A MECHANICAL ASPIRATOR FOR SAFE TRANSFER OF ARBOVIRUS-INFECTED MOSQUITOES WITHIN CONTAINMENT CHAMBERS[1]

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In the laboratory, aspirating devices are frequently used to transfer small flying insects like mosquitoes from container to container. Gentle aspiration is often the preferred means of manipulating such insects, as the need for anesthetics is obviated. Aspirators in common use are either lung-powered, or are small battery-powered units providing suction only (Service 1976). Aspiration of mosquitoes infected with vertebrate pathogens poses special challenges, due to the additional demands for biocontainment. First, lung-powered aspirators expose the operator to potentially infectious aerosols or particulate debris, and many battery-powered aspirators require the operator to detach holding tubes in order to expel aspirated mosquitoes by exhalation. Secondly, use of any type of aspirator for transferring infected mosquitoes within an open room exposes the operator to risk because of the possibility of occasional escapes, even if the transfers are conducted in a room that is part of a biocontainment facility. Safety can be virtually assured by making such transfers within sealed containment chambers, such as glove boxes. Unfortunately, common battery-powered aspirators are very difficult to use within such confines, particularly when it is necessary to expel aspirated insects individually to separate holding containers. Use of such aspirators is further hindered if the operator is wearing heavy rubber gloves to preclude exposure to infectious bites.

Spielman (1964) described two aspirators that can be used to transfer mosquitoes within glove boxes, since attached air pressure and suction lines can be made to exit the boxes via sleeved exit holes that are also used for the operator's arms. However, one of Spielman's aspirators requires that the operator use a mouth tube for expulsion of aspirated mosquitoes, while his other design can be difficult or tiring when used for long periods. The mechanical aspirator described here avoids the limitations of previous devices and can be used for extended periods without fatigue.

The new aspirator consists of four major components, a rotary vane pressure-suction pump rated at 37 liters/min free air displacement (Gast Model No. SA55JZGTD-4144) (Fig. 1A), a valve assembly (Fig. 1B) and foot pedal assembly (Fig. 1C) for switching the aspirator from suction to aspiration modes, and a wooden box (Fig. 1D). The valve-pedal assembly is secured to this box, and the pump is placed within it to provide sufficient weight to hold the device stationary on the floor during use. The pump includes pressure and suction gauges, and bleeder valves that permit regulation of pressure and aspiration to yield airflow that is appropriate for transfers, but not strong enough to injure mosquitoes. While it may be possible to use pumps other than those of the rotary vane type, I have found that piston pumps of similar capacity tend to produce a pulsating suction that is more likely to damage aspirated mosquitoes.

The valve-pedal assembly includes two metal three-way stopcocks (B-D No. 3195) (Fig. 2A), soldered to 8 × 32, 1.9 cm flat-head mounting bolts on the flat side opposite the side port. The stopcocks are then bolted to two 2.5 cm horizontal slots in the mounting plate to facilitate accurate linkage of their operating levers; minimum separation of the slots is 3.8 cm. The mounting plates are constructed of 0.48 cm aluminum (Fig. 1E) and are connected by bolts to aluminum angles (Fig. 1F). Operating levers of both stopcocks are mechanically linked by a 5.1 cm section of lucite rod (1.9 cm diam), countersunk and slotted to receive them (Fig. 2B). When properly connected, the stopcocks can be operated smoothly by 90° rotation of the lucite rod, changing the routing of airflow almost instantaneously in each stopcock. A tubing clamp (Fig. 2C) attached to the center of the lucite rod serves for attachment of the operating cable (Fig. 2D) and the return spring (Fig. 2E). A bolt attached to the mounting plate serves as a valve stop (Fig. 2F), the height of which is adjusted to assure accurate alignment of air channels in the stopcocks when in the "down" position. The foot pedal (Fig. 1C) is made of aluminum (0.64 × 5.1 × 20.3 cm) attached to a common door hinge at the pivot point (Fig. 1G). Excessively long bolts (3.2 cm) are used to attach the hinge to the pedal and the mounting plate; bolts attached to the

