CONTAMINATION CONTROL
THROUGH FILTRATION OF MICROORGANISMS

TECHNOLOGY SUMMARY
(Task 12)

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by

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FOREWORD

This document is a state-of-the-art summary prepared by Exotech Systems, Inc. in partial fulfillment of Contract NASw-2062, under the cognizance of the NASA Planetary Quarantine office.

The initial purpose of the report had been to summarize the technological advances in the uses of filtration, which had been developed under the NASA Planetary Quarantine program. After a review of a preliminary draft, however, the scope of this paper was expanded to include the technological advances developed for non-space applications. This further information is reflected in our list of references and bibliography.

This state-of-the-art report should, therefore, be useful in evaluating the potential for new and further uses of the process of filtration in Planetary Quarantine applications.
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INTRODUCTION

Requirements for decontamination and sterilization of spacecraft have resulted in continuing investigations of gas and liquid cleaning systems, which can effectively control the spacecraft assembly environment. For the requirements of Planetary Quarantine, gas and liquid cleaning systems are of interest. They reduce the number of particles in the air of assembly room facilities, and on the surfaces of spacecraft hardware, hence lessening the microbial load to allow a relaxation of sterilization requirements. Among those cleaning systems, filtration is one of the simplest and most feasible.

Filters may be divided into two categories: those used for filtering gas, and those used for filtering liquid. Gas filters are generally used in clean rooms to aid in controlling contamination. They may also be used to assure a sterile supply of gas, such as nitrogen, which is used in conjunction with dry heat for sterilization purposes. Liquid filters are used to remove microorganisms from liquids that may be heat labile, or sensitive to chemical sterilants.

Filters have numerous non-space applications. The field of medicine and the pharmaceutical industry have used filters extensively and, as a result, have prompted advances in the state-of-the-art of filtration.

This report describes the various kinds of gas and liquid
filters, explains the filtration mechanisms, outlines the important characteristics of filter materials, and the factors affecting filter performance, summarizes filter testing and evaluation techniques, and relates the possible application of filters to the field of spacecraft sterilization.

A survey of the literature indicates that filters provide an efficient means of controlling contamination, so long as they are properly monitored and adequately maintained. The degree of monitoring and maintenance is commensurate with the efficiency required from the filter. For space applications, where maximum efficiencies are necessary, the requirements for continuous maintenance and monitoring could be a severe limitation, since these are definitely money and time consuming.

Appended to the report is a bibliography citing documents not directly referenced in the text. These documents are intended to serve the reader in obtaining more detailed information on specific aspects of the state-of-the-art on filtration.
I. GAS FILTRATION

A. COLLECTION MECHANISMS

The screening or sieving mechanism for filtering gases has very limited applicability because the interstices of a filter would have to be smaller than the smallest particles to be removed; the resistance to airflow correspondingly would be high (ref. 1). As suspended material accumulates on the filter surface, resistance increases, and ultimately airflow stops as all of the interstices become plugged. All practical gas filters consist of fibers of various materials oriented in such a way that most of the open spaces or interstices are much larger than the diameter of the particles to be removed. The filtering action depends upon the particles coming in contact with, and adhering to, the fibers or collecting surfaces.

There are several collection mechanisms that may cause airborne particles to impact on the fibers. These include inertial effect, diffusion, electrostatic effect, direct interception, and deposition in accordance with Stokes' law. The last two are less effective in removing particles by filtration than are the first three mechanisms (ref. 1)

B. GAS FILTERS

Gas filters may be divided into four categories according to their efficiency and use. These categories are: roughing filters,
medium efficiency filters, high efficiency filters, and high efficiency particulate air filters (HEPA).

Roughing filters are generally used when large amounts of contamination and debris are in the air. These filters remove the bulk of large airborne particles and about 10 to 60 percent of the bacteria and other particles of a similar size (1-to 5 micron ($\mu$m)* diameter). Roughing filters are also used as prefilter, to reduce loading of the higher efficiency and more expensive filters (ref. 2).

