambient gas means the gas from the environment surrounding the container. Typically this will be the gas from within the incubator in which the container is housed.

15 The portable storage apparatus may further comprise a vent connector configured to connect to the outlet port, and the valve means is arranged to open the outlet port when the vent connector is connected and to close the outlet when the vent connector is disconnected.

20 The portable storage apparatus may further comprise a controller for controlling the gas circulation device when connected to the container. In the first mode of operation the gas circulation device supplies ambient gas to the inlet port and the controller is configured to stop the gas circulation device from circulating ambient gas through the container in response to a signal indicative of a variation in the ambient gas levels in order to maintain the atmosphere within the container. Stopping the gas circulation device may comprise turning the device off, or closing the supply pathway between the circulation unit and the inlet port.

The controller is preferably configured to stop the circulation of gas through the container in response to a signal indicative of a change in ambient oxygen levels.

The controller may also be configured to restart the gas circulation device based on an input indicative of the ambient gas levels having returned to a desired level or set point. In this way, gas ambient gas is not circulated through the container when the ambient gas levels are not at the levels desired within the container and circulation is only  
5 recommenced when the required levels are restored.

A door sensor may be provided that is configured to determine when the door of an incubation chamber within which the container is housed has been opened or closed. The controller is configured to stop the gas circulation device in response to a signal from the  
10 door sensor which is the signal indicative of the door being opened.

The controller is preferably configured to restart the gas circulation device based on a signal from the door sensor indicative of the door being closed and an input indicative of the ambient gas levels having returned to a desired level following closure of the door.  
15 The gas circulation device is restarted only after the door is closed and the gas levels have returned to the desired level.

The input indicative of the ambient gas levels having returned to a desired level following closure of the door is provided by a timer configured to indicate when a predetermined  
20 time has elapsed following closure of the door. The timer may be integrated into the controller and is initiated when the signal is received from the door sensor when the door is closed. The time period after which the gas circulation is restarted is pre-set and may be varied by the user via controls provided on the gas circulation device. Preferably the controller is integrated in the gas circulation device.

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The portable storage apparatus may further comprise a rapid purge device configured to connect to the inlet port of the container and supply gas to the container for a controlled period to cause a change in the gaseous environment within the container. The term 'rapid' is relative and means that gas replacement within the container occurs more  
30 rapidly than is achieved with the gas circulation device.

In another aspect of the invention there is provided a method of storage and transport of live biological material such as cell cultures or microorganisms, the method comprising providing a portable storage apparatus according to any preceding claim; placing the biological material within the container; connecting a gas circulation device to the inlet  
5 port of the container; and locating the container within a controlled gaseous environment and operating the gas circulation device to circulate ambient gas through the container.

The method may further comprise disconnecting the gas circulation device from the container when it is required to transport the container, the step of disconnecting the gas  
10 circulation device causing the valve means to close and seal the inlet and outlet ports to maintain the gaseous environment within the container the container. The container is then removed from the controlled gaseous environment and transported.

The method may further comprise the steps of detecting a condition indicative of a  
15 change in the gas levels within the controlled gaseous environment and causing the gas circulation device to stop circulating air through the container in response to the detected change.

The controlled gaseous environment may be a sealed chamber or cabinet such as an  
20 incubator or workstation, and the detected condition may be the opening of a door to the sealed chamber.

The method may further comprise the steps of detecting when the door has been closed; and reactivating the gas circulation device once a predetermined time period has elapsed  
25 following the closure of the door. Alternatively the method may comprise detecting the gas levels within the chamber and reactivating the gas circulation device once it is determined that the gas levels have reached the required level.

A purge gas may be supplied to the container prior to the container being placed in the  
30 controlled gaseous environment and connected to the gas circulation device, the supply of purge gas being controlled to change the gaseous environment within the container

such it is the same or similar to the controlled gaseous environment into which the container is to be placed.

In another aspect of the invention there is provided a portable storage apparatus for live  
5 biological material such as cell cultures or microorganisms. The portable storage  
apparatus comprises a container having an opening for receiving biological material to be  
stored within the container. A closure is arranged to close and seal the opening of the  
container. An inlet port is arranged to allow gas to flow into the container from a gas  
10 source and an outlet port arranged to allow gas to flow out of the container. A gas  
circulation device is provided that is arranged to connect to at least one of the inlet port  
and outlet port to circulating gas through the container.

The gas circulation device is configured to circulate through the container ambient gas  
from the surrounding atmosphere.  
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The gas circulation device is detachably connectable to said one of the inlet port and the  
outlet port and the gas circulation device and the container are configured such that said  
one of the inlet port and the outlet port are automatically closed when the gas circulation  
device is disconnected. The other of the inlet port and the outlet port is configured to be  
20 closed when the gas circulation device is disconnected.

