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(54) **Title**: HEAD-ONLY AND/OR WHOLE BODY INHALATION EXPOSURE CHAMBER

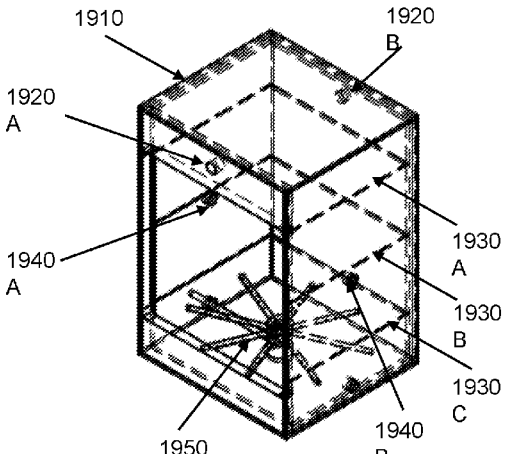


FIG. 19A

(57) **Abstract**: At least one embodiment of the invention provides a system comprising an inhalation exposure chamber and an aerosol delivery line connected to the inhalation exposure chamber. The aerosol delivery line is configured to produce a bi-directional and symmetrical presentation of aerosol to the inhalation exposure chamber. A laminar flow element is configured to create an antechamber where complete and turbulent mixing of the aerosol occurs. A radial exhaust and the laminar flow element enable laminar flow of the aerosol through the inhalation exposure chamber.

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Head-Only and/or Whole Body Inhalation Exposure Chamber

[001] This PCT international application claims benefit of U.S. Patent Application Serial No. 62/292,355 filed on 7 February 2016 with the U. S. Patent and Trademark Office, which is hereby incorporated by reference.

[002] This invention was made with support from the United States Department of the Army. The United States Government has certain rights in this invention.

I. Field of the Invention

[003] The present invention relates to a head-only inhalation exposure chamber for use on nonhuman primates. The present invention also relates to a whole body inhalation exposure system.

II. Background of the Invention

[004] Nonhuman primates (NHPs) are frequently challenged with infectious aerosols by head-only inhalation. In traditional head-only inhalation systems, a sedated NHP is positioned in a supine orientation and its head is inserted inside a rectangular chamber. The head is supported on a wire grid and a flexible piece of dental dam material is sealed around the animal's neck and between the animal's neck and the chamber. Aerosol is directed into the top of the chamber, flows past the animal's head, and is exhausted out of the bottom of the chamber. In examining this traditional method, testing with visible aerosols revealed that the unidirectional presentation of the aerosol resulted in persistent turbulent flow patterns throughout the chamber. Additionally, the asymmetrical exhaust manifold contributed to an abbreviated T99 and an increased dead space.

[005] The current art utilizes a uni-directional, asymmetrical aerosol delivery line and symmetrical exhaust line that result in an acutely short observed T99 (time to 99% equilibrium concentration) and a large amount of turbulent flow and dead space in the chamber. An observed T99 that is less than the theoretical T99 is indicative of plug flow (short circuiting) in the chamber. The dead space percentage is an indication of unequal distribution (non-homogeneity) of aerosol in the chamber.

[006] The various embodiments of this invention resolve several design weaknesses of the current art in head-only exposure chambers adversely affecting chamber operational efficiency.

III. Summary of the Invention

[007] Embodiments of the invention include a head-only chamber comprising an opposed normal aerosol delivery line, a laminar flow element and a radial exhaust. The opposed normal delivery line produces a bi-directional and symmetrical presentation of aerosol to the chamber. The laminar flow element creates an ante-chamber where complete, turbulent mixing occurs. The laminar flow element and radial exhaust enable laminar flow of the aerosol through the chamber and past the test system's head. Improvements in chamber operational efficiency have been

confirmed using video and pictures of a dense aerosol in the chamber, by calculation of T99 and determination of the aerosol temporal stability of in the chamber.

[008] At least one embodiment provides a system having an inhalation exposure chamber. The inhalation exposure chamber can include four sidewalls, where one of the sidewalls includes an aperture dimensioned to receive a primate head therethrough. The system can include four cages in the inhalation exposure chamber. An aerosol delivery line can be connected to the inhalation exposure chamber, where the aerosol delivery line is configured to produce a bi-directional and symmetrical presentation of aerosol to the inhalation exposure chamber. The aerosol delivery line can be centrally located at the top of the inhalation exposure chamber. The aerosol delivery line can be a 12.0 inch long pipe having a 1.5 inch diameter. The aerosol delivery line can include two 0.25 inch wide by 8.0 inch long slots positioned 180° apart.

[009] In at least one embodiment, the system includes a laminar flow element configured to create an ante-chamber where complete and turbulent mixing of the aerosol occurs, where the laminar flow element is located below the aerosol delivery line. The laminar flow element can include a sheet having a plurality of perforations. For example, the laminar flow element includes an 8.0 inch x 8.0 inch x 0.735 inch honeycomb. In another example, the laminar flow element includes an 8.0 inch x 8.0 inch x 0.0156 inch sheet with 0.25 inch perforations. In yet another example, the laminar flow element includes an 8.0 inch x 8.0 inch x 0.0625 inch sheet with 0.0625 inch diameter perforations. The system can include at least one additional laminar flow element including a sheet having a plurality of perforations.

[0010] The system can include a radial exhaust, where the radial exhaust and the laminar flow element can enable laminar flow of the aerosol through the inhalation exposure chamber. The radial exhaust can be centrally located at the bottom of the inhalation exposure chamber. The radial exhaust can include eight tubes positioned radially around a central hub, where each tube can include a sealed distal end and a plurality of lateral holes. The radial exhaust can include four first tubes having a first length and four second tubes having a second length, where the second length is greater than the first length. The radial exhaust can include 0.375 inch diameter tubes positioned radially around a 1.5 inch diameter x 2.0 inch high central hub. Each of the tubes can include a sealed distal end and five 0.125 inch lateral apertures. The system can include one or more sample collection ports located at a midpoint of a wall of the inhalation exposure chamber.

[0011] Given the following enabling descriptions of the drawings, the invention should be evident to one ordinarily skilled in the art.

IV. Brief Description of the Drawings

[0012] FIG. 1 is an overhead plan view of one embodiment of the radial exhaust of this invention.

[0013] FIG. 2 is a photograph of turbulent and rotational chamber flow.

[0014] FIG. 3 is a photograph of turbulent and rotational chamber flow showing wall sampler location.

[0015] FIG. 4 is a photograph of one embodiment of the chamber assembly.

[0016] FIG. 5 is a photograph of one embodiment of the aerosol delivery line inlets.

[0017] FIG. 6 is a photograph of one embodiment of the expanded metal laminar flow elements.

[0018] FIG. 7 is a photograph of one embodiment of the honeycomb laminar flow element.

[0019] FIG. 8 is a photograph of one embodiment of the plastic laminar flow element.

[0020] FIG. 9 is a photograph of one embodiment of the head-only chamber radial exhaust.

[0021] FIG. 10 is a schematic diagram of one embodiment of the modified chamber test system.

[0022] FIG. 11 is a photograph of the turbulent aerosol flow in antechamber.

[0023] FIG. 12 is a photograph of a well-mixed chamber at steady state equilibrium.

[0024] FIG. 13 is a schematic diagram of an embodiment of the novel head-only chamber test system.

[0025] FIG. 14 is a side-view photograph of an embodiment of the novel head-only exposure chamber.

[0026] FIG. 15 is a top-view photograph of an embodiment of the novel opposed, normal, bi-directional aerosol delivery line.

[0027] FIG. 16 is a photograph of time lapse comparison of low dynamics between the prior art exposure chamber and an embodiment of the present invention.

[0028] FIG. 17 is a photograph of laminar flow.

[0029] FIG. 18 illustrates an inhalation exposure chamber system according to an embodiment of the invention.

[0030] FIG. 19A is a top perspective view of an inhalation exposure chamber system according to another embodiment of the invention. FIG. 19B is a top view of the inhalation exposure chamber system illustrated in FIG. 19A. FIG. 19C is a side view of the inhalation exposure chamber system illustrated in FIG. 19A. FIG. 19D is a side cross-sectional view of the inhalation exposure chamber system illustrated in FIG. 19A.

[0031] FIG. 20 is a flow diagram illustrating a method for controlling aerosol respiratory exposure according to an embodiment of the invention.

[0032] FIG. 21 is a flow diagram illustrating a method for controlling aerosol respiratory exposure according to an embodiment of the invention.

[0033] FIG. 22 illustrates a computer program product and computer implementation according to an embodiment of the invention.

[0034] FIG. 23 illustrates a schematic of one embodiment of the computer system of this invention.

[0035] FIG. 24 illustrates a schematic of one embodiment of the computer system of this invention.

V. Detailed Description of the Drawings

[0036] In order to provide a clear and consistent understanding of the specification and claims, including the scope to be given such terms, the following definitions are provided.

[0037] As used herein, "communication means" refers to means for conveying digital information, synchronously and asynchronously, as an electromagnetic signal from one electronic device to another, to include without limitation: Universal Serial Bus (USB) ports, FIREWIRE TM, wired technology, optical fiber, wireless (including BLUETOOTH TM, ANT + TM), analog-to-digital conversion (e.g. through a microphone), cellular, microwave, satellite, infrared, Large Area Networks, Local Area Networks, Internet/Web, and radio.

[0038] The term "data-entry means" refers to any device for accepting data from a subject, to include without limitation: keyboards, keypads, touchpads, tactile sensors, capacitive sensing devices, and conductance sensing devices, regardless of whether the data entry means requires the use of a stylus or similar writing implements.

[0039] The term "statistical methods" refers to both descriptive and inferential statistics well known in the art, to include without limitation those statistical methods, tests (parametric and non-parametric) and procedures particular to the fields of biostatistics and psychological statistics.

[0040] The term "storage means" refers to any means for storing data that is accessible by an electronic digital computer to include, without limitation: volatile memory (RAM, SRAM, DRAM, Z-RAM, TTRAM, A-RAM and ETA RAM) and non-volatile memory (ROM, flash memory, magnetic computer storage devices and optical discs), whether or not the data is stored in a database.

[0041] As used herein, the term "subject" means any living organism, including humans, and mammals.

[0042] As will be appreciated by one skilled in the art based on this disclosure, aspects of the present invention may be embodied as a system, method or computer program product. Accordingly, aspects of the present invention may take the form of an entirely hardware embodiment, a processor operating with software embodiment (including firmware, resident software, micro-code, etc.) or an embodiment combining software and hardware aspects that may all generally be referred to herein as a "circuit," "module" or "system." Furthermore, aspects of the present invention may take the form of a computer program product embodied in one or more computer readable medium(s) having computer readable program code embodied thereon.

[0043] Any combination of one or more computer readable medium(s) may be utilized. The computer readable medium may be a computer readable signal medium or a computer readable

storage medium. A computer readable storage medium may be, for example, but not limited to, an electronic, magnetic, optical, electromagnetic, infrared, or semiconductor system, apparatus, or device, or any suitable combination of the foregoing. More specific examples (a non-exhaustive list) of the computer readable storage medium would include the following: an electrical connection having one or more wires, a portable computer diskette, a hard disk, a random access memory (RAM), a read-only memory (ROM), an erasable programmable read-only memory (EPROM or Flash memory), an optical fiber, a portable compact disc read-only memory (CD-ROM), an optical storage device, a magnetic storage device, or any suitable combination of the foregoing. In the context of this disclosure, a computer readable storage medium may be any tangible medium that can contain, or store a program for use by or in connection with an instruction execution system, apparatus, or device.

[0044] A computer readable signal medium may include a propagated data signal with computer readable program code embodied therein, for example, in baseband or as part of a carrier wave. Such a propagated signal may take any of a variety of forms, including, but not limited to, electro-magnetic, optical, or any suitable combination thereof. A computer readable signal medium may be any computer readable medium that is not a computer readable storage medium and that can communicate, propagate, or transport a program for use by or in connection with an instruction execution system, apparatus, or device.

[0045] Computer program code for carrying out operations for aspects of the present invention may be written in any combination of one or more programming languages, including an object oriented programming language such as Java, Smalltalk, C++, C#, Transact-SQL, XML, or the like and conventional procedural programming languages, such as the "C" programming language or similar programming languages. The program code may execute entirely on the user's computer, partly on the user's computer, as a stand-alone software package, partly on the user's computer and partly on a remote computer or entirely on the remote computer or server. In the latter scenario, the remote computer may be connected to the user's computer through any type of network, including a local area network (LAN) or a wide area network (WAN), or the connection may be made to an external computer (for example, through the Internet using an Internet Service Provider).

[0046] Aspects of the present invention are described herein with reference to flowchart illustrations and/or block diagrams of methods, apparatus (systems) and computer program products according to embodiments of the invention. It will be understood that each block of the flowchart illustrations and/or block diagrams, and combinations of blocks in the flowchart illustrations and/or block diagrams, can be implemented by computer program instructions. These computer program instructions may be provided to a processor of a general purpose computer,

special purpose computer, or other programmable data processing apparatus to produce a machine, such that the instructions, which execute with the processor of the computer or other programmable data processing apparatus, create means for implementing the functions/acts specified in the flowchart and/or block diagram block or blocks.

[0047] These computer program instructions may also be stored in a computer readable medium that can direct a computer, other programmable data processing apparatus, or other devices to function in a particular manner, such that the instructions stored in the computer readable medium produce an article of manufacture including instructions which implement the function/act specified in the flowchart and/or block diagram block or blocks.

[0048] The computer program instructions may also be loaded onto a computer, other programmable data processing apparatus, or other devices to cause a series of operational steps to be performed on the computer, other programmable apparatus or other devices to produce a computer implemented process such that the instructions which execute on the computer or other programmable apparatus provide processes for implementing the functions/acts specified in the flowchart and/or block diagram block or blocks.

[0049] For purposes of clarity of disclosure, and not by way of limitation, the detailed description of the invention is described by way of the demonstrative examples discussed below, and as further described in FIGS. 1-24.

[0050] FIG. 1 is an overhead plan view of one embodiment of the radial exhaust 100 of this invention. The radial exhaust 100 can include eight tubes 110a – 110h positioned radially around a central hub 120. Each tube can include a sealed distal end and a plurality of lateral holes (not shown). Aerosol is delivered to the chamber bi-directionally using an opposed normal inlets from a common feed. A laminar flow element is placed under the inlet nozzles to create a mixing ante-chamber; and a radial exhaust manifold is implemented to improve flow symmetry. The radial exhaust contributes to less dead space and a T99 that more closely approximated the theoretical T99.