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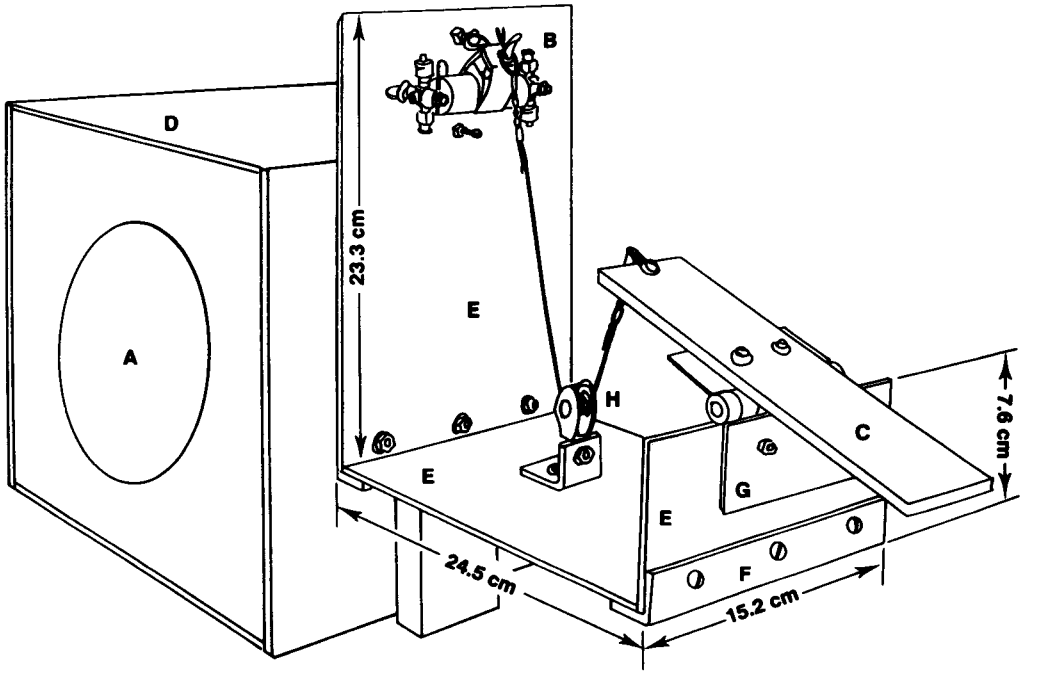

Fig. 1. Mechanical aspirator, tubing connections and aspirator handle not shown; A. Gast No. SA55JZGTD-4144 rotary vane pressure-suction pump, B. valve assembly, C. foot pedal, D. wooden box, E. aluminum mounting plates, F. aluminum angles, G. door hinge, H. pulley.

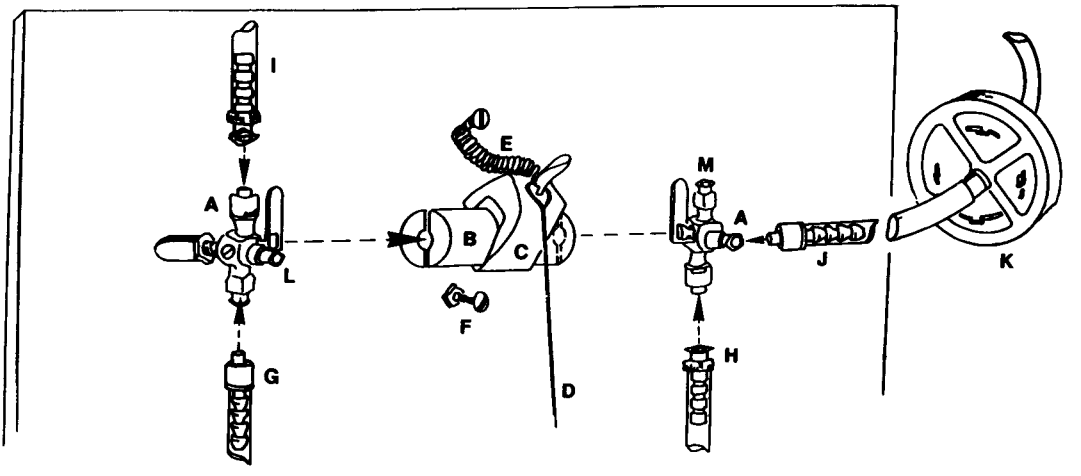

Fig. 2. Valve assembly; A. B-D No. 3195 three-way stopcocks, B. lucite rod connector, C. tubing clamp, D. operating cable, E. return spring, F. valve stop, G. pressure line from pump, H. suction line from pump, I. aspirator line attached to stopcock top port, J. aspirator line attached to stopcock side port, K. Gelman No. 4402 "Vacushield" filter, L. stopcock side port, M. stopcock top port.

pedal contact those attached to the mounting plate, serving as a stop for the pedal when foot pressure is not being applied. The operating cable consists of 13.6 kg-test (30 lb) nylon-coated stainless steel fishing leader, routed through a 1.9 cm diam pulley (Fig. 1H) mounted on the bottom plate. While the mounting plates could be made of material other than aluminum, they should be sufficiently rigid to withstand the pressure of foot operation and the pulling action of the operating cable.