The present invention will now be described by way of example only with reference to  
the following illustrative figures in which:

25 Figure 1 shows a portable storage apparatus according to an  
embodiment of the invention;

Figure 2 shows an alternative view of the portable storage  
apparatus of Figure 1;

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Figure 3 shows a portable storage apparatus connected to a gas circulation device; and

5 Figure 4 shows a portable storage apparatus connected to a purge gas device.

Referring to Figure 1, a portable controlled atmosphere apparatus 1 comprises an enclosure 2 having a main body section 4. The main body section 4 is formed from a thermally insulating material, which helps maintain the temperature within the enclosure when it is moved from a thermally regulated environment. The main body may for  
10 example be formed of a structural foam or cellular plastic, such as polypropylene, having a outer surface that is denser than the inner core. The core has a honeycomb structure and is less dense than the outer surface. The use of structural foam provides a light weight body with a smooth outer surface, which is structurally robust. In addition,  
15 polypropylene has antimicrobial properties and inhibits the formation of microbes within the enclosure. The main body section 4 includes side wall sections 6 and end wall sections 8. The side walls 6 and end walls 8 and integrally moulded and continuous, having a substantially rectangular, annular form. The main body section 4 further includes an upper surface 9 and a lower surface 11, which are substantially planar. The enclosure  
20 further comprises a base 10 and a lid 12. The base 10 and lid 12 are formed from glass or a similar transparent and durable material such as Perspex which allows the contents of the enclosure to be visible from above and below. The base 10 and lid 12 seat against the planar lower surface 11 and upper surface 9 respectively.

25 The main body section 4 has an inner surface that defines the inner wall 14 of the enclosure 2, having an upper peripheral edge 16 and a lower peripheral edge 18. The inner wall 14 extends continuously around the inner surface of the main body 4, and the corners have a curved radius to avoid sharp geometries within the enclosure and make enclosure easier to clean. The upper peripheral edge 16 of the inner wall 14 defines the opening to the enclosure. A resilient, compressible gasket sealing element 20 is provided  
30 around the upper peripheral edge 16 of the inner wall 14 on the upper surface 9. The

sealing element 20 has a curved cross sectional profile, and is seated within a correspondingly shaped groove formed in the upper surface 9, such that the sealing element 20 protrudes convexly from the upper surface 9. The sealing element 20 is compressed by and seals against the glass lid 12. A similar seal is provided on the lower surface 11 and seals against the glass base 10.

The upper surface 9 of the main body 4 includes a retaining plate 22 at a first end 23. The retaining plate extends across the width of the first end 23 and is secured to the upper surface 9 by screws or other suitable releasable fixings. A gap 25 is defined beneath the lower surface of the retaining plate 22 and the upper surface 9 of the main body 4. The gap 25 between the retaining plate 22 and the main body 4 is configured to receive the first end 24 of the lid 12 such that the first end 24 is held beneath the retaining plate 22 to vertically retain the first end 24 of the lid 12.

A pivot latch 26 is provided at the second end 27 of the enclosure 2 for locking the second end 25 of the lid 12 in the closed position. The second end 25 of the lid 12 includes a latch plate having a tongue 28 secured to end extending from the second end 25 of the lid 12. The second end 25 of the lid 12 includes a scalloped recess located beneath the tongue 28 such that the tongue is able to extend from the lids 12 without projecting past the end wall 8 and remains within the rectangular footprint of enclosure 1. The latch 26 is configured to pivot upwardly to an engagement position in which it hooks over the tongue 28. When the latch 26 is returned to the closed position in pulls the tongue 28 downwardly to move the lid 12 to the locked position.

The height of the lower surface of the retaining plate 22 is lower than the uppermost surface of the sealing element 20. As such, when the retaining strip 24 at the first end of the lid 12 is inserted in the gap 25 and the second end of the lids 12 is pulled downwardly to the locked position, the lower surface of the lid 12 is moved to a position lower than height of the uncompressed sealing element 20, causing the lid to compress against the sealing element to create a seal. The base 10 is fixed in position by releasable fixings such as screws which are used to urge the base 10 against the lower sealing element to seal

the base 10 to the main body 4. With the lid 12 in the locked position and the base 10 secured, the enclosure 2 is sealed and fully air tight.

A gas inlet 30 and gas outlet 32 are provided in the end wall 8 at the first end 23 of the enclosure. The inlet 30 and outlet 32 extend through the end wall 8 and provide a means of gas flow into and out of the sealed enclosure 2. The gas inlet 30 includes a filter 34 at its internal end arranged to filter the incoming gas flow. A similar filter may be provided on the internal end of the outlet 32 to prevent contaminants from within the enclosure 2 from being released into the atmosphere.

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As shown in the Figure 2, the external ends of the inlet 30 and outlet 32 comprise male pneumatic valve couplings 36 and 38 respectively. The male valve couplings 36,38 include integral check valves arranged such that the valve couplings are self-sealing. This means that the inlet 36 and outlet 38 valve couplings close and seal the enclosure 2 when a gas supply line and/or outlet line are not connected. The gas connections to the enclosure 2 include corresponding self-sealing female valve couplings having integral check valves arranged to seal the gas supply and gas outlet when disconnected from the enclosure 2.