[0051] The head-only inhalation exposure chamber design currently in use at USAMRIID was evaluated using flow visualization techniques and industry standard aerosol tests to ascertain efficiency. The head-only chamber utilizes a 1.5 inch diameter by 12.0 inch long, uni-directional aerosol delivery line located at the top of the chamber, an aerosol sample collection port located at the midpoint of one wall of the chamber and a 1.5 inch diameter by 12.0 inch long opposed normal exhaust line. The aerosol delivery and exhaust lines are centrally located, continuous and have two 0.25 inch wide by 8.0 inch long slots positioned 180° apart. FIGS. 2 and 3 show the results of flow visualization tests that identified highly turbulent and rotational flow throughout the chamber that was directionally biased toward the location of the site where the nonhuman

primate head rests (**FIG. 2**) and showed inefficient aerosol mixing at the aerosol sample collection location (**FIG. 3**). Tests using aerosol generated from a saline solution indicated that the measured time-to-99% stead-state aerosol concentration (T99) was less than the theoretical T99 which may be indicative of plug flow. Additionally, the percent dead space, a metric used to estimate chamber mixing efficiency (homogeneity), as measured from the wall sample collection port was 47.7%. FIG. 4 also illustrates a perforated metal sheet between the laminar flow element and the radial exhaust, which can be used to support the test head. In at least one embodiment, the head-only chamber in the system is replaced with a whole-body chamber.

[0052] Based on flow visualization images and data collected from this head-only chamber, design modifications for the aerosol delivery line (inlet), a laminar flow element, the exhaust line were devised. A prototype chamber that included a simulated nonhuman primate head was assembled as shown in **FIG. 4**. The following modifications were tested:

[0053] An opposed normal, bi-directional aerosol delivery line strategy was implemented. One configuration used the existing 1.5 inch diameter continuous aerosol delivery line which was slotted on each side. The slots were 0.25 inches in width and located perpendicularly to the vertical axis of the chamber. Other configurations included dual, non-continuous 1.5 inch diameter stainless steel tubing, dual non-continuous 0.5 inch diameter inlets and dual non-continuous 0.125 inch wide by 1.0 inch long slotted inlets. Aerosol Delivery Line Inlets tested are shown in **FIG. 5**.

[0054] Three types of laminar flow elements were tried below the aerosol delivery line forming an antechamber. Laminar flow elements tested included an 8.0 x 8.0 x 0.735 inch aluminum honeycomb (FIG. 6), an 8.0 x 8.0 x 0.0156 inch thick expanded metal sheet with 0.25 inch perforations (FIG. 7), and an 8.0 x 8.0 x 0.0625 inch thick plastic sheet with 0.0625 inch diameter perforations (FIG. 8).

[0055] The modified exhaust line consisted of eight 0.375 inch diameter stainless steel tubes positioned radially around a 1.5 inch diameter x 2.0 inch (length) Plexiglas central hub. The distal end of each radial tube was sealed. Five 0.125 inch diameter jets were drilled along the length of each radial tube. In order to equilibrate the relative area each jet would exhaust, their locations along the length of the tube were calculated as:

$$r_m = l\sqrt{\frac{2m-1}{2n}} \qquad 0 < m \leq n$$

Where: r = radial location
m = number of jets to be located
n = total number of jets
l = length of the tube

A single 0.375 inch diameter stainless steel tube was connected to the internal cavity of the central hub and supplied vacuum exhaust. For this study, the jets in each radial tube were aligned in a downward orientation. The modified radial exhaust is shown in **FIG. 9**.

[0056] Flow visualization tests identified highly turbulent flow throughout the chamber that were directionally biased toward the location of the site where the NHP head rests and inefficient aerosol mixing at the aerosol sample collection location. Tests using aerosol generated from a saline solution indicated that the measured time-to-99% stead-state aerosol concentration (T99) was less than the theoretical T99, which may be indicative of plug flow. Additionally, the percent dead space, a metric used to estimate chamber mixing efficiency (homogeneity), as measured from the wall sample collection port was 47.7%.

[0057] Each opposed normal delivery line configuration tested showed an improved distribution of the aerosol in the chamber as compared to the original, uni- directional design. The 1.5 inch diameter, slotted continuous inlet provided turbulent mixing in the antechamber, but produced a profound plug flow region in the center of the chamber and uneven mixing in the distal areas. The dual, non-continuous 1.5 inch diameter stainless steel tubing provided the least amount of turbulent mixing in the antechamber. The dual, non-continuous 0.5 inch diameter inlets and dual non-continuous 0.125 inch wide by 1.0 inch long slotted inlets provided the best turbulent mixing in the antechamber with the 0.5 inch diameter round jets providing the best mixing.

[0058] The aluminum honeycomb did not provide retention time of the aerosol in the antechamber and produced significant plug flow into the main chamber. The plastic sheet with 0.0625 inch perforations provided for adequate turbulent mixing in the antechamber and laminar flow in the main chamber, it restricted the flow of aerosol to the point that large globules of aerosol would plunge through the interior. The expanded metal sheet with 0.25 inch perforations performed the best of the laminar flow elements tested. It allowed for adequate retention time resulting in turbulent mixing in the antechamber and laminar flow in the main chamber. However, the 0.25 inch perforations restricted the flow in the center of the antechamber, resulting in the appearance of plug flow in the center of the main chamber. Nonetheless, the combination of a dual, non-

continuous 0.5 in diameter jet inlet with the expanded metal sheet with 0.25 inch perforations provided for the best overall mixing, laminar flow, stability and emptying observed during visual testing.

[0059] The radial exhaust resulted in improved laminar flow during chamber filling and chamber evacuation.

[0060] FIG. 10 is a schematic diagram of one embodiment of the modified chamber test system. Compressed air can flow into a gas flow controller and a nebulizer. Ambient air can flow through a HEPA filter. The ambient air and compressed air from the nebulizer can flow into a radial mixer. The radial mixer can receive input from a gas flow controller. Air from the radial mixer can flow into a quadtrack diffusion dryer and into an opposed normal delivery line. The air can enter into the exposure chamber, out and through a gas flow controller, and into a vacuum pump.

[0061] Flow visualization photographs of the modified head-only inhalation exposure chamber demonstrate a remarkable improvement in chamber flow dynamics. **FIG. 11** shows turbulent mixing in the antechamber from the dual, non-continuous 0.5 inch diameter inlets. **FIG. 17** demonstrates laminar flow of the aerosol as it penetrates the laminar flow element. **FIG. 12** shows a well-mixed chamber at steady state equilibrium.

[0062] In one embodiement of the invention, a standard push-pull orientation was constructed using Alicat Gas Flow Controllers MCRS-100SLPM-TFT, S/N 63412 (vacuum exhaust), MC-30SLPM-D S/N 25245 (radial mixer) and MC-30SLPM-TFT S/N 94001 (nebulizer). A Model 11310, Lot D328105 Hope Nebulizer was filled with 30 mL of Silahydrocarbon MLO 86-348. Total exhaust volumetric flow through the chamber was set at 16.0 L/min. The volumetric flow rate of the radial mixer was set at 5.0 L/min and the volumetric flow rate through the nebulizer was set at 7.5 L/min. Aerosol was generated into the plenum until the concentration reached an approximate (visual) equilibrium and then stopped. The chamber aerosol concentration exponential rise, equilibrium and exponential decay were photographed and videotaped for subsequent analysis. A schematic of the modified chamber test system is shown in **FIG. 13.** A picture of one embodiment of the novel head-only exposure chamber is shown in **FIG. 14**.

[0063] In another embodiment of the invention, the opposed normal, bi-directional aerosol delivery line strategy was implemented. One configuration used the existing 1.5 inch diameter continuous aerosol delivery line which was slotted on each side. Aerosol was delivered to each end of the delivery line. Other configurations included dual, non-continuous 1.5 inch diameter stainless steel tubing, dual non-continuous 0.5 inch diameter inlets and dual non-continuous 0.125 inch wide by 1.0 inch long slotted inlets. A picture of a 0.375 inch noncontiguous opposed aerosol delivery lines are shown in FIG. 15.

[0064] In another embodiment of the invention, three types of laminar flow elements were tested. A 0.25 inch thick aluminum honeycomb did not provide retention time of the aerosol in the ante-chamber and produced significant plug flow into the main chamber. A 0.0625 inch plastic sheet with 0.0625 inch perforations provided for adequate turbulent mixing in the ante-chamber and laminar flow in the main chamber, however, it restricted the flow of aerosol to the point that large globules of aerosol would plunge through the interior. An expanded metal sheet with 0.25 inch perforations performed the best of the laminar flow elements tested. It allowed for adequate retention time resulting in turbulent mixing in the ante-chamber and laminar flow in the main chamber. However, the 0.25 inch perforations restricted the flow in the center of the ante-chamber resulting in the appearance of plug flow in the center of the main chamber. Nonetheless, the combination of a dual, non-continuous 0.375 in diameter aerosol inlet and the expanded metal sheet with 0.25 inch perforations provided for the best overall mixing, laminar flow, stability and emptying observed during visual testing.

[0065] The modified exhaust line consisted of eight 0.375 inch diameter stainless steel tubes positioned radially around a 1.5 inch diameter x 2.0 inch (length) Plexiglas central hub. The distal end of each radial tube was sealed. Five 0.125 inch diameter jets were drilled along the length of each radial tube. In order to equilibrate the relative area each jet would exhaust, their locations along the length of the tube were calculated as:

$$r_m = l\sqrt{\frac{2m-1}{2n}} \qquad 0 < m \leq n$$

Where: r = radial location
m = number of jets to be located
n = total number of jets
l = length of the tube

[0066] A single 0.375 inch diameter stainless steel tube was connected to the internal cavity of the central hub and supplied vacuum exhaust. For this study, the jets in each radial tube were aligned in a downward orientation.

[0067] As seen in FIG. 16, a time lapse comparison of flow dynamics for (A) original chamber design and (B) chamber design with modifications. The original chamber design geometry produces large amounts of void space, even at steady state (Panel A3). The modified chamber design has a shorter time to steady state, and aerosol distribution is more uniform at steady state (panel B3).

[0068] Chamber homogeneity (% Dead Space) in the current style head-only exposure chamber was determined using the equation:

$$\left(1 - \frac{T_{measured}}{T_{theoretical}}\right) \times 100\%$$

Sample ID	V (L)	Q (L/min)	$T_{theoretical}$ (sec)	k (sec^{-1})	$T_{measured}$ (sec)	Dead Space (%)
1w	16.256	16.000	60.9600	0.0304	32.8696	46.1
2w	16.256	16.000	60.9600	0.0285	35.1246	42.4
3w	16.256	16.000	60.9600	0.0363	27.5614	54.8
			MEAN	0.0317	31.8519	47.7
			STDEV	0.0041	3.8829	6.4
			% CV	12.8	12.2	13.3

Sample ID	V (L)	Q (L/min)	$T_{theoretical}$ (sec)	k (sec^{-1})	$T_{measured}$ (sec)	Dead Space (%)
1c	16.256	16.000	60.9600	0.0382	26.1536	57.1
2c	16.256	16.000	60.9600	0.0500	20.0182	67.2
3c	16.256	16.000	60.9600	0.0402	24.8825	59.2
4c	16.256	16.000	60.9600	0.0441	22.6783	62.8
			MEAN	0.0431	23.4331	61.6
			STDEV	0.0052	2.6915	4.4
			% CV	12.0	11.5	7.2

[0069] Triplicate aerosol samples were collected from either a wall sampling location (w) or from a sample probe positioned at the approximate location of the animal's nose (c).

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ID	V (L)	Q (L/min)	$T_{theoretical}$ (sec)	k (sec^{-1})	$T_{measured}$ (sec)	Dead Space (%)
1	16.256	16.000	60.9600	0.0209	47.8469	21.5
2	16.256	16.000	60.9600	0.0230	43.4783	28.7
3	16.256	16.000	60.9600	0.0251	39.8406	34.6
			MEAN	0.0230	43.7219	28.3
			STDEV	0.0021	4.0087	6.6
			% CV	9.1	9.2	23.3

Triplicate aerosol samples were collected from a sample probe positioned at the approximate location of the animal's nose (c).

	Current Chamber Design T_{99} (min)	Modified Chamber Design T_{99} (min)
	2.9	3.7
	2.8	3.3
	2.7	3.1
MEAN	2.8	3.4
STDEV	0.1	0.3

[0070] The observed time-to-99% (T99) steady state aerosol concentration was determined in the current and modified head-only exposure chambers using 2 μm (nominal) polystyrene latex microspheres. Triplicate samples were collected from each chamber style. The mean T99 for the current head-only style exposure chamber was 2.8 ± 0.1 min. The mean T99 for the modified head-only style exposure chamber was 3.4 ± 0.3 min. The theoretical T99 for each style of chamber, as calculated using the equation given by Cheng and Moss (1), was 4.63 min.

[0071] Redesigning the aerosol exposure chamber with a turbulent mixing antechamber and a radial exhaust to facilitate fluid dynamic symmetry in the direction of flow within the chamber resulted in a more efficient design. Incorporating the new design elements led to a decrease in dead space and a measured T99 that more closely approximated the theoretical T99. Testing with a visible aerosol confirmed a turbulent mixing zone in the antechamber and laminar aerosol flow throughout the rest of the chamber.

[0072] Additional optional embodiments to the above-described embodiments include, but are not limited to: modifications of the dual, non-continuous opposed normal aerosol delivery line to maximize turbulent mixing; modifications may include slightly offsetting the jets and adjusting the length of the tubes; modifications of the laminar flow element will include adjusting the diameter of the perforations in the expanded metal sheet and overlaying multiple sheets to restrict flow enough to allow for optimal mixing while providing laminar flow with minimized globule formation; and, any combination of these modifications. Tests with the radial exhaust will include different orientations of the jets in the radial tubes. In at least one embodiment, additional tests include the addition of a 0.25 inch diameter stainless steel sample probe and ventilation and temperature regulation of the simulated nonhuman primate head to determine the effects on chamber laminar flow and mixing.

[0073] **FIG. 18** illustrates a system 1800 according to an embodiment of the invention, where the system 1800 includes an inhalation exposure chamber 1810, aerosol delivery lines 1820A and 1820B, a laminar flow element 1830, an aerosol sample collection port 1840 (also referred to

herein as a sampling tube), a radial exhaust 1850, cage 1860A, cage 1860B, cage 1860C, and cage 1860D. Each cage can include an upper wall, a bottom wall, and four side wall. The sidewalls can be formed from a transparent material (e.g., plastic, glass). The upper wall and the bottom wall can be formed from a transparent material or a perforated sheet. The inhalation exposure chamber 1810 can include a door 1812 and handle 1814.