The two most commonly used types of roughing filters are the viscous-coated, and the dry. Viscous-coated filters are composed of woven metal screens or loosely packed fibers of animal hair, hemp, glass wool, or synthetics. The fibers are often coated with an adhesive substance, usually an oil, which aids in retaining the trapped particles. In certain cases, these filters are constructed for indefinite use and can be cleaned and reoiled when the fibers become loaded. A metal screen filter consisting of a metal screen belt that moves across the air stream is automatically cleaned by passing through an oil or water bath at the bottom of the filter unit, where the screen is cleaned and rewetted. The dust collects as sludge in the bath (ref. 2).

The dry roughing filter consists of loosely packed glass or other fibers, cotton batting, or paper. In general, it offers more resistance to the passage of air and has a higher filtration efficiency than

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* $\mu$m is used throughout this document to denote microns, according to the S. I. system of units.
the viscous type. However, dry filters cannot be recleaned and must be discarded when the resistance to air flow becomes excessive.

Medium efficiency filters remove 60 to 90 percent of the bacteria and other particles in the 1 μm to 5 μm diameter range. They are composed of compressed glass fibers or a good grade of paper fiber. Resistance to air flow is slightly higher than that of roughing filters and increases little when the filters are loaded with dust. Medium efficiency filters must be discarded when they are loaded. They are generally used where removal of large particles is desired, and relatively clean air, without a large reduction in flow rate is required. Medium efficiency filters are used also as prefilters to reduce loading of higher efficiency filters.

High efficiency filters remove 90 to 99 percent of all particles in the 1 μm to 5 μm diameter range. Their filter media are mainly glass fibers, good grades of fiber paper, and asbestos fibers. The diameter of the fiber ranges from 1 to 5 μm. Resistance to air flow is higher than that of medium efficiency filters, and increases considerably as the filter loading increases. The air flow resistance of these filters may increase fourfold or more before discarding is necessary (ref. 2).

High efficiency particulate air filters (HEPA) are used for maximum removal of small biological and radioactive particles from air. Their efficiency is greater than 99.99 percent for removing bacterial particles having a diameter of 1 to 5 μm. The HEPA filters
have a higher resistance to air flow than the less efficient filters but, like the high efficiency filters, their replacement is necessary only when resistance increases more than fourfold. Some of the materials used at present for HEPA filters are cellulose-asbestos fiber papers, ceramic fiber paper, compressed glass fibers, and composite beds of glass wool pads.

Although HEPA filters are excellent for removing all particles down to at least 1 μm, it is uneconomical to use them alone to remove large quantities of dust and other particles larger than 5 μm in diameter. The use of roughing, medium-efficiency, and possibly high efficiency filters, ahead of the more efficient and expensive HEPA filters, places the bulk of the loading on the less expensive filters, extends the life of the HEPA filters considerably, and reduces total operating costs.

C. CHARACTERISTICS OF MATERIALS TO FILTER GAS

Important among the characteristics of filter materials are: high filtration efficiency; low resistance to flow; maximum strength; low cost; and high porosity. For certain applications, the filter materials should also be resistant to heat and chemicals and should withstand the mechanical stresses to which they are subjected during manufacturing and handling processes (ref. 3).

Ideally, the filters must be capable of producing absolutely
clean air, of delivering a constant flow at the outlet, and be free of leakages both in the filter paper itself and in the installation. A high efficiency filter is required to remove an aerosol given to it because it is dangerous, toxic or simply troublesome (as in the case of air conditioning). Specifications for clean room environments require the air supply to be completely dust-free. This implies, among other things, that the filter itself must not be a dust generator which may produce particles by blow-off. To investigate this point, clean air is blown through the filter supplied by the manufacturer. The presence or absence of contamination caused by the filter under test is then measured in the effluent (ref. 4).

Laminar flow, as it is used in relation to filters, should not be understood in the Reynolds-number sense but as a property of an air flow, such as when a fine stream of smoke injected into a point flows out in a plume which opens gradually, but whose axis remains well defined and, in general, parallel to a given direction (in the case of a laminar flow from wall to wall, it would be horizontal). The system is one of micro-turbulence escaping along a known direction. To characterize it quantitatively, the velocity profile is drawn up with a non-directional device such as a hotwire anemometer. An average speed is thus obtained, integrated in a certain solid angle around the direction of flow. Federal Standard 209 (ref. 5) specifies that, in an actual filter installation, the velocity distribution must have extreme values not
diverging more than 20 percent from the average value.