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As shown in Figure 3, the controlled atmosphere apparatus 1 includes a circulation unit 40 configured to supply gas to the enclosure 2. The circulation unit 40 includes a fan arranged to draw air from the surrounding environment in which the circulation unit 40 is located, and blow the air through an outlet port 42. As can be seen, the circulation unit 40 includes a plurality of outlet ports 42, enabling it to circulate gas through a plurality of enclosures simultaneously. The circulation unit 40 further includes a power supply 44 and a plurality of sensor inputs 46 connected to a plurality of sensor output cables 48. A connection tube 50 is connected to the outlet port 42. The connection tube 50 includes a self-sealing female valve coupling 52 as described above. The self-sealing connector is particularly advantageous where the gas circulation unit 40 is connected to multiple enclosures. Upon disconnection of one or more of the enclosures the connection tube from the gas circulation unit 40 is automatically closed, which prevents the gas flow to the remaining enclosures from dropping. The female valve coupling 52 is connected to

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the male valve coupling 36 of the enclosure 2 in a push fit manner, which opens the valve couplings and locks the valve couplings 36,52 together. Air is blown into the enclosure 2 by the circulation unit 40 at a controlled low flow rate. The air flow may be varied via a controller on the circulation unit 40 depending on the refresh rate required within the enclosure 1. A second female valve coupling 54 is connected to the male outlet valve coupling 38. The second valve coupling 54 opens the outlet valve coupling 38 upon connection to allow air to flow out of the enclosure 2 at the same rate as it is blown in.

In use a sample is placed within the enclosure or container 2, and the lid 12 is locked to seal the enclosure 2. Due to the low flow rate of the circulation unit 40, it would take a significant time to lower the oxygen levels in the enclosure 2 to the required level. As shown in Figure 4, a purge unit 60 is provided to precondition the atmosphere within the enclosure 2. The purge unit 60 flows a stream of pressurised gas, or mixture or gases, which may for example be nitrogen, through the enclosure 2 to dilute the air and more rapidly lower the oxygen level within the enclosure 2 to the required level. This rapid preconditioning of the enclosure 2 reduces stress and shock to the cultured cells or microbes, encouraging normal development. The pressurised gas supply of the purge unit 60 is provided at a higher flow rate than the circulation unit 40. A supply tube 62 and female valve coupling 64 connect the purge unit 60 to the male valve coupling 36 of the inlet 30 of the enclosure 2. A nitrogen supply 66 is connected to the purge unit 60, and the purge unit 60 provides a flow of nitrogen to the enclosure 2 via the inlet 30. As the nitrogen is streamed into the enclosure 2, gas from within the enclosure 2 exits via the outlet 32. The purge unit 60 includes a timer control 68 that enables the duration of the purge to be controlled.

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The preconditioned, sealed enclosure 2 is placed into the incubator and connected to the circulation unit 40. The female valve coupling 52 of the supply tube 50 is connected to the male valve coupling 36. The outlet female valve coupling 54 is connected to the outlet male valve coupling 38. Connection of the outlet female valve coupling 54 opens the outlet. The outlet female valve coupling 54 is not connected to an outlet line, and gas is vented directly through the outlet female valve coupling 54 to the surrounding

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atmosphere. However, it will be appreciated that in an alternative embodiment an outlet line may connect to the outlet female valve coupling 54 and may be directed for example to a filtration unit before being vented from the incubator.

5 The enclosure 2 is connected to the circulation unit 40 and housed within the incubator cabinet. Due to the pre-conditioning the oxygen levels are close to the desired level at this stage. Inside the incubator the circulation unit 40 circulates air through the enclosure 2 from within the incubator. The oxygen level within the enclosure 2 is therefore controlled by controlling the environment within the incubator. The incubator also  
10 controls the temperature and humidity within the enclosure. As previously described, the oxygen levels within the incubator are affected when the door to the incubator is open. The low flow rate of the circulation unit dampens this effect as the air within the enclosure is replaced at a low rate, allowing the incubator time to recover the required oxygen levels. The culture is therefore subject to the same rapid spikes in oxygen levels  
15 experienced within the open environment of the incubator. Nonetheless, the fluctuating oxygen levels within the incubator would result in a corresponding fluctuation of oxygen levels within the enclosure 2. To address this, a door sensor is provided that is arranged to sense when the door is opened and closed. The circulation unit also includes a controller that is arranged to determine when the door has been opened based on a  
20 signal the door sensor, and to control the circulation unit 40 to stop circulating air to the enclosure 2 when the door is opened to maintain the atmosphere within the enclosure 2. The controller also determines when the door has been closed on the basis of the door sensor. Once the door has been closed, the controller is programmed to keep the circulation deactivated for a pre-determined period corresponding to the time required  
25 for the atmosphere with in the incubator to return to the required oxygen level. This pre-programmed delay in re-activating the circulation unit 40 is based on observed data taken from the incubator.