[0074] **FIG. 19A** is a top perspective view of an alternative inhalation exposure chamber system 1900 according to another embodiment of the invention. **FIG. 19B** is a top view of the inhalation exposure chamber system illustrated in **FIG. 19A**. **FIG. 19C** is a side view of the inhalation exposure chamber system illustrated in **FIG. 19A**. **FIG. 19D** is a side cross-sectional view of the inhalation exposure chamber system illustrated in **FIG. 19A**. The inhalation exposure chamber system 1900 can include an inhalation exposure chamber 1910, aerosol delivery ports 1920A and 1920B, laminar flow element 1930A, 1930B and 1930C, aerosol sample collection ports 1940A and 1940B, and a radial exhaust 1950.

[0075] At least one embodiment of the invention provides an Integrated Modular Bioaerosol Respiratory Exposure System (IMBRE) that includes a pneumatic/electronic control box and accompanying control computer program instructions that can utilize existing aerosol exposure chambers (e.g., COTS nose-only exposure chambers). The IMBRE (also referred to herein as the "system") can interface with particle sizing and counting devices that provide data streams to the computer program instructions, which can enable the control of exposure duration based on aerosol particle count (which can be correlated with presented dose).

[0076] Additionally, the system can count aerosol particles and correlate particle counts with presented dose and determine particle size distribution. This feature can be used to determine the time required for "dose-calculated" exposures. More specifically, aerosol particles can be diluted and collected from one sample tube using an in-line aerosol particle counter. Aerosol particles can be collected from a second sample tube using filters and liquid impingers. Samples from each mask can be collected over different lengths of time. The filter samples can be weighed and a mass per unit volume aerosol concentration calculated. The liquid impinger samples can be microbiologically assayed and a count per unit volume aerosol concentration can be calculated. The mass and count per unit volume aerosol concentrations can be plotted against the count data from the in-line particle counter and a mathematical regression correlation can be determined. The mathematical correlation can be used to determine dosimetry of subsequent exposures.

[0077] At least one embodiment of the invention provides a compact and modular system that runs aerosol exposures using a head-only exposure chamber or a whole-body exposure chamber. The system can expose animal experimental models to biological, chemical, and radiological agents for pathogenesis and/or toxicity studies for therapeutic, vaccine, and/or prophylactic development.

The system can also be used to test aerosolized drug formulations in animal models to facilitate the development of inhalable drugs within the pharmaceutical industry.

[0078] In at least one embodiment, the system provides improved spray accuracy, including integrated real-time plethysmography, particle-based tracking of concentration (oronasal mask module), real-time adjustment of exposure time, and/or continuous spray improvement based on previous run data across all systems (animal, agent, agent media, etc.). The system may also provide spray analysis and reporting, integrated calibration of all components, and/or system health detection and analysis. Specifically, the system may detect the likelihood of component failure before any run and ensure that all components and/or accessories are in calibration before each spray, where component heath and calibration can be transferable to other systems.

[0079] **FIG. 20** is a flow diagram illustrating a method for controlling aerosol respiratory exposure according to an embodiment of the invention, where the exposure chamber includes a differential pressure transmitter. The differential pressure transmitter can detect positive pressure pulses and negative pressure pulses in the exposure chamber 2010. Specifically, a differential pressure transducer can generate a pressure differential waveform and transmit the pressure differential waveform to a processor for further calculation.

[0080] Plethysmography software in the processor can determine tidal volume and frequency based on the detected positive and negative pressure pulses 2020. The processor can also calculate the respiratory minute volume 2030, which may be a product of the tidal volume and the frequency. In at least one embodiment, the respiratory minute volume is an average integral over a respiratory waveform, where the respiratory waveform is measured, integrated over time, and averaged.

[0081] The processor can calculate the cumulative inspired volume 2040, which may be a product of the respiratory minute volume and an exposure duration. The cumulative inspired volume can be calculated by integrating the respiratory waveform over time in real-time until the desired presented dose is achieved. The processor can also calculates the desired inhaled volume of aerosol with the cumulative inspired volume and the theoretical aerosol concentration 2050. The total presented dose can be an integral of the product of a concentration function and a volume function over time when aerosol concentration changes over time.

[0082] **FIG. 21** is a flow diagram illustrating a method for controlling aerosol respiratory exposure according to an embodiment of the invention. The method can also determine aerosol particle size distribution (mass median aerodynamic diameter (μm)) and geometric standard deviation or particle counts. An in-line aerosol particle counter can collect first aerosol particles from a first aerosol sample collection port for a first time duration 2110. A second aerosol sample collection port can collect second aerosol particles using one or more filters and one or more liquid impingers

2120. The second aerosol particles can be collected for a second time duration that is different from the first time duration.

[0083] In at least one embodiment, the weights of the filters in the second aerosol sample collection port is determined 2130; and, a mass per unit volume aerosol concentration is calculated based on the weights of the filters 2140. Second aerosol particles from the liquid impinger can be microbiologically assayed 2150; and, a count per unit volume aerosol concentration can be calculated based on the microbiological assay 2160.

[0084] The mass per unit volume aerosol concentration and the count per unit volume aerosol concentration can be plotted against count data from the in-line aerosol particle counter to correlate a presented dose with the particle count 2170. A mathematical regression correlation can be determined based on the plotting of the mass per unit volume aerosol concentration and the count per unit volume aerosol concentration against the count data from the in-line aerosol particle counter 2180. Dosimetry of subsequent exposures can be determined based on the mathematical regression correlation 2190.

[0085] Referring now to **FIG. 22**, a representative a hardware environment for practicing at least the first embodiment of the invention is depicted. This schematic drawing illustrates a hardware configuration of an information handling/computer system in accordance with at least one embodiment of the invention. The system comprises at least one processor or central processing unit (CPU) **2210**. The CPUs **2210** are interconnected with system bus **2212** to various devices such as a random access memory (RAM) **2214**, read-only memory (ROM) **2216**, and an input/output (I/O) adapter **2218**. The I/O adapter **2218** can connect to peripheral devices, such as disk units **2211** and tape drives **2213**, or other program storage devices that are readable by the system. The system can read the inventive instructions on the program storage devices and follow these instructions to execute the methodology of at least one embodiment of the invention. The system further includes a user interface adapter **2219** that connects a keyboard **2215**, mouse **2217**, speaker **2224**, microphone **2222**, and/or other user interface devices such as a touch screen device (not shown) to the bus **2222** to gather user input. Additionally, a communication adapter **2220** connects the bus **2222** to a data processing network **2225**, and a display adapter **2221** connects the bus **2222** to a display device **2223** which may be embodied as an output device such as a monitor, printer, or transmitter, for example.

[0086] The flowchart and block diagrams in the FIGS. illustrate the architecture, functionality, and operation of possible implementations of systems, methods and computer program products according to various embodiments of the present invention. In this regard, each block in the flowchart or block diagrams may represent a module, segment, or portion of code, which comprises one or more executable instructions for implementing the specified logical function(s).

It should also be noted that, in some alternative implementations, the functions noted in the block may occur out of the order noted in the FIGS. For example, two blocks shown in succession may, in fact, be executed substantially concurrently, or the blocks may sometimes be executed in the reverse order, depending upon the functionality involved. It will also be noted that each block of the block diagrams and/or flowchart illustration, and combinations of blocks in the block diagrams and/or flowchart illustration, can be implemented by special purpose hardware-based systems that perform the specified functions or acts, or combinations of special purpose hardware and computer instructions.

[0087] The terminology used herein is for the purpose of describing particular embodiments only and is not intended to be limiting of the invention. As used herein, the singular forms “a”, “an” and “the” are intended to include the plural forms as well, unless the context clearly indicates otherwise. It will be further understood that the root terms “include” and/or “have”, when used in this specification, specify the presence of stated features, integers, steps, operations, elements, and/or components, but do not preclude the presence or addition of one or more other features, integers, steps, operations, elements, components, and/or groups thereof.

[0088] The corresponding structures, materials, acts, and equivalents of all means plus function elements in the claims below are intended to include any structure, or material, for performing the function in combination with other claimed elements as specifically claimed. The description of the present invention has been presented for purposes of illustration and description, but is not intended to be exhaustive or limited to the invention in the form disclosed. Many modifications and variations will be apparent to those of ordinary skill in the art without departing from the scope and spirit of the invention. The embodiments were chosen and described in order to best explain the principles of the invention and the practical application, and to enable others of ordinary skill in the art to understand the invention for various embodiments with various modifications as are suited to the particular use contemplated.

[0089] Although the present invention has been described in terms of particular example embodiments, it is not limited to those embodiments. The embodiments, examples, and modifications which would still be encompassed by the invention may be made by those skilled in the art, particularly in light of the foregoing teachings.

[0090] As used above "in fluid communication" is intended to indicate that two components are in fluid communication with each other and that fluid is able to pass from one component to another component directly or indirectly.

[0091] As used above “substantially,” “generally,” and other words of degree are relative modifiers intended to indicate permissible variation from the characteristic so modified. It is not intended to be limited to the absolute value or characteristic which it modifies but rather possessing

more of the physical or functional characteristic than its opposite, and preferably, approaching or approximating such a physical or functional characteristic.

[0092] Those skilled in the art will appreciate that various adaptations and modifications of the exemplary and alternative embodiments described above can be configured without departing from the scope and spirit of the invention. Therefore, it is to be understood that, within the scope of the appended claims, the invention may be practiced other than as specifically described herein.

[0093] While particular embodiments of the present invention have been shown and described, it will be obvious to those skilled in the art that changes and modifications may be made without departing from this invention. Therefore, it is intended that the claims herein are to include all such obvious changes and modifications as fall within the true spirit and scope of this invention.

VI. INDUSTRIAL APPLICABILITY

[0094] A system featuring a head-only or whole body inhalation exposure chamber and a method of opposed, normal, bi-directional aerosol delivery. The system includes a laminar flow element and a radial exhaust.

VII. REFERENCE

[0095] The teachings of the references cited herein are incorporated herein in their entirety: McClellan, R.O and Henderson, R.E. (edited). Concepts in Inhalation Toxicology. Hemisphere Publishing Corporation, p 45-48. 1989.

What is claimed:

1. A system comprising:

an inhalation exposure chamber;

an aerosol delivery line connected to said inhalation exposure chamber, said aerosol delivery line configured to produce a bi-directional and symmetrical presentation of aerosol to said inhalation exposure chamber;

a laminar flow element configured to create an ante-chamber where complete and turbulent mixing of the aerosol occurs; and

a radial exhaust, said radial exhaust and said laminar flow element enable laminar flow of the aerosol through said inhalation exposure chamber.

2. The system according to claim 1, wherein said aerosol delivery line is centrally located at a top of said inhalation exposure chamber, wherein said laminar flow element is located below said aerosol delivery line, and

wherein said radial exhaust is centrally located at a bottom of said inhalation exposure chamber.

3. The system according to claim 1, further comprising at least one sample collection port located at a midpoint of a wall of said inhalation exposure chamber.

4. The system according to claim 1, wherein said laminar flow element includes a sheet having a plurality of perforations.

5. The system according to claim 1, further including at least one additional laminar flow element, said at least one additional laminar flow element includes a sheet having a plurality of perforations.

6. The system according to any one of claims 1-5, wherein said radial exhaust includes eight tubes positioned radially around a central hub, wherein each tube includes a sealed distal end and a plurality of lateral holes.

7. The system according to claim 6, wherein said radial exhaust includes four first tubes having a first length and four second tubes having a second length, wherein said second length is greater than said first length.

8. The system according to any one of claims 1-5, wherein said aerosol delivery line is a 12.0 inch long pipe having a 1.5 inch diameter.

9. The system according to any one of claims 1-5, wherein said aerosol delivery line includes two 0.25 inch wide by 8.0 inch long slots positioned 180° apart.

10. The system according to any one of claims 1-5, wherein said laminar flow element includes an 8.0 inch x 8.0 inch x 0.735 inch honeycomb.

11. The system according to any one of claims 1-5, wherein said laminar flow element includes an 8.0 inch x 8.0 inch x 0.0156 inch sheet with 0.25 inch perforations.

12. The system according to any one of claims 1-5, wherein said laminar flow element includes an 8.0 inch x 8.0 inch x 0.0625 inch sheet with 0.0625 inch diameter perforations.

13. The system according to any one of claims 1-5, wherein said radial exhaust includes 0.375 inch diameter tubes positioned radially around a 1.5 inch diameter x 2.0 inch high central hub.

14. The system according to claim 13, wherein each of said tubes includes a sealed distal end and five 0.125 inch lateral apertures.

15. The system according to any one of claims 1-5, further comprising at least two cages in said inhalation exposure chamber.

16. The system according to any one of claims 1-5, further comprising four cages in said inhalation exposure chamber.

17. The system according to any one of claims 1-5, wherein said inhalation exposure chamber includes four sidewalls, wherein one of said sidewalls includes an aperture dimensioned to receive a primate head therethrough.

