SPUN GLASS AIR FILTERS FOR BACTERIOLOGICAL CABINETS, ANIMAL CAGES, AND SHAKING MACHINE CONTAINERS

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To minimize the chance of injury or disease from toxic or infectious substances, laboratory personnel are increasingly confining hazardous techniques to enclosed ventilated work cabinets. The well established use of the fume hood for chemistry has been extended to other laboratory sciences. The Atomic Energy Commission has been particularly active in this progress.

The bacteriologist has recently become aware of dangers in techniques previously considered safe. Johansson and Ferris (1946) have demonstrated by high speed photography and air sampling that such operations as pipetting, pouring, and vigorous agitation of dilution blanks often produce bacterial contamination of the surrounding air and surfaces. Anderson et al. (1950) and others (Stein et al., 1949; Reitman et al., 1951; Stein and Rogers, 1946, 1950) have reported that flaming an inoculating loop, mixing broth cultures with a pipette by alternate suction and blowing, making microscopic slide agglutination tests, autopsy of infected animals, cooling a hot loop by immersing the loop in a culture, use of the lyophilizer, and other bacteriological manipulations release bacterial aerosols.

A survey of laboratory acquired infections in the United States conducted by Sulkin and Pike (1950) tabulates a total of 1,334 infections presumably acquired in the laboratory.

When a cabinet is used to confine accidentally released bacterial aerosols, it is desirable that the exhaust air be purified. Depending on the type of cabinet and volume of air, various methods may be used. One of the cheapest and most convenient is the use of spun glass\(^1\) air filters.

The use of spun glass as an efficient bacterial and viral filter has been reported by Decker et al. (1950). A one-inch layer was found to have an efficiency of 98 per cent or better in removing aerosolized clouds of *Serratia indica* and of *Escherichia coli* bacteriophage T-3. The readily available supply of spun glass, the low pressure drop across the filter pad, low cost, and the ease with which the spun glass can be installed in a filter frame suggested its use as a filter for bacteriological safety cabinets (Decker et al., 1951). A practical, compactly arranged, spun glass filter unit having a capacity up to 250 cubic feet per minute and an over-all resistance of 2 inches of water has been developed (figure 1). The over-all size of the filter cabinet is 2\(\frac{1}{2}\) by 3\(\frac{1}{2}\) feet by 8 inches. Tests have been performed using *S. indica* and *E. coli* bacteriophage T-3 to determine the filtration efficiency.

\(^1\) This material consists of fibers with an average diameter of 1.25 \(\mu\). It is manufactured by the Owens Corning Fiberglass Corporation and is known commercially as “aerocor PF 105”.

377
The test equipment is illustrated in figure 2. The organisms were nebulized into a cloud chamber where the cloud of bacteria or virus was mixed with air, then passed into the pre-filter sampling chamber, through the test filter at a face velocity of 20 feet per minute into a post-filter sampling chamber, and finally exhausted by means of a blower to the outside air.

Liquid impingers sampled the air at the pre-filter sampling point. When the anticipated recovery of the test organism exceeded 300 organisms per plate, liquid impingers were used post-filter also, since the liquid sample could be diluted and plated. When low concentrations of the test organism were expected, sieve samplers were used for post-filter sampling.

The liquid impingers were constructed with a critical orifice which permitted air to be drawn through the collecting media at approximately 0.5 cubic feet per minute. When S. indica was the test organism, the collecting medium consisted of 20 ml of nutrient broth and 6 to 8 drops of olive oil. One-tenth ml of the sample was streaked on a corn steep agar plate. In addition, 1 ml samples from the liquid impingers were serially diluted, and one-tenth ml samples of the dilutions were streaked on plates for incubation and counting.

When E. coli bacteriophage T-3 was the test organism, 20 ml of tryptose phosphate broth were used in the liquid impinger, and this same medium was employed for serial dilutions. One ml of each dilution was mixed with 5 ml melted tryptose phosphate agar which had been inoculated with 0.3 ml of an 18 to 24-hour E. coli culture, and the mixture was poured on a substrate of

Figure 1. Spun glass filter unit
tryptose phosphate agar. After solidifying, the plates were placed in an incubator at 37 C, and plaques were counted at the end of 5 and 18 hours' incubation.

For detection of S. indica, air samples taken with the sieve type sampler were collected by drawing air through each sieve sampler at 1 cubic foot per minute. The organisms were impinged on corn steep agar plates in the sieve sampler, and the plates were then incubated and examined for growth.

When recovering coliphage T-3 particles with sieve samplers, tryptose phosphate agar plates were used in the sampler, and the organisms were impinged on the medium. Upon completion of the run, 5 ml of melted tryptose phosphate agar inoculated with 0.3 ml of an 18 to 24-hour E. coli culture were poured on the substrate, and plaques were counted at the end of 5 and 18 hours of incubation at 37 C.

RESUL TS

Table 1 shows the results of testing the spun glass filter for removal of S. indica from an air stream. The weighted average efficiency of the filter for an average concentration of 350 organisms per cubic foot of air was 99.68 per cent, while for a concentration of 69,650 organisms per cubic foot the filter efficiency was 99.95 per cent. Concentrations of phage particles varying from 30,870 to 298,000 per cubic foot of air were removed with an efficiency of 99.74 per cent. It is believed that these efficiencies are sufficient to permit use of this filter in all situations in which air containing minor aerosols incidental to common laboratory techniques is to be filtered and discharged to the air outside a building.