Alternatively, the incubator may include a sensor or sensors configured to measure and  
30 monitor the gas levels within the incubator, and in particular to monitor the oxygen concentration within the incubator. The circulation unit 40 may be connected to the

atmospheric sensors of the incubator and the controller may be configured to operate the circulation unit 40 depending on information on the atmospheric conditions within the incubator received from the atmospheric sensors. The door sensor and/or atmospheric sensors therefore enable the atmospheric conditions, including oxygen level, temperature and humidity within the enclosure to be maintained when there is an undesired fluctuation in the atmospheric conditions within the incubator, commonly due to opening the incubator door.

When it is required to remove the enclosure from the incubator, the male valve couplings 36,38 of the enclosure 2 are disconnected from the female valve couplings 52,54. Upon disconnection the valve couplings 36,38 automatically close and seal the enclosure 2 to maintain the atmospheric conditions within the enclosure 2. The enclosure 2 may then be safely removed from the controlled atmosphere of the incubator. During transit and analysis the atmosphere within the sealed enclosure 2 is maintained at the required levels. In the case of microscope analysis, the glass base 10 enables the enclosure 2 to be placed directly on the stage of an inverted microscope, allowing the culture to be viewed from below using the microscope while the glass lid allows the culture to be lit from above. The culture is therefore able to be studied with a microscope without the need to remove the culture from the enclosure. The atmosphere within the enclosure is maintained through the analysis, enabling live-cell imaging of the culture under in vivo condition. The enclosure 2 is also sized such that it is able to fit within the interlock of a workstation, for example to enable the cell cultures to be accessed and for division. Once the enclosure is placed within the controlled atmosphere of the workstation the lid may be opened to allow access to the cell culture. Once work is completed, the lid is resealed and the atmosphere within the chamber is sealed under the same conditions as the workstation. The enclosure 2 remains sealed, and the atmosphere is maintained, until the enclosure 2 is re-connected to the circulation unit 40 within the incubator and the pre-set time period has passed within the closed incubator for the atmosphere to reach set point, at which point the circulation unit 40 restarts.

It will be appreciated that while the above described specific embodiment relates to studies under reduced oxygen conditions, the invention may applied to studies in any non- atmospheric environment. For example, the gas levels within the incubator may be controlled to maintain certain cultures at raised oxygen conditions, or to vary the level of other gases within the incubator. The controlled atmosphere apparatus 1 is able to ensure that whatever the storage environment within the incubator, the atmosphere within the enclosure 2 is maintained in the event that the incubator environment is lost due to door opening or for any other reason.

## CLAIMS

1. A portable storage apparatus for live biological material such as cell cultures or microorganisms, the portable storage apparatus comprising:
  - 5 a container having an opening for receiving biological material to be stored within the container;
  - a closure arranged to close and seal the opening of the container;
  - an inlet port arranged to allow gas to flow into the container from a gas supply and an outlet port arranged to allow gas to flow out of the container; and
  - 10 valve means configured to open the inlet port and outlet port in a first mode of operation to allow gas to circulate through the container by entering through the inlet and venting through the outlet, and to close the inlet port and outlet port in a second more of operation to seal the container and maintain the gaseous environment therein.
- 15 2. A portable storage apparatus according to claim 1 wherein the inlet port is configured for detachable connection to a gas source, and the valve means is configured to automatically close the inlet port when the inlet port is disconnected from the gas source.
- 20 3. A portable storage apparatus according to claim 1 or 2 wherein the valve means comprises an inlet valve coupling connected to the inlet port and an outlet valve coupling connected to the outlet port.
- 25 4. A portable storage apparatus according to claim 3 wherein the inlet valve coupling is configured for detachable connection to a gas source coupling and is configured to open when connected to said gas source coupling in the first mode of operation and to automatically close when detached from said gas source fluid coupling to seal the inlet port in the second mode of operation.

5. A portable storage apparatus according to claim 3 or 4 wherein the outlet valve coupling is configured for detachable connection to a complimentary mating coupling, and configured to open when connected to said fluid coupling in the first mode of operation and to automatically close when detached from said fluid couplings to seal the outlet port in the second mode of operation.
6. A portable storage apparatus according to any preceding claim wherein the container includes a base, a plurality of walls upstanding from the base, and an upper surface forming an enclosure, and at least a portion of at least one of the base and the upper surface comprises a transparent material arranged such that the contents of the container are visible through said transparent material.
7. A portable storage apparatus according to claim 6 wherein the base and the upper surface both comprise a transparent material arranged to such that the contents of the container are visible from above and below the container to enable the contents of the container to be studied using an optical instrument such as a microscope.
8. A portable storage apparatus according to claim 6 or 7 wherein the closure means comprises a lid formed at least in part of a transparent material and movable between an open position and a closed position.
9. A portable storage apparatus according to claim 8 further comprising securing means for holding the lid in the closed position.
10. A portable storage apparatus according to any preceding claim further comprising a gas circulation device for circulating gas through the container, the gas circulation device being detachably connectable to one of the inlet port and the outlet port to circulate gas through the container and the valve means being configured to open said inlet port or outlet port when the gas circulation means is connected and close said inlet port or outlet port when the gas circulation device is disconnected.