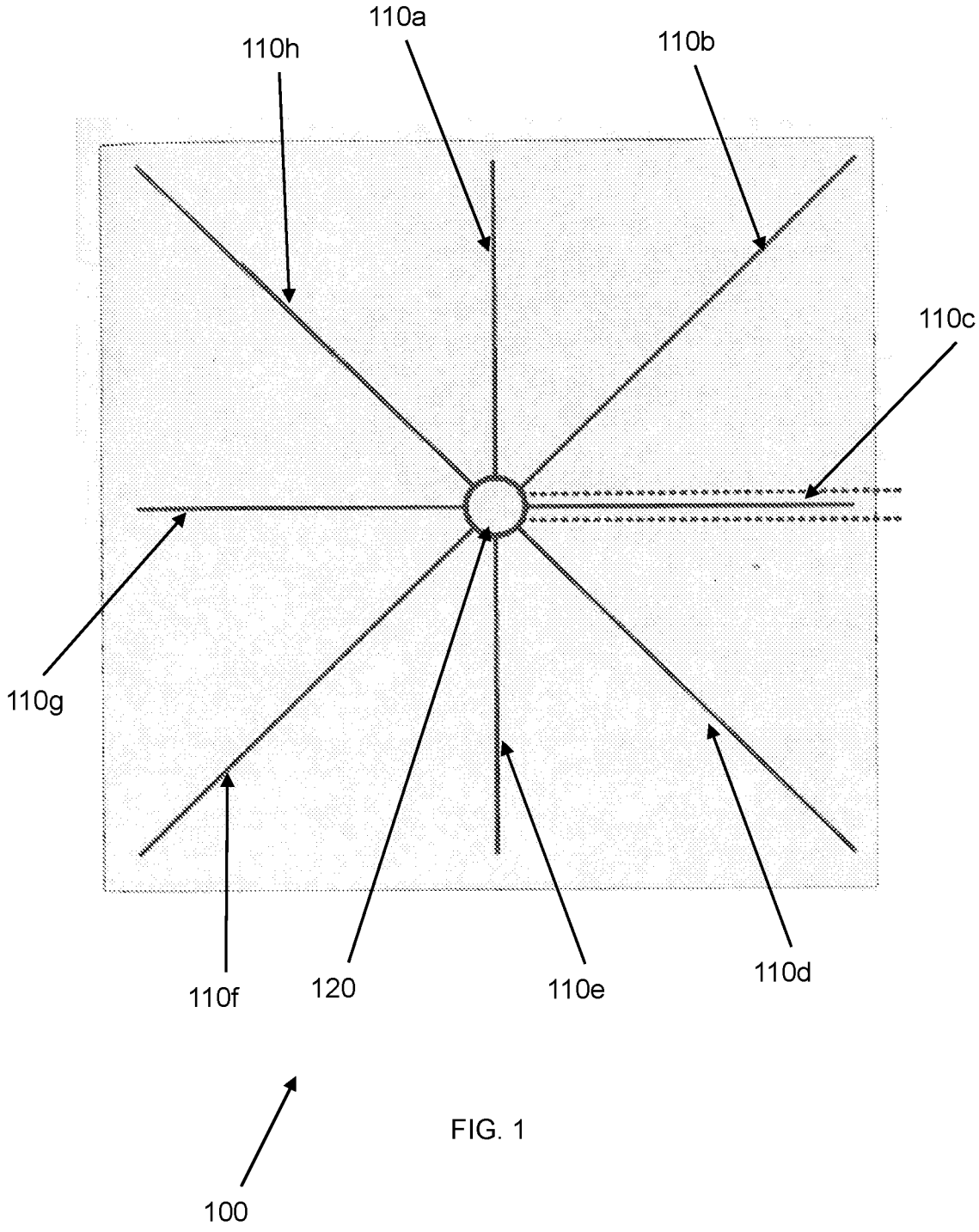


FIG. 1



FIG. 2

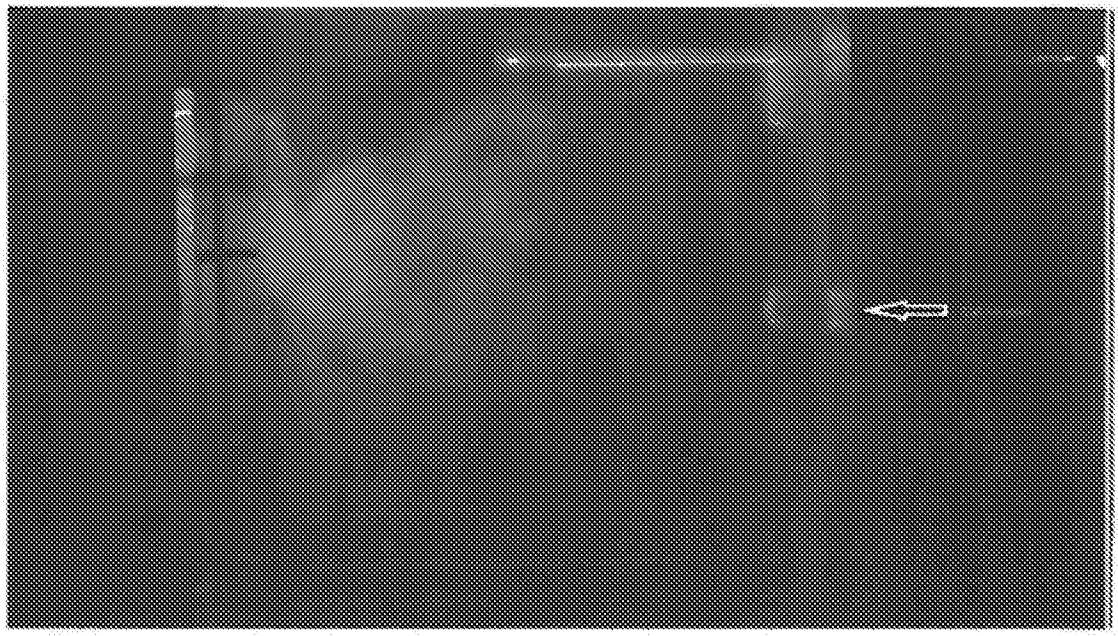


FIG. 3

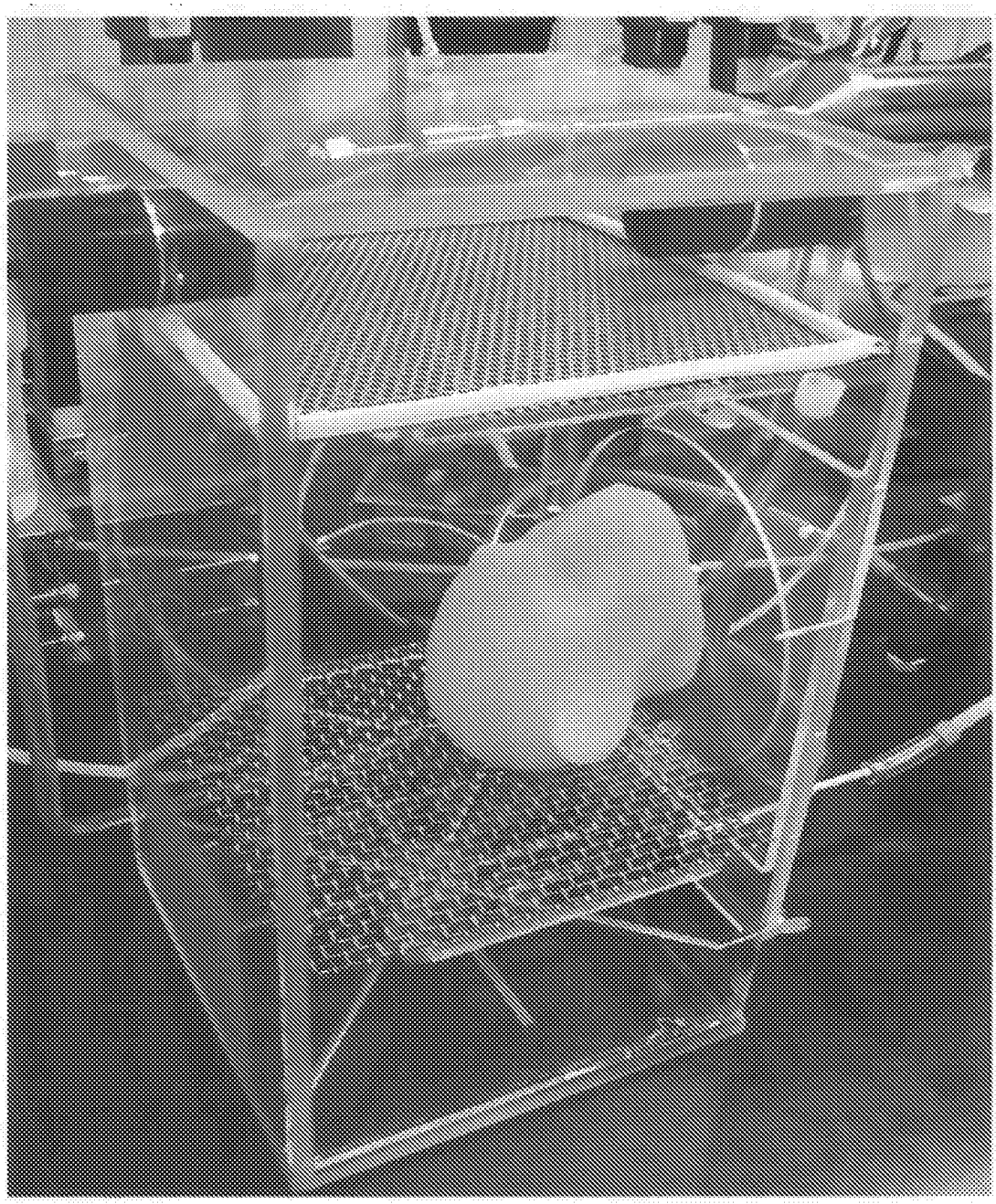


FIG. 4

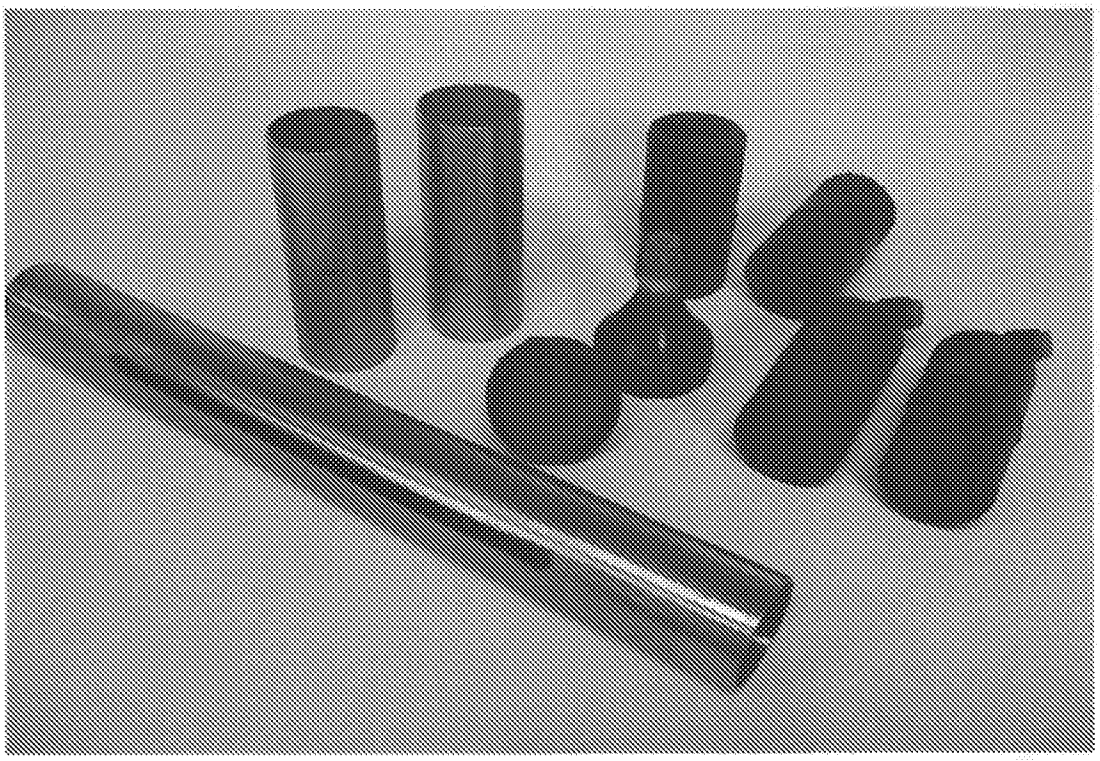


FIG. 5

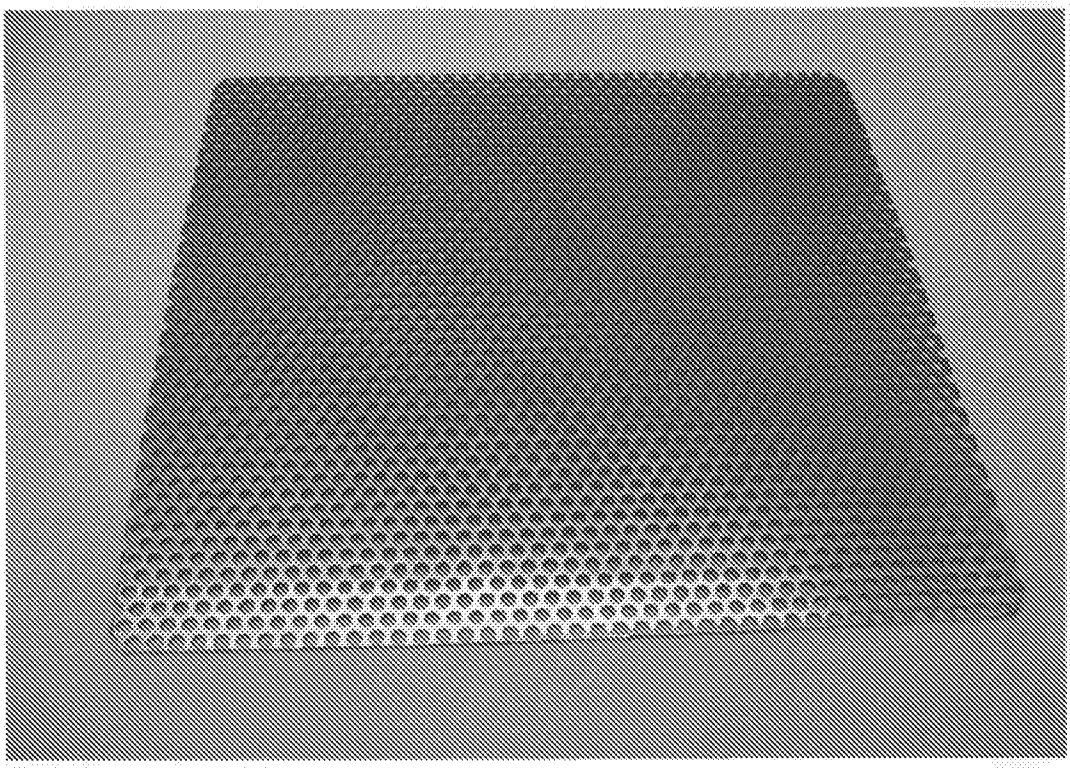


FIG. 6

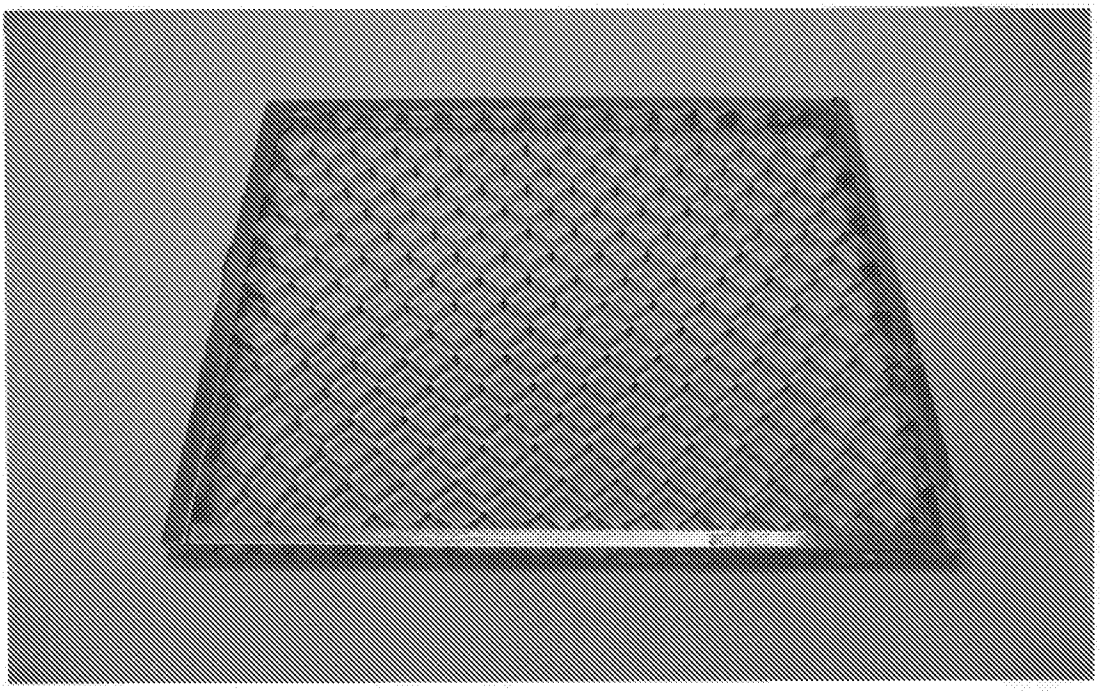


FIG. 7

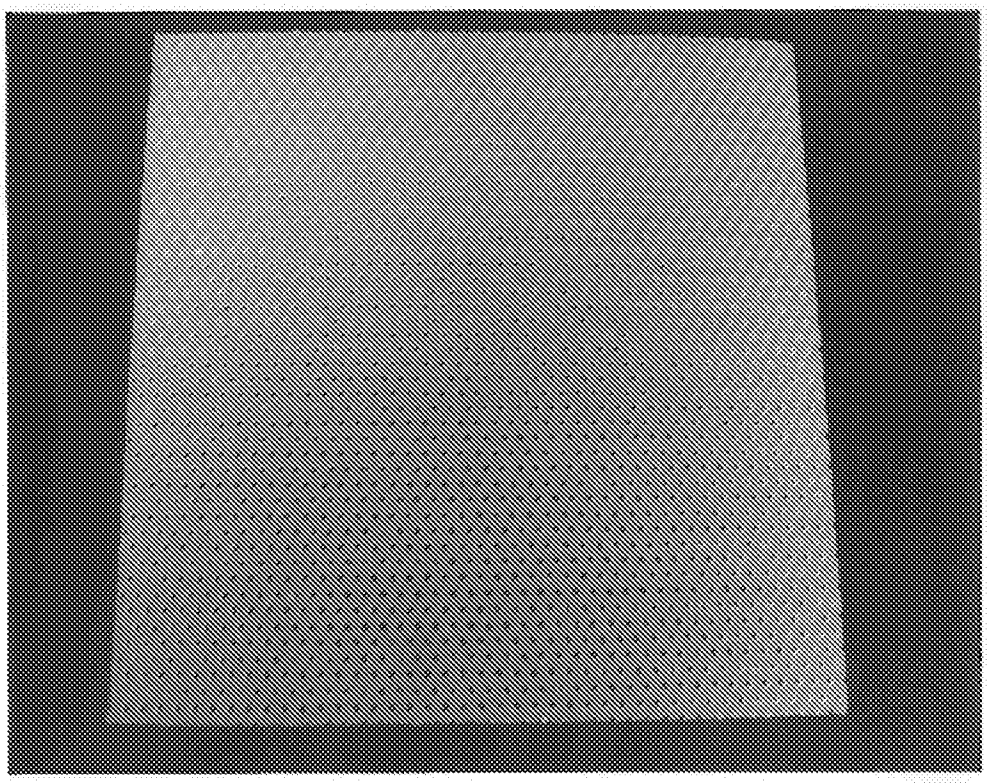