Consideration had to be given to protective measures required in changing the spun glass filter pads. To avoid exposure of personnel to contaminated filter material, it would be desirable to decontaminate the filter in place. Accordingly,
the filter unit was designed with six 250-watt strip heaters capable of heating the interior of the filter cabinet to more than 200°C. This unit could be connected to a standard 110-volt outlet since the total wattage did not exceed 1,500. To prevent heat loss, the 6 sides of the filter unit were lined with 4 layers of spun glass, which is excellent insulating material.

Time and temperature studies have shown that after the interior of the filter has reached 200°C, an exposure of 20 minutes is sufficient to destroy Bacillus globigii spores. To allow sufficient time for reaching and maintaining 200°C or higher, the thermostat installed on the filter unit was set at 230°C, and the heaters were operated for two hours. All tests showed complete destruction of B. globigii at the end of this time.

### Table 1

**Efficiency of two-layer PF 105 spun glass filter unit**

<table>
<thead>
<tr>
<th>SERIES NUMBER</th>
<th>NUMBER OF TESTS PER SERIES</th>
<th>Number of organisms per cu ft of air before filtration</th>
<th>PERCENT FILTRATION EFFICIENCY</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Maximum</td>
<td>Minimum</td>
</tr>
<tr>
<td>A</td>
<td>10</td>
<td>596</td>
<td>130</td>
</tr>
<tr>
<td>B</td>
<td>14</td>
<td>701</td>
<td>100</td>
</tr>
<tr>
<td>C</td>
<td>18</td>
<td>438</td>
<td>56</td>
</tr>
<tr>
<td>Avg</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Weighted Avg</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

|               |                             |          |         |         | 99.95                      |
| D             | 18                          | 102,500 | 10,200  | 39,800  | 99.95                      |
| E             | 19                          | 168,000 | 13,300  | 93,500  | 99.94                      |
| Avg           |                             |          |         | 69,650  | 99.95                      |
| Weighted Avg  |                             |          |         |         | 99.95                      |

| F             | 5                           | 298,000  | 30,870  | 91,600  | 99.74                      |

Data have also been obtained on the effect of moisture, steam, and chemical fumes, such as formaldehyde, on spun glass. It has been found that unless the air becomes saturated, there is no apparent effect on the efficiency of filtration. However, if there is more moisture in the air than is required for complete atmospheric saturation, the excess moisture can collect in the filter pad, causing conditions favorable to the growth of organisms. This characteristic is common to all filter materials unless environmental factors are such that growth of biological life is inhibited.

**Use of spun glass in animal cages.** It has been reported by Leavel and Amoss (1931) that brucellae can be isolated from the feces and urine of infected animals. This would indicate the possibility of airborne infection from animal cages. One way to minimize this danger is to ventilate cages in such a manner that the air is filtered as it enters and leaves. Use of a filter serves several purposes: (1) it provides clean air to the animals; (2) it lowers the possibility of cross contamination between cages; and (3) in the event of a failure of the mechanical air supply...
system, contaminated air does not escape but the animals still receive enough circulation of air through the filters to prevent suffocation. Figure 3 shows a photograph of the ventilated animal cage. The filter for incoming air consists of a bored rubber stopper with a grooved section into which are fitted a true-arc inside retaining ring, a 32 mesh screen, two layers of spun glass, a second 32 mesh screen, and a second true-arc ring. This arrangement provides a tight fit which prevents leakage of organisms between the spun glass and the sides of the rubber stopper. The air leaving the cage may pass through a similar filter or be piped to a larger filter unit.

![Mechanically ventilated animal cage](image)

**Figure 3.** Mechanically ventilated animal cage

*Use of spun glass in a shaking machine.* The breakage of culture flasks on a mechanical shaker is a problem which becomes a hazard in laboratories working with pathogenic agents. Loss of plugs from the culture flasks being shaken has also been reported. The possibility of accidentally dropping a flask during transfer to or from the shaker must not be overlooked. To avoid the dangers in these possibilities, a shaking machine container has been constructed (figure 4) which completely encloses the flask, yet provides sufficient air through the spun glass filter for satisfactory bacterial growth.

The shaking machine container is cylindrical and equipped with a spring device which securely holds a 250 ml or a 500 ml flask in place. The head is made of Pyrex glass and aluminum. A circular opening in the center of the head houses a spun glass filter held in position by Pyrex glass secured in a groove with a true-arc retaining ring. Several tests were conducted to determine whether enclosure
of a culture flask would affect the growth rate of bacteria. All results indicated that there was no effect upon the growth rate of the bacteria.

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FILTERS FOR BACTERIOLOGICAL CABINETS

SUMMARY

A spun glass filter unit having a capacity of 250 cubic feet of air per minute has been constructed and tested. The unit was found capable of removing 99 per cent of the test organisms from an air stream. This efficiency will permit the discharge of air, originally containing infectious bacteria and viruses, to the outside atmosphere, except when there are several million organisms per cubic foot of air. However, filtration efficiencies of 99 per cent cannot be considered adequate when the air is to be recirculated.

An animal cage having a spun glass filtered air supply has been developed. Use of this cage may prevent possible cross-contamination and avoid the release of pathogens to the atmosphere.

A shaking machine container is described which completely encloses the flask, yet provides through a spun glass filter sufficient air for satisfactory bacterial growth.

REFERENCES