11. A portable storage apparatus according to claim 10 wherein the gas circulation device is connected to the inlet port and further comprising a vent connector configured to connect to the outlet port, wherein the valve means is arranged to  
5 open the outlet port when the vent connector is connected and to close the outlet when the vent connector is disconnected.
12. A portable storage apparatus according to claim 10 or 11 wherein the gas circulation device is configured to circulate ambient gas from the atmosphere  
10 surrounding the container.
13. A portable storage apparatus according to claim 12 further comprising a controller for controlling the gas circulation device, wherein in the first mode of operation the gas circulation device circulates ambient gas through the container and the  
15 controller is configured to stop the gas circulation device from circulating ambient gas through the container in response to a signal indicative of a variation in the ambient gas levels in order to maintain the atmosphere within the container.
14. A portable storage apparatus according to claim 13 wherein the controller is  
20 configured to restart the gas circulation device based on an input indicative of the ambient gas levels having returned to a desired level.
15. A portable storage apparatus according to any one of claims 13 or 14 further comprising at least one door sensor configured to determine when a door of a  
25 controlled atmosphere chamber within which the container is housed has been opened or closed, and wherein the controller is configured to stop the gas circulation device in response to a signal from the door sensor indicative of the door being opened.
- 30 16. A portable storage apparatus according to claim 15 wherein the controller is configured to restart the gas circulation device based on a signal from the door sensor indicative of the door being closed and an input indicative of the ambient gas levels having returned to a desired level following closure of the door.

17. A portable storage apparatus according to claim 16 wherein the input indicative of the ambient gas levels having returned to a desired level following closure of the door is provided by a timer configured to indicate when a predetermined time has elapsed following closure of the door.
- 5
18. A portable storage apparatus according to claim 17 wherein the input indicative of the ambient gas levels having returned to a desired level following closure of the door is provided by and a gas sensor configured to detect the levels of one or more gases within the controlled atmosphere chamber.
- 10
19. A storage apparatus according to any preceding claim further comprising a rapid purge device configured to connect to the inlet port of the container and supply gas to the container for a controlled period to cause a change in the gaseous environment within the container.
- 15
20. A method of storage of live biological material such as cell cultures or microorganisms, the method comprising:
- providing a storage apparatus according to any preceding claim;
  - 20 placing the biological material within the container;
  - connecting a gas circulation device to the inlet port of the container;
  - locating the container within a controlled gaseous environment and operating the gas circulation device to circulate ambient gas through the container.
- 25
21. A method according to claim 20 further comprising disconnecting the gas circulation device from the container and causing the valve means to close and seal the inlet and outlet ports to maintain the gaseous environment within the container the container and removing the sealed container from the controlled gaseous environment.
- 30
22. A method according to claim 21 further comprising the steps of detecting a condition indicative of a change in the gas levels within the controlled gaseous

environment and causing the gas circulation device to stop circulating air through the container in response to the detected change.

23. A method according to claim 22 wherein the controlled gaseous environment is a sealed chamber such as an incubator and the detected condition is the opening of a door to the sealed chamber.

24. A method according to claim 23 further comprising the steps of:  
detecting when the door has been closed; and  
reactivating the gas circulation device once it has been determined the gas levels within the controlled gaseous environment have returned to a desired level and/or wherein the gas circulation device is reactivated once a predetermined time period has elapsed following the closure of the door indicative of the gas levels within the controlled gaseous environment having returned to a desired level.

15

25. A method according to any one of claims 20 to 24 wherein a purge gas is supplied to the container prior to the container being placed in the controlled gaseous environment and connected to the gas circulation device.