FIG. 8

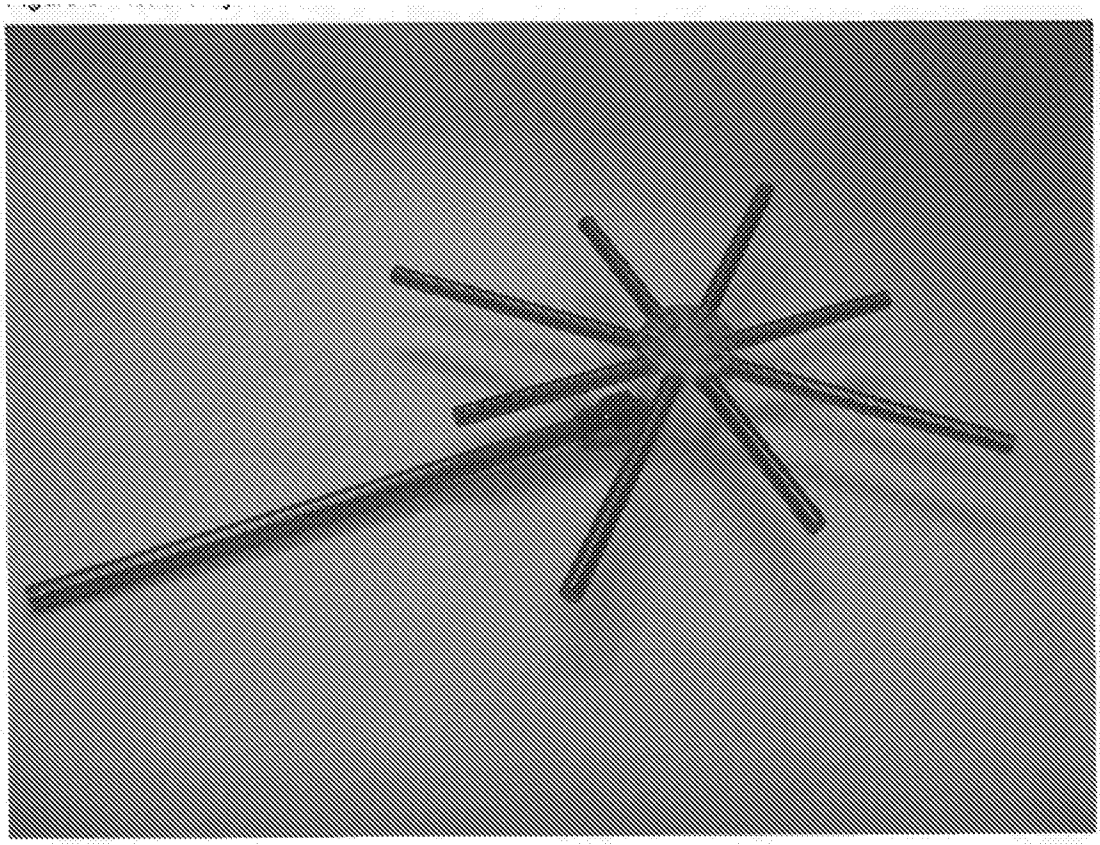


FIG. 9

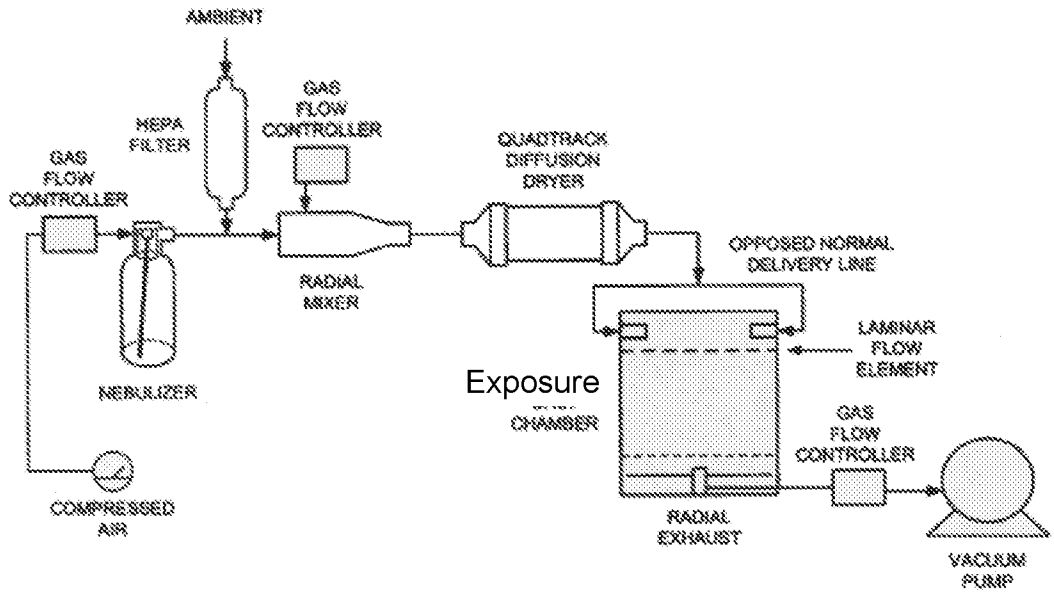

AMBIENT
HEPA FILTER
GAS FLOW CONTROLLER
GAS FLOW CONTROLLER
QUADTRACK DIFFUSION DRYER
OPPOSED NORMAL DELIVERY LINE
RADIAL MIXER
NEBULIZER
Exposure CHAMBER
LAMINAR FLOW ELEMENT
GAS FLOW CONTROLLER
COMPRESSED AIR
RADIAL EXHAUST
VACUUM PUMP