Industrial filters are frequently less efficient than the paper of which they are made. Generally, this is due to the difficulties encountered when folding and fitting the paper into its case; sometimes it is a result of imperfect assembly of the filter into the filter frame. An aerosol of dioctyl phthalate (DOP) generated by atomization, is led from upstream and, point by point, by means of an optical particle counter, the dust in the effluent is investigated downstream of the filter. This gives an indication of any weakness of the cell, and also of the uniformity of its efficiency over its entire surface. If this uniformity is absent, it is taken into account later when making determination tests of the filtering efficiency.

D. FACTORS AFFECTING THE PERFORMANCE OF GAS FILTERS

Although aerosol particle size, aerosol charge, air flow, and exposure to high humidity have significant effects in filter performance, they are of secondary importance as compared with the manufacturer's quality control procedures. Performance of filter units is mainly a function of fabrication rather than the efficiency of the filter paper.

An evaluation of HEPA filters with submicron $T_1$ bacteriophage aerosols having a number median diameter (NMD) of 0.12 $\mu$m, and with aerosols of Bacillus subtilis var. niger spores with a NMD
of 1 µm was reported by Harstad (ref. 6). The filters used for this evaluation included all-glass, and glass-asbestos, HEPA filters. Each filter was given the standard dioctyl phthalate (DOP) test by the manufacturer, and was further scan tested with DOP for pinhole leaks.

Penetration of submicron phage aerosols through the filters increased markedly with an increase in velocity. Neutralizing phage aerosol with bipolar air ions resulted in two- to fivefold increases in penetration, with the larger increases occurring at the lower velocities. Penetration of bacterial spore aerosols through the filters was 0.00005 percent. The comparable value for submicron phage aerosol penetration was 0.00095 percent. Exposure to high humidity (> 95%) that simulated conditions under which filter units are decontaminated with steam-formaldehyde resulted, roughly, in a threefold increase in penetration.

No filter medium can be better than the tightness of its installation. Air seeks the path of least resistance; thus the volume of air bypassing a loosely installed filter bank will increase with the build-up of dust and associated resistance in the filter bed. At the same time, low manometer readings may give the erroneous impression that the filter bed is operating well below its maximum permissible pressure drop, when actually the effectiveness may be nil.

Unlike an element in a liquid piping system, a faulty air filter assembly shows no immediate sign of leakage. Because the load of particles entering the air intake during any short period of
time is small, dust and bacteria may accumulate before spilling over into a channel. If alternate moisture and drying exist at an upstream location, bacteria which have proliferated during the moist phase may strike the filter face en masse during the dry phase. Detection of leakage would thus depend either on prolonged sampling, or fortuitous sampling during a period of spillage, or on performance tests using a standard challenge of dust or bacteria.

E. CLEAN ROOMS

The foremost application of gas filters is in the supply of air for clean rooms. Raw incoming air is drawn through a blower system into modular plenum chambers, then forced through roughing or pre­filters to remove gross airborne contaminants (ref. 7). These pre­filters, usually of polyurethane foam construction, immediately precede the HEPA filters. By having the same outside dimensions as the HEPA filters, the prefilters can have a high efficiency rating and still not produce an excessive pressure drop.

Immediately downstream from each prefilter is a HEPA filter which constitutes the final barrier against the contaminated incoming air. This filter is generally composed of a pleated medium of fiberglass-asbestos separated by rows of corrugated spacers, which impart to the now virtually pure air a laminar outflow characteristic.

Prefilters are serviced at regular intervals, and this ex-
tends the life of the HEPA filters. However, when the flow rate through the HEPA filters falls to a prespecified level, they are replaced.

There are standards in the field of medicine which define minimum requirements for filter applications in hospital clean rooms. Reference 8 is a compilation of such standards. It calls for a minimum of two filter beds for ventilating systems serving sensitive areas such as operating rooms, isolation rooms and laboratory sterile rooms. The first filter bed should be located upstream of the conditioning equipment and should have a minimum efficiency of 30 percent. The second filter bed should be located downstream of the conditioning equipment, with a minimum efficiency of 90 percent. The exhausts from all laboratory hoods, where infectious or radioactive materials are processed, should be equipped with filters having a 99 percent efficiency.