20

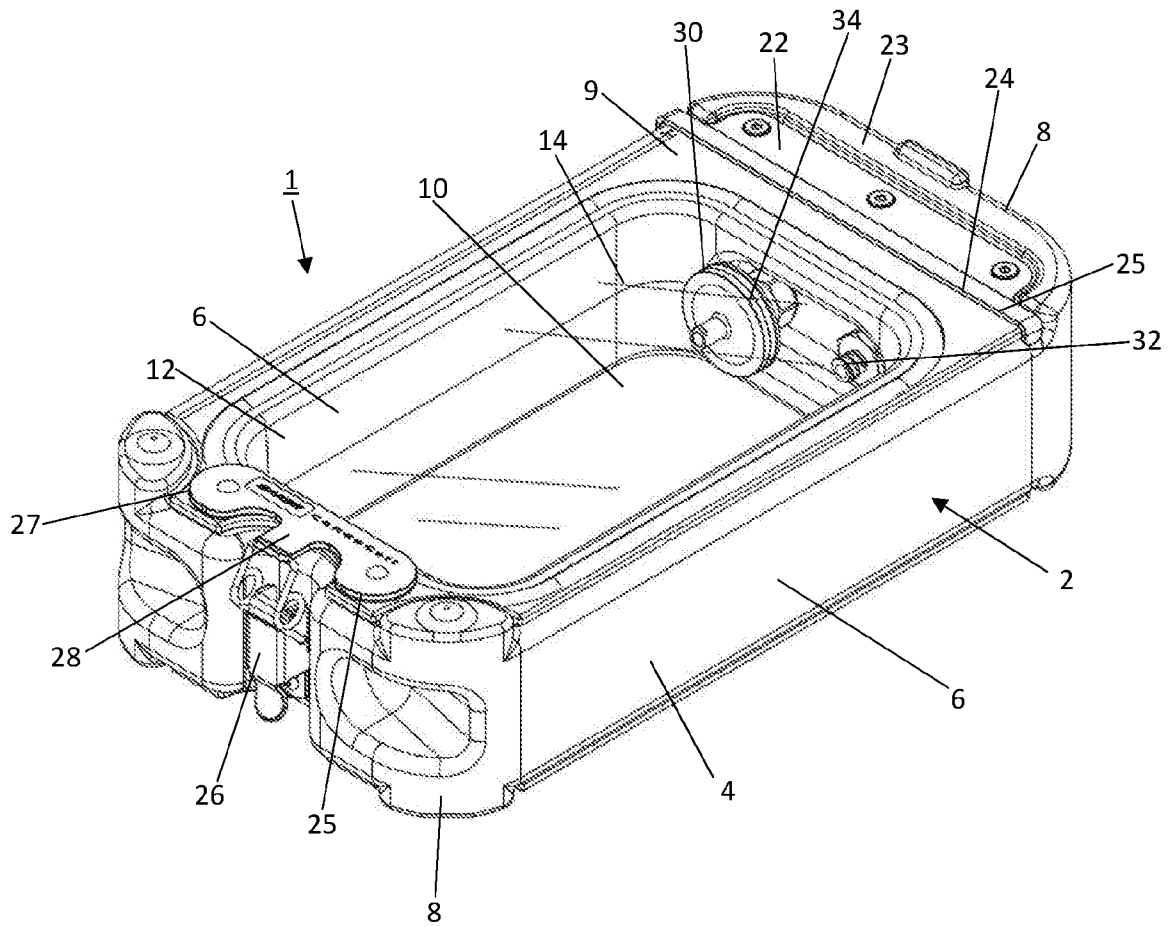
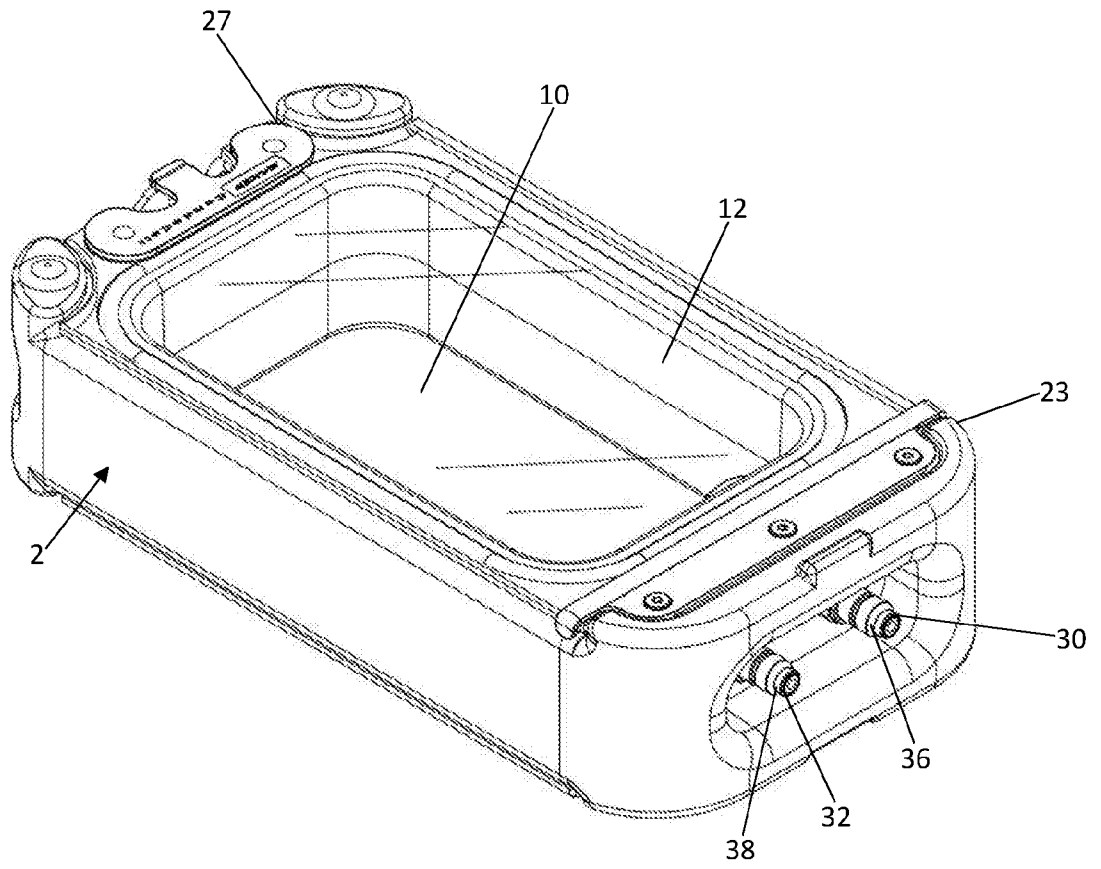