FIG. 10

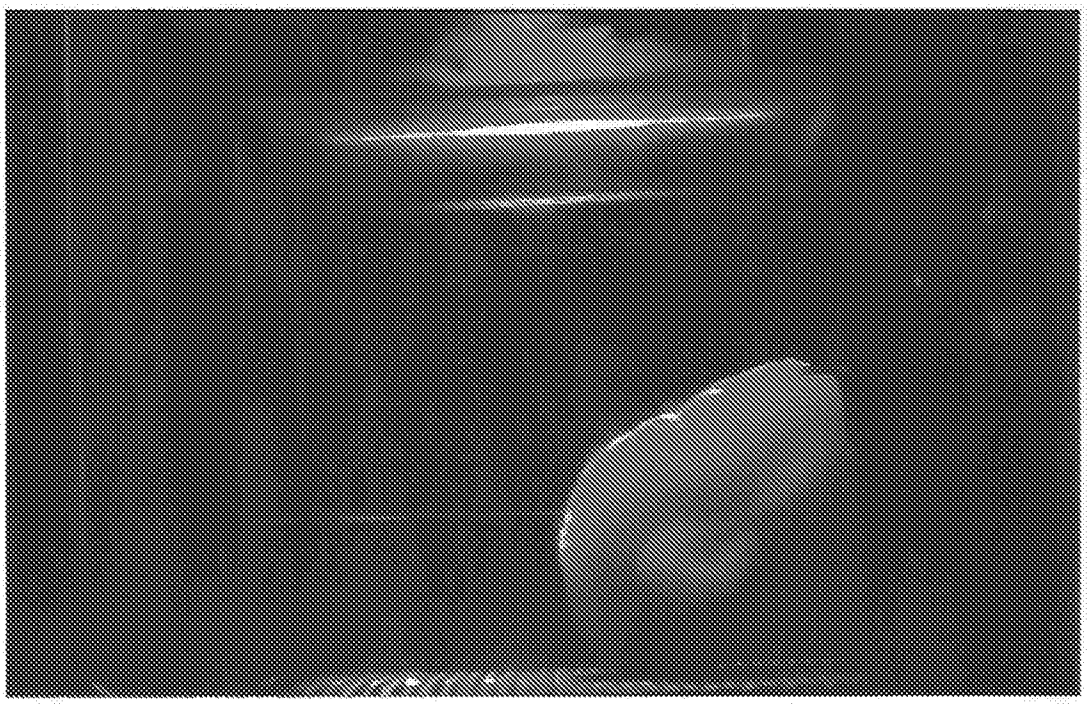


FIG. 11

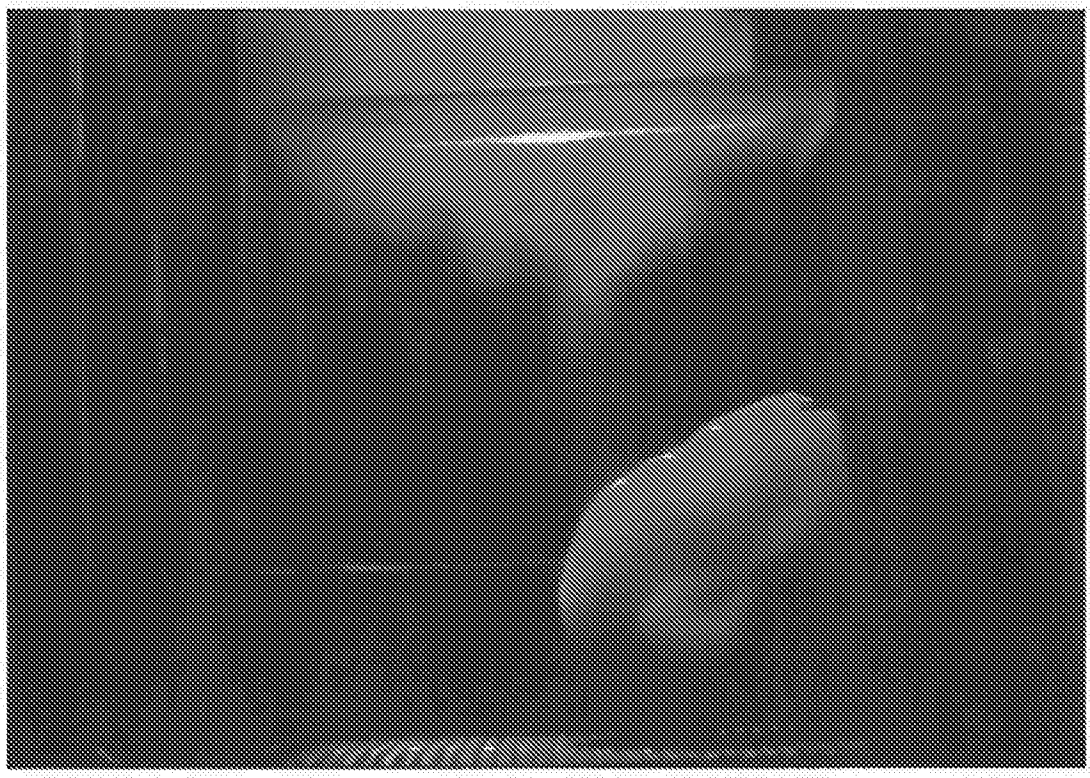


FIG. 12

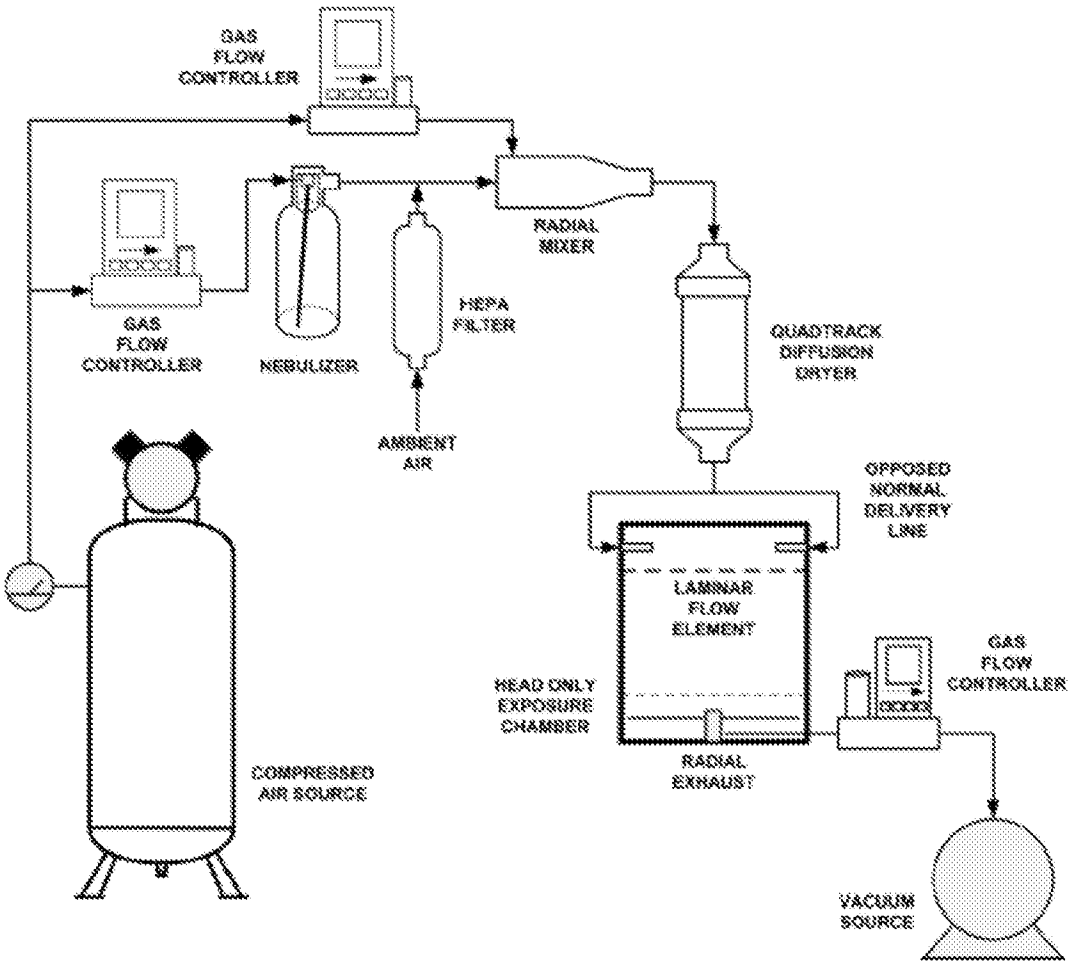

GAS FLOW CONTROLLER
GAS FLOW CONTROLLER
NEBULIZER
HEPA FILTER
AMBIENT AIR
RADIAL MIXER
QUADTRACK DIFFUSION DRYER
OPPOSED NORMAL DELIVERY LINE
LAMINAR FLOW ELEMENT
HEAD ONLY EXPOSURE CHAMBER
RADIAL EXHAUST
GAS FLOW CONTROLLER
COMPRESSED AIR SOURCE
VACUUM SOURCE