Pressure (Fig. 2G) and suction (Fig. 2H) lines from the pump are connected to the valve assembly as shown, using, respectively, male (B-D No. 9067) and female (B-D No. 9040) Luer-Lok connectors. It is critical that tubing be connected to

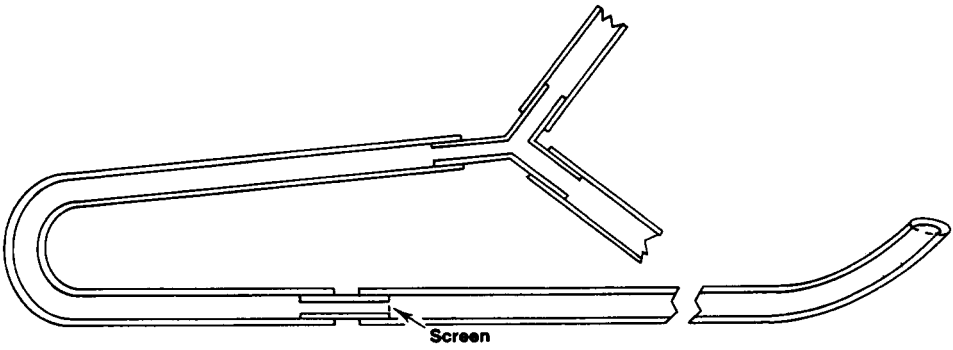

Fig. 3. Aspirator handle assembly.

valve outlets exactly as shown in Fig. 2 to achieve proper switching action. The aspirator handle (Fig. 3) consists of 0.95 cm (inner diam) × 0.16 cm wall plexiglass tubing, 30 cm long and with a slight curve in the terminal 10 cm. One layer of nylon organdy is placed over the end of a 3.8 cm section of 0.80 cm (outer diam) tubing, which is then snugly inserted (half-way) into the straight end of the larger tubing. The organdy serves as a screen to retain aspirated mosquitoes. A section of plastic tubing approximately 200 cm long is attached at one end to the completed aspirator handle, and at the other end to a plastic Y-connector. A 45-cm section of tubing is attached to one branch of the Y-connector and attached by a female Luer-Lok adapter to the top port of the left stopcock as illustrated (Fig. 2I). This branch supplies air from the pump pressure line to expel mosquitoes. A 45 cm section of tubing is connected to the remaining branch and attached by a male Luer-Lok adapter to the side port of the right stopcock (Fig. 2J) to provide suction. A Gelman No. 4402 "Vacushield" filter may be inserted in the suction branch (Fig. 2K) to contain aspirated infectious aerosols.

In use, the glove box in which transfers are to be made is placed on a bench where the operator can sit with arms comfortably placed within the glove box. The aspirator assembly is then set on the floor where the operator can comfortably use the foot pedal. The aspirator handle is inserted into one of the arm holes of the glove box before turning on the pump. The aspirator is normally in "aspiration" mode. With no foot pressure on the operating pedal, the return spring maintains the stopcocks in the "up" position. This routes air from the aspirator handle to the righthand stopcock, and then through the suction line to the pump (Fig. 2J and Fig. 2H). Air from the pump pressure line enters the lefthand stopcock (Fig. 2G) and is exhausted through its side port (Fig. 2L).

The operator depresses the foot pedal to switch the aspirator instantaneously to "expulsion" mode, drawing the operating levers of both stopcocks "down" 90° and so changing the routing of the air channels. Now, air from pressure line G (Fig. 2) enters the lefthand stopcock and exits to the aspirator handle through the top port (Fig. 2I), expelling any mosquitoes contained downstream in the handle. The pump suction line (Fig. 2H) now draws air through the top port of the righthand stopcock (Fig. 2M). The foot pedal is released to return to aspiration mode.

For maximum comfort the foot pedal should be positioned above the floor so that the operator can maintain his foot poised over the pedal without having to maintain an unnatural posture. Correct height can be attained by altering the vertical position of the valve-foot pedal assembly at its point of attachment to the box housing the pump. The prototype aspirator uses a sliding rail to adjust pedal height.

The aspirator has been trouble-free after extensive use in transfer of mosquitoes. It requires no regular maintenance, aside from examination of the organdy screen in the aspirator handle on each day of use. The screen tends to accumulate lint, and it should be cleaned or replaced regularly. After extensive use the stopcocks will require cleaning and relubrication to assure smooth operation.

Many of the materials used to construct the aspirator are generally available in local hardware stores. The pump, stopcocks, Luer-Lok hose fittings and filter can be obtained through scientific supply houses. The author (an inexpert machinist) was able to construct the aspirator, using only a band saw, soldering gun, drill press and lathe; among which the latter is helpful, but not essential.

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