FIG. 1



**FIG. 2**

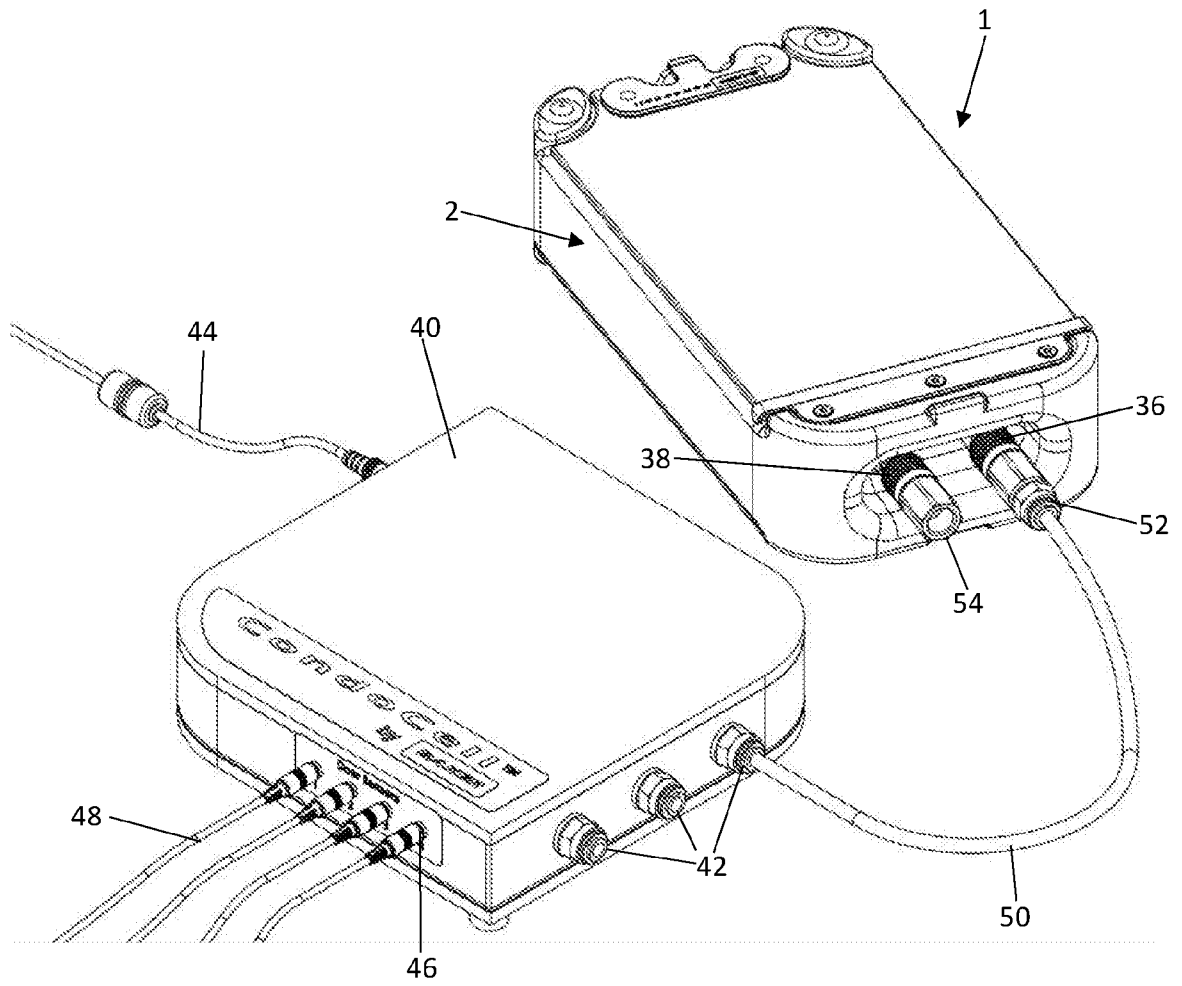


FIG. 3

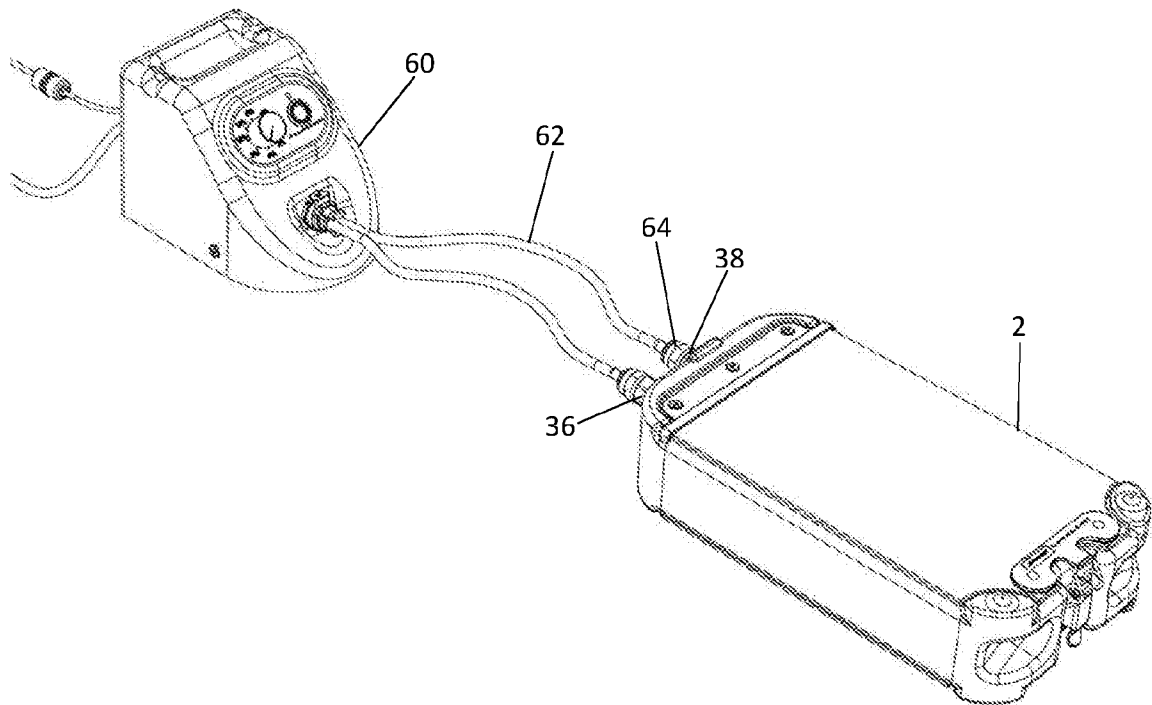


FIG. 4

INTERNATIONAL SEARCH REPORT

International application No  
PCT/GB2019/051800

A. CLASSIFICATION OF SUBJECT MATTER  
INV. C12M3/00 C12M1/12 C12M1/00  
ADD.  
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED  
Minimum documentation searched (classification system followed by classification symbols)  
C12M

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

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)  
EPO-Internal, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

| Category* | Citation of document, with indication, where appropriate, of the relevant passages  | Relevant to claim No. |
|-----------|---|-----------------------|
| X         | WO 01/40436 A2 (CELLOMICS INC [US];<br>FRIEDMAN ALEXANDER L [US] ET AL.)<br>7 June 2001 (2001-06-07)<br>page 11, lines 6-9; claims 1-5; figures<br>1-3<br>page 16, lines 9-12 | 1-25                  |
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| A         | -----<br>US 2017/058246 A1 (GRIER JR ROBERT DIXON<br>[US] ET AL) 2 March 2017 (2017-03-02)<br>claim 1; figure 1   | 1-25                  |

Further documents are listed in the continuation of Box C.

See patent family annex.

\* Special categories of cited documents :

|   |  |
|---|--|
| "A" document defining the general state of the art which is not considered to be of particular relevance  | "T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention  |
| "E" earlier application or patent but published on or after the international filing date   | "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone   |
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| "P" document published prior to the international filing date but later than the priority date claimed  |  |

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| Date of the actual completion of the international search<br><br>10 September 2019 | Date of mailing of the international search report<br><br>18/09/2019 |
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| Name and mailing address of the ISA/<br>European Patent Office, P.B. 5818 Patentlaan 2<br>NL - 2280 HV Rijswijk<br>Tel. (+31-70) 340-2040,<br>Fax: (+31-70) 340-3016 | Authorized officer<br><br>Jones, Laura |
|--|--|

# INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No

PCT/GB2019/051800

| Patent document cited in search report | Publication date | Patent family member(s) | Publication date         |
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|  |                  |                         | US 6365367 B1 02-04-2002 |
|  |                  |                         | WO 0140436 A2 07-06-2001 |
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| GB 2499372                             | A                | 21-08-2013              | NONE                     |
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| US 2017058246                          | A1               | 02-03-2017              | NONE                     |
| -----                                  |                  |                         |                          |