FIG. 13

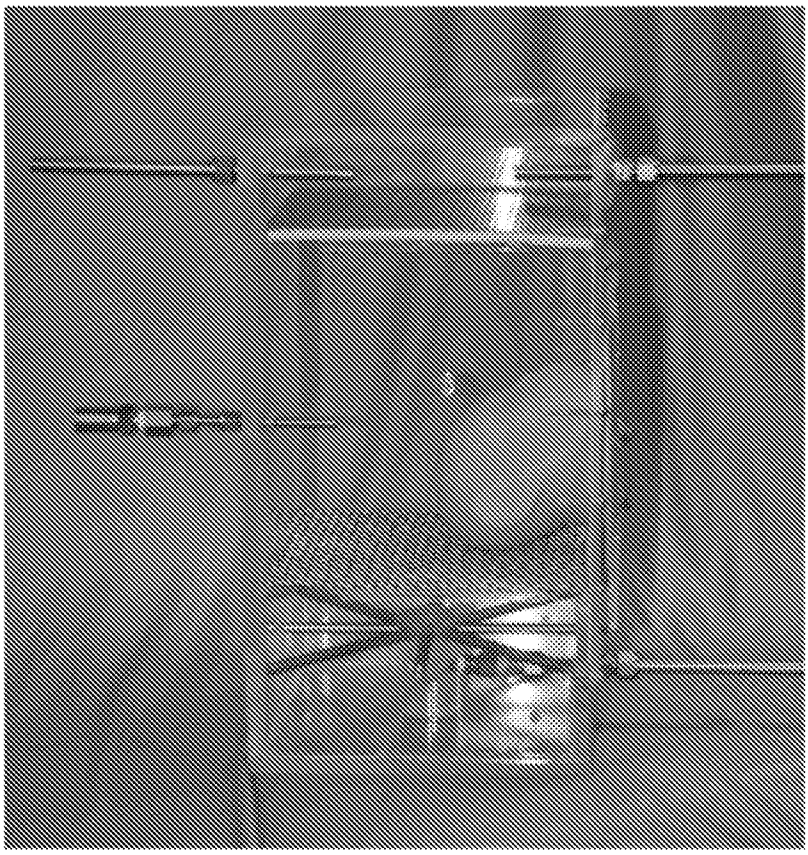


FIG. 14

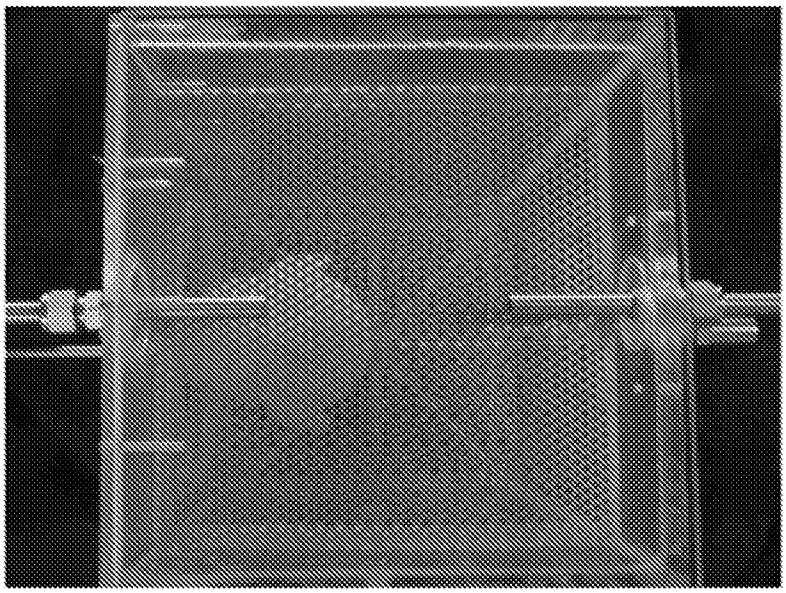


FIG. 15

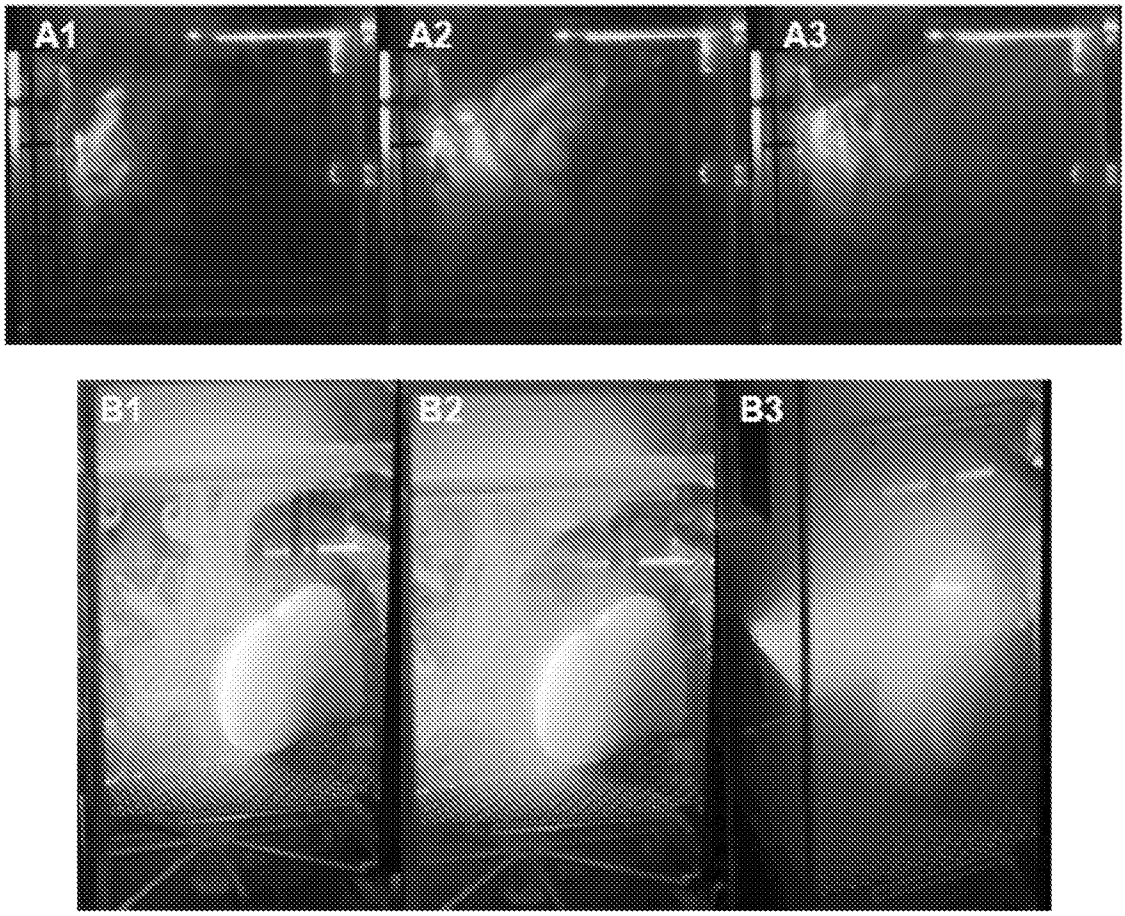


FIG. 16

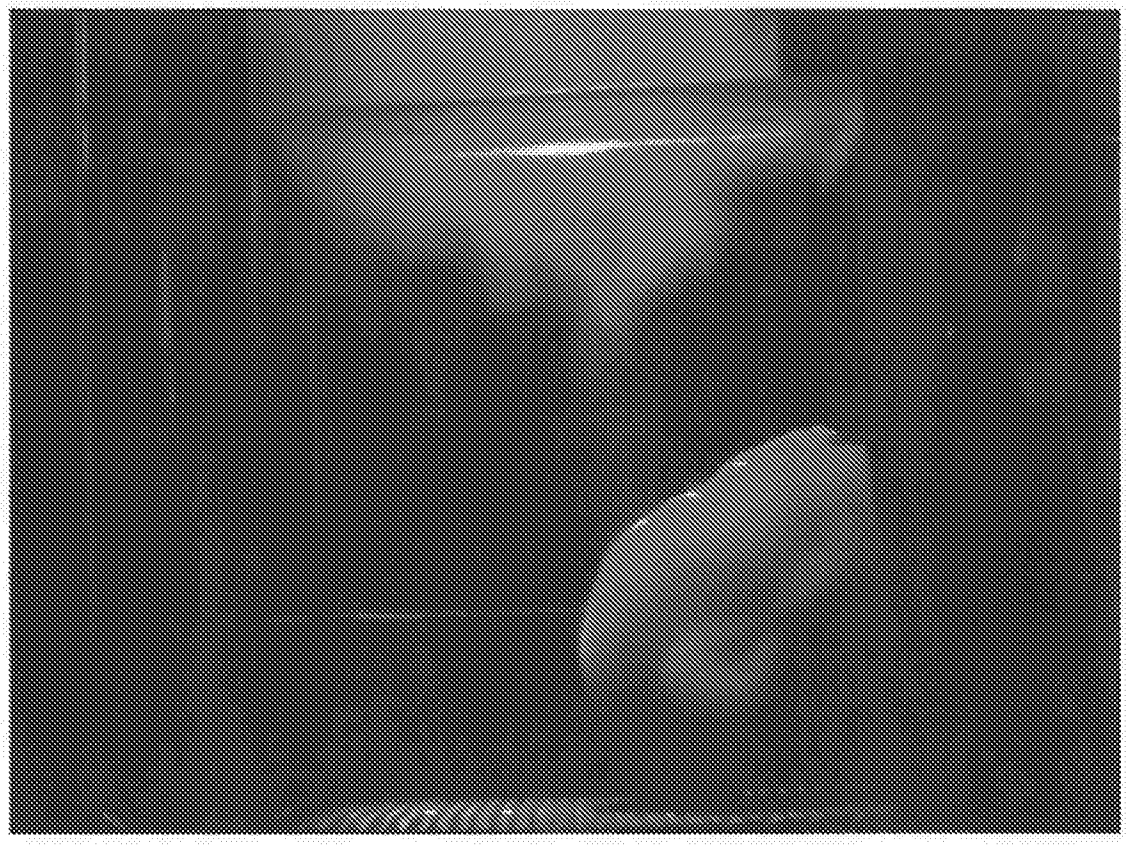


FIG. 17

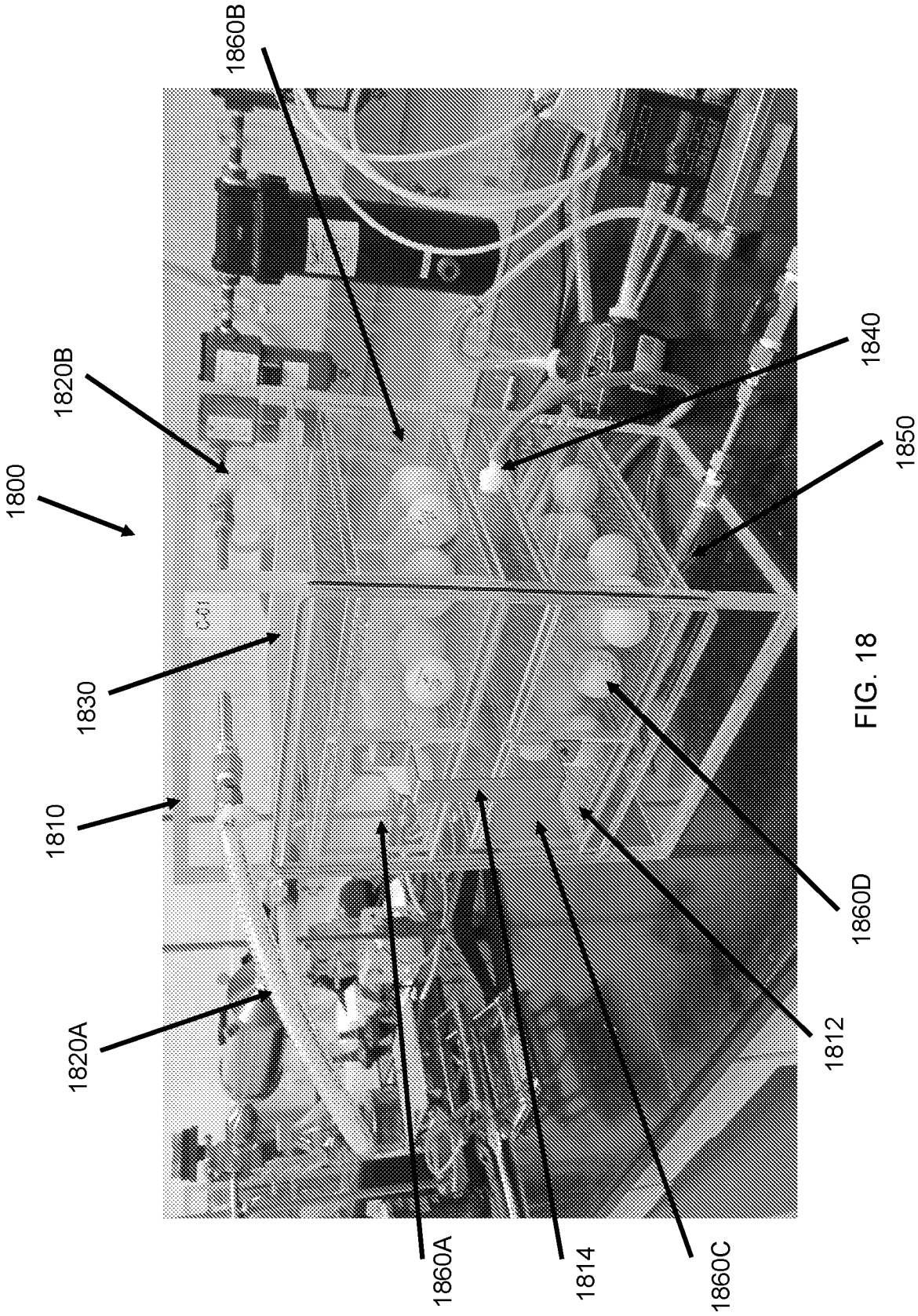

1860B
1820B
1800
1840
1850
1830
1810
1820A
1860D
1812
1860A
1814
1860C

FIG. 18

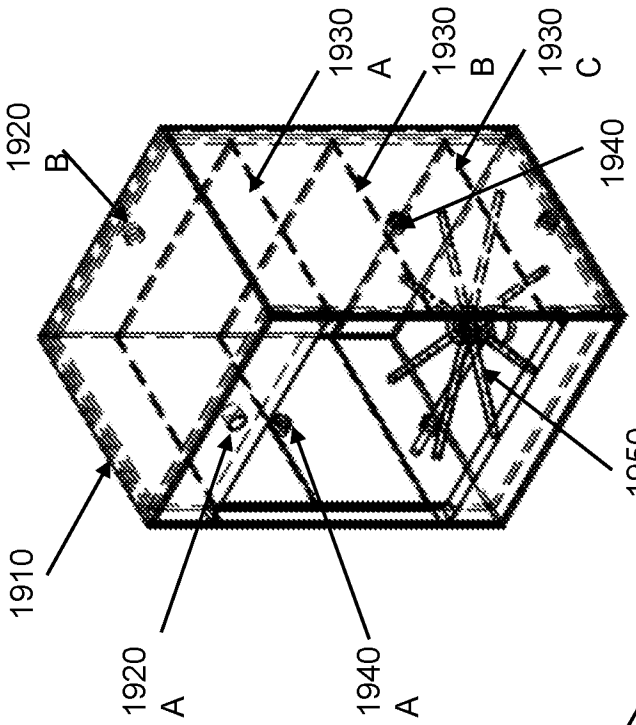


FIG. 19A

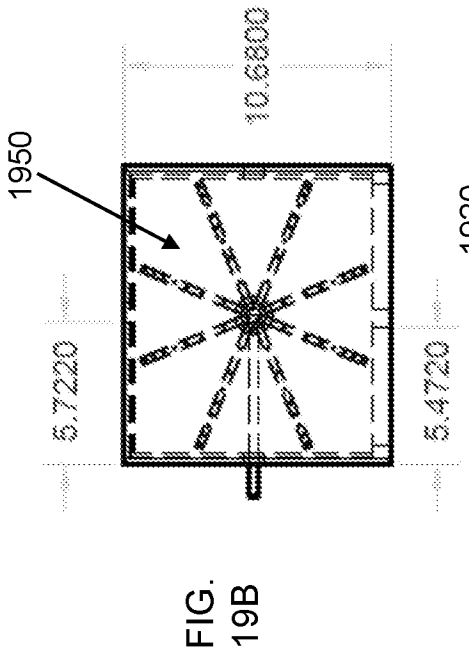


FIG. 19B

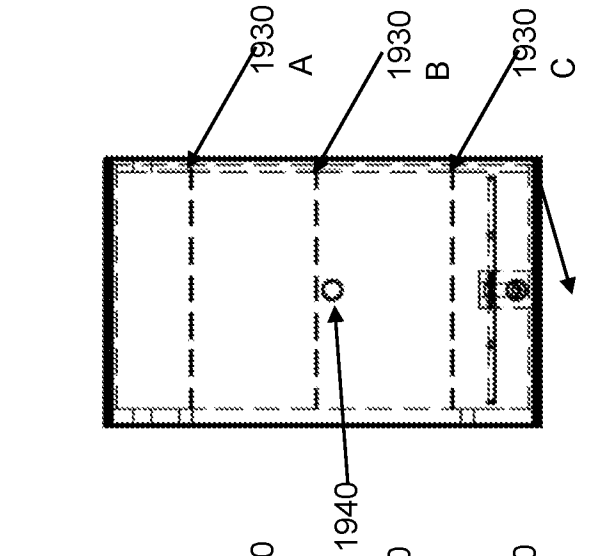


FIG. 19D

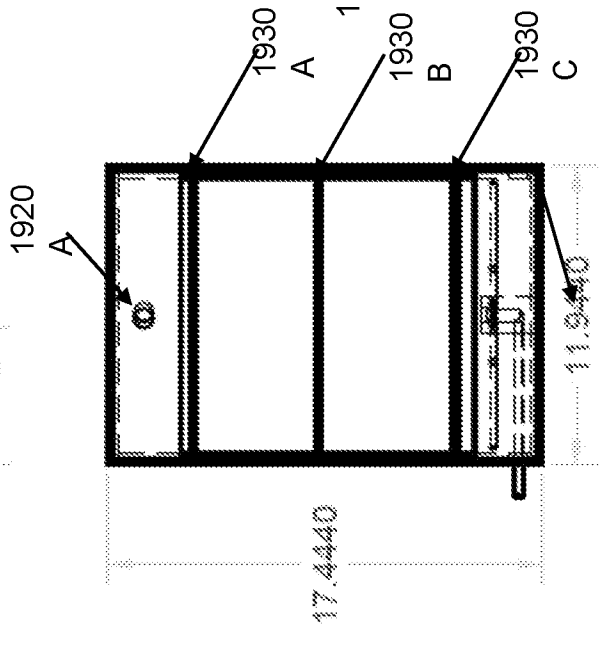


FIG. 19C

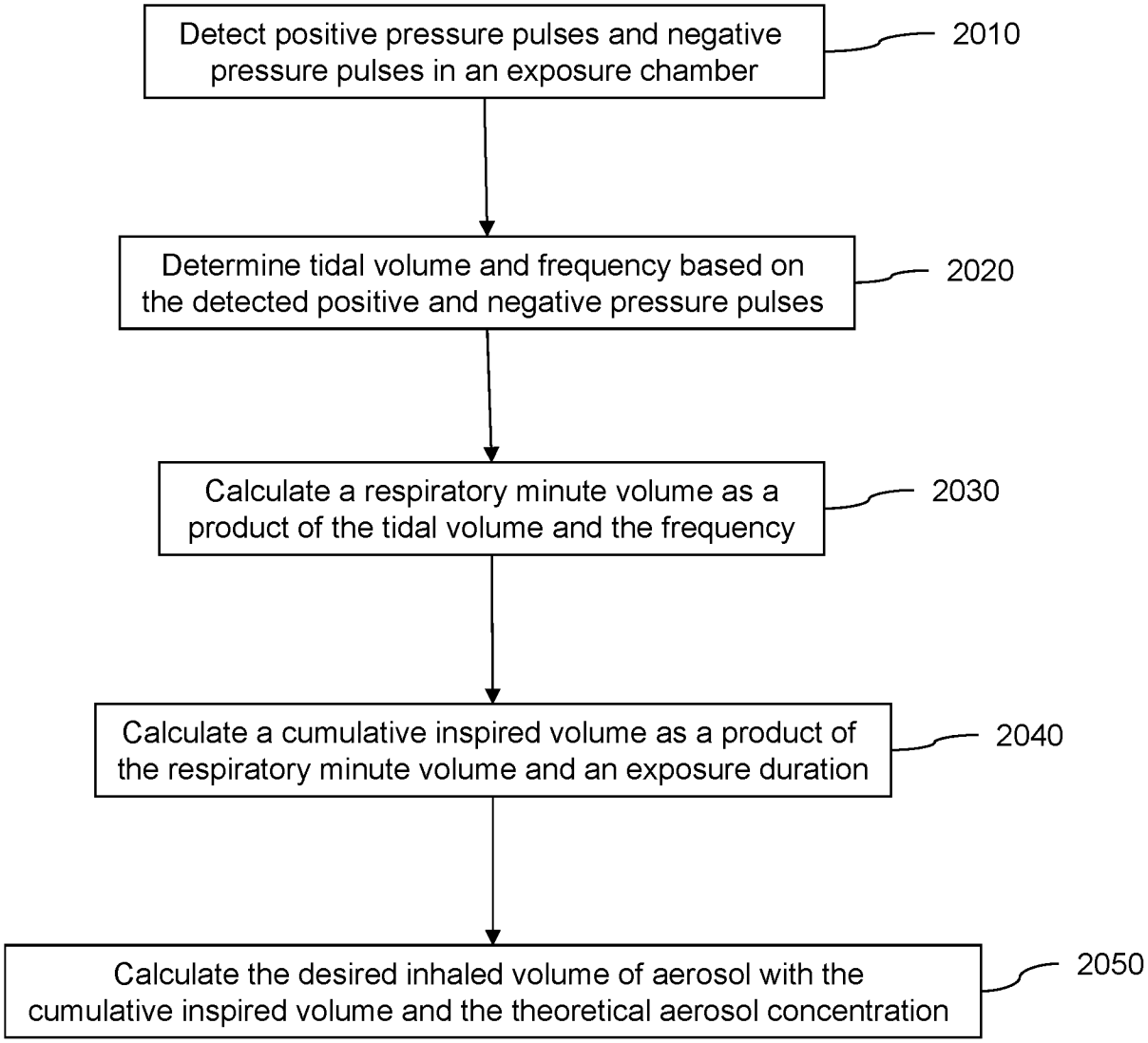

Detect positive pressure pulses and negative pressure pulses in an exposure chamber
2010
Determine tidal volume and frequency based on the detected positive and negative pressure pulses
2020
Calculate a respiratory minute volume as a product of the tidal volume and the frequency
2030
Calculate a cumulative inspired volume as a product of the respiratory minute volume and an exposure duration
2040
Calculate the desired inhaled volume of aerosol with the cumulative inspired volume and the theoretical aerosol concentration
2050

FIG. 20

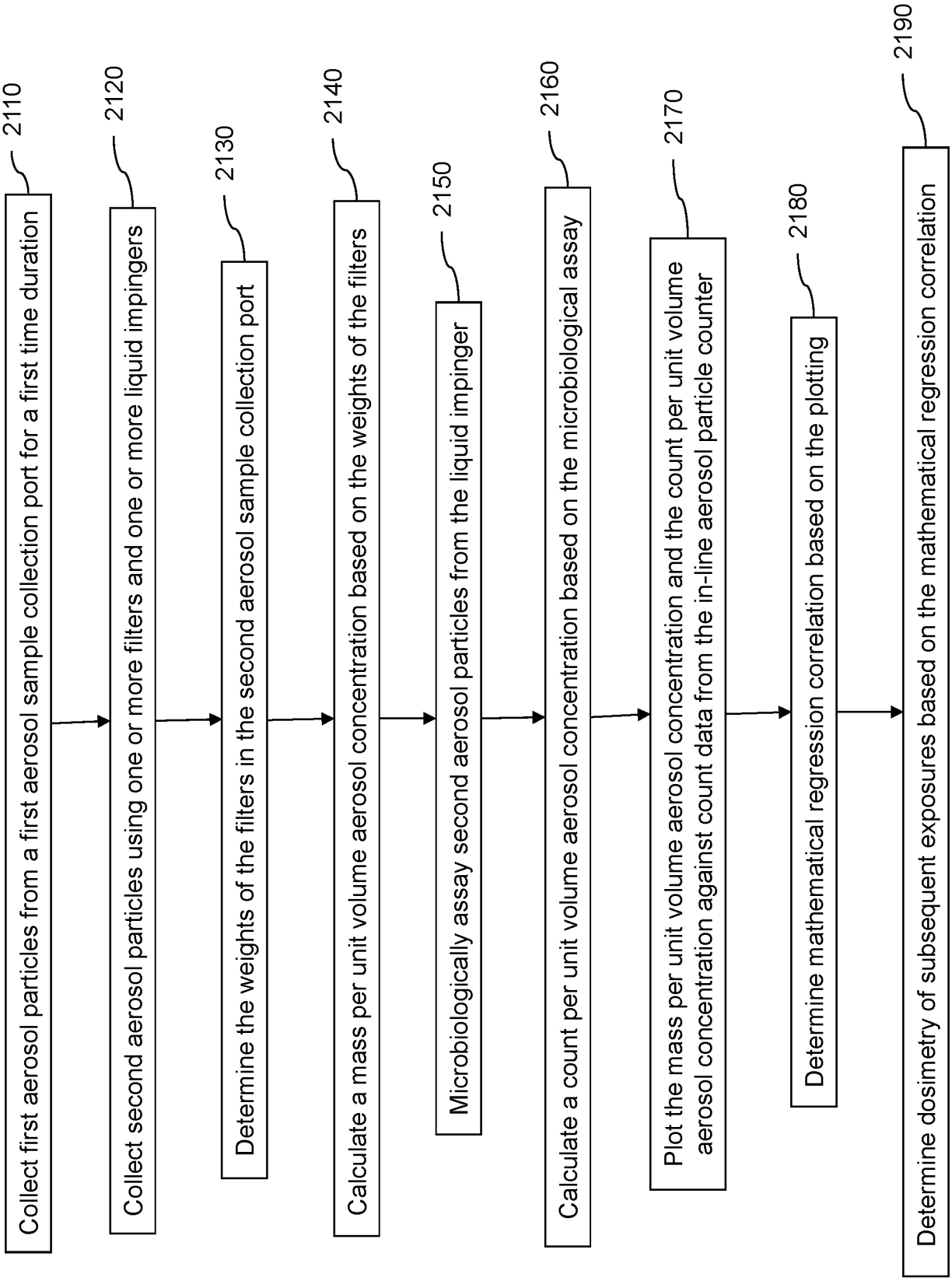

2110
Collect first aerosol particles from a first aerosol sample collection port for a first time duration
2120
Collect second aerosol particles using one or more filters and one or more liquid impingers
2130
Determine the weights of the filters in the second aerosol sample collection port
2140
Calculate a mass per unit volume aerosol concentration based on the weights of the filters
2150
Microbiologically assay second aerosol particles from the liquid impinger
2160
Calculate a count per unit volume aerosol concentration based on the microbiological assay
2170
Plot the mass per unit volume aerosol concentration and the count per unit volume aerosol concentration against count data from the in-line aerosol particle counter
2180
Determine mathematical regression correlation based on the plotting
2190
Determine dosimetry of subsequent exposures based on the mathematical regression correlation

FIG. 21

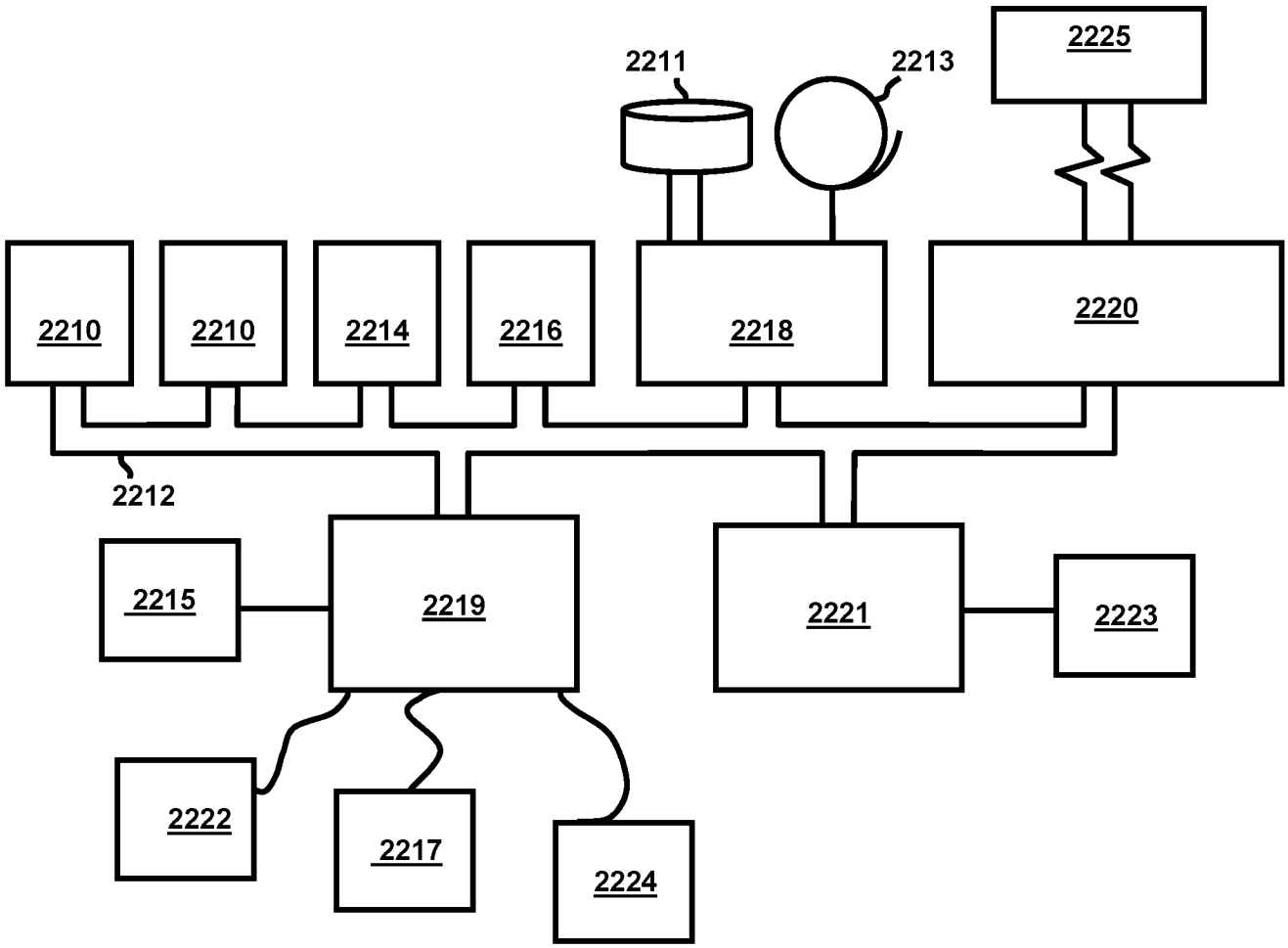

2225
2211
2213
2210
2210
2214
2216
2218
2220
2212
2215
2219
2221
2223
2222
2217
2224

FIG. 22

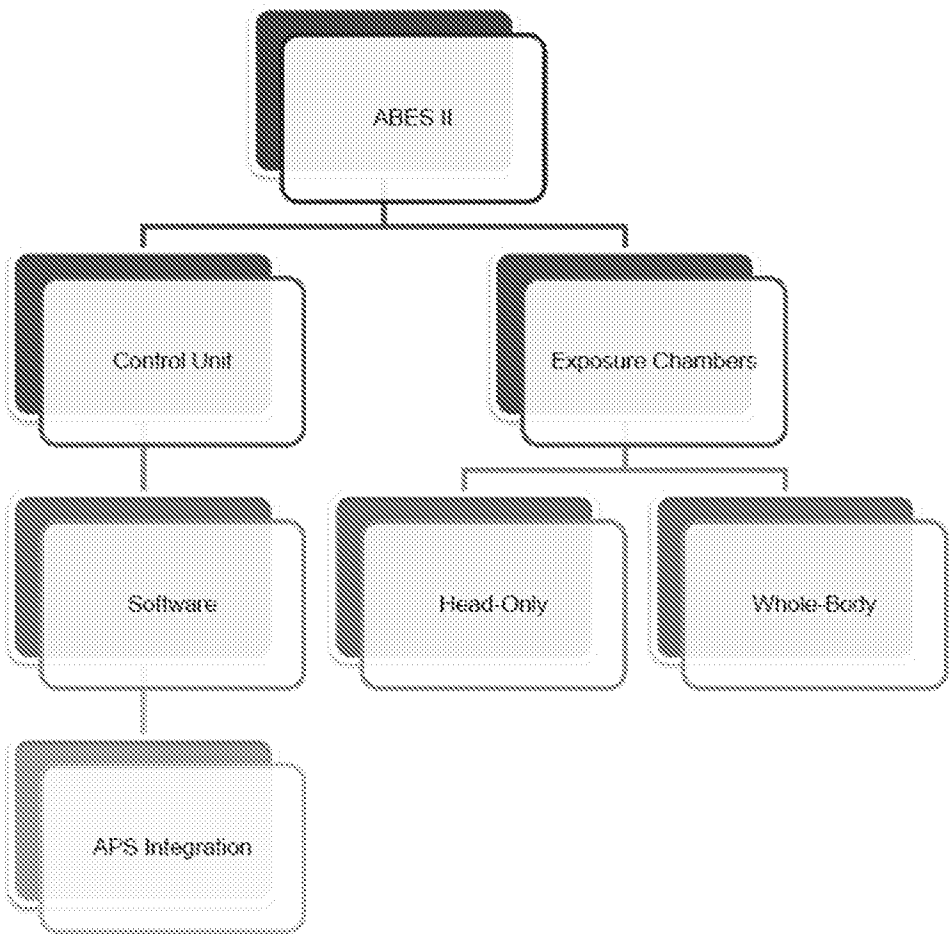

ABES II
Control Unit
Exposure Chambers
Software
Head-Only
Whole-Body
APS Integration

FIG. 23

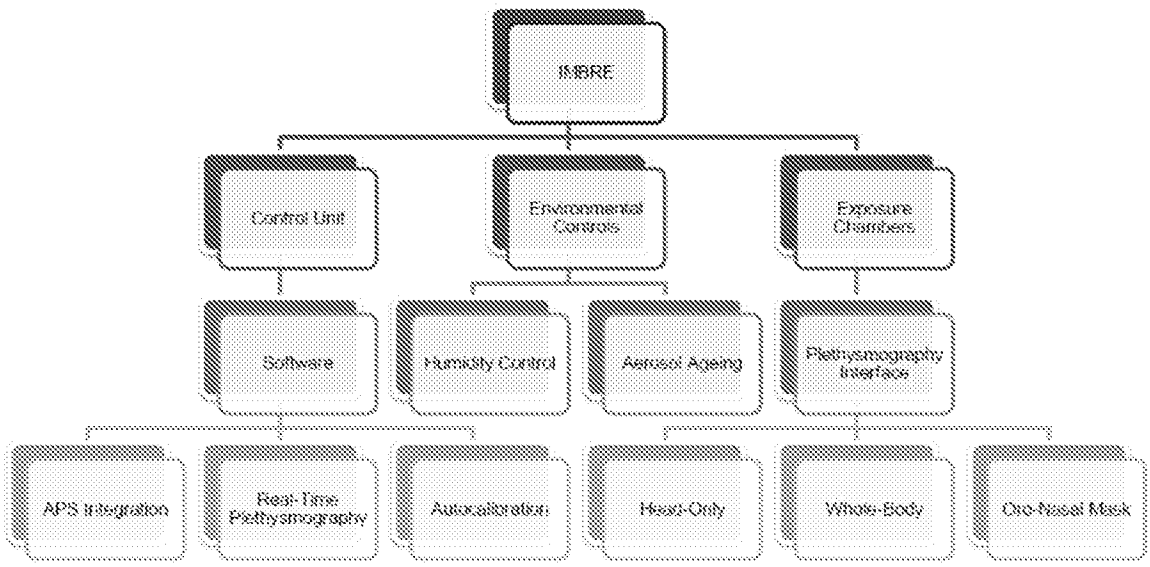

IMBRE
Control Unit
Environmental Controls
Exposure Chambers
Software
Humidity Control
Aerosol Ageing
Plethysmography Interface
APS Integration
Real-Time Plethysmography
Autocalibration
Head-Only
Whole-Body
Oro-Nasal Mask

FIG. 24

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US2017/016811

A. CLASSIFICATION OF SUBJECT MATTER

IPC(8) - A61M 13/00; A01K 1/00; A61D 7/00; A61D 7/04; A61M 11/00; A61M 11/06 (2017.01)

CPC - A61M 15/0086; A61D 7/00; A61D 7/04; A61M 11/00; A61M 15/00; A61M 15/0048; A61M 2202/064; A61M 2206/16 (2017.02)

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)

See Search History document

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

USPC - 119/420; 128/200.14; 128/200.18; 128/200.23; 128/200.24; 128/203.12; 128/203.26 (keyword delimited)

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

See Search History document

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US 5,297,502 A (JAEGER) 29 March 1994 (29.03.1994) entire document	1-17
Y	US 8,985,100 B2 (MINOCCHIERI et al) 24 March 2015 (24.03.2015) entire document	1-17
Y	US 5,379,777 A (LOMASK) 10 January 1995 (10.01.1995) entire document	4, 5, 11, 12
Y	US 5,487,378 A (ROBERTSON et al) 30 January 1996 (30.01.1996) entire document	10

☐ 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

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"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

"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

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
23 March 2017	14 APR 2017
Name and mailing address of the ISA/US Mail Stop PCT, Attn: ISA/US, Commissioner for Patents P.O. Box 1450, Alexandria, VA 22313-1450 Facsimile No. 571-273-8300	Authorized officer Blaine R. Copenheaver PCT Helpdesk: 571-272-4300 PCT OSP: 571-272-7774