Filter frames should be durable and carefully dimensioned, and should provide an airtight fit with the enclosing ductwork. All joints between filter segments and the ductwork should be gasketed or sealed to provide a positive seal against air leakage (ref. 8).
II. LIQUID FILTRATION

A. RETENTION MECHANISMS

Retention mechanisms of liquid filters may be separated into two principal steps: a step to carry particles across the streamlines so that they come adjacent to the pore wall, and a step causing the particles to adhere to the wall. The former is referred to as the transport mechanism, the latter as the attachment. Other actions within the filter pore which may be important in certain cases are flocculation, which may alter particles to be more amenable to transport or attachment, and scour, which may detach particles or aggregates of particles to return them into suspension.

The transport mechanisms include diffusional, gravitational, interceptive and hydrodynamic actions. Inertia, which is very important in gas filtration, has a negligible effect in liquid filtration. The attachment mechanisms include molecular forces, electrical double layers, and mutual absorption. Both transport and attachment mechanisms are explained in detail in reference 9.

B. LIQUID FILTERS

There are five main types of filters used to remove bacteria from liquids. They are the diatomaceous earth filters, the unglazed porcelain filters, the asbestos-pad filters, the fritted-glass filters,
and the membrane filters. In the past, industries such as the pharmaceutical, vaccine producers, and fermentation industries made use of asbestos pad and porcelain types of filters. More recently there has been a conversion to membrane filters.

Table 1 summarizes the types of liquid filters, provides examples for each type and includes an estimate of the minimum size of particles trapped in the filters.

C. REVERSE OSMOSIS

Reverse Osmosis is a membrane separation process. It is used for removing dissolved salts from aqueous solutions, or vice versa. The degree of removal or concentration is a function of the feed supply characteristics. It is not a process for the removal of suspended matter nor for fractionation (i.e. separation of one set of molecules from another).

From a given feed supply, two streams are produced: the permeate and the concentrate (see figures 1 and 2). The permeate constitutes the major stream, and passes through the membrane virtually free of colloidal, particulate, and microbiological matter. It has a relatively low content of dissolved organic and inorganic matter. The concentrate constitutes the minor stream in which the initial colloidal and particulate matter has been substantially concentrated by the barrier properties of the membrane (ref. 10).
<table>
<thead>
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<th>TYPES</th>
<th>EXAMPLES</th>
<th>MIN. PART. TRAPPED (μm)</th>
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<tr>
<td>1. Solid Fabrications</td>
<td>Scalloped Washers, Wire-Bound Tubes</td>
<td>5</td>
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<tr>
<td>2. Rigid Porous Media</td>
<td>a. Ceramics &amp; Stoneware</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>b. Sintered Metal</td>
<td>3</td>
</tr>
<tr>
<td>3. Metal Sheets</td>
<td>a. Perforated</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>b. Woven Wire</td>
<td>5</td>
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<tr>
<td>4. Porous Plastics</td>
<td>a. Plastic Pads, Sheets, etc.</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>b. Membranes</td>
<td>0.005</td>
</tr>
<tr>
<td>5. Woven Fabrics</td>
<td>Cloths of natural &amp; synthetic fibers</td>
<td>10</td>
</tr>
<tr>
<td>6. Cartridges</td>
<td>Yarn-Wound Spools, Graded Fibers</td>
<td>2</td>
</tr>
<tr>
<td>7. Non-Woven Sheets</td>
<td>a. Felts, Lap, etc.</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td>b. Paper: Cellulose</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td>Glass</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>c. Sheets &amp; Mats</td>
<td>0.5</td>
</tr>
<tr>
<td>8. Loose Solids</td>
<td>a. Fibers, e.g. Asbestos, Cellulose</td>
<td>SUBMICRON</td>
</tr>
<tr>
<td></td>
<td>b. Powders</td>
<td>&quot;</td>
</tr>
<tr>
<td></td>
<td>-diatomaceous earth</td>
<td>&quot;</td>
</tr>
<tr>
<td></td>
<td>-expanded perlite</td>
<td>&quot;</td>
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<tr>
<td></td>
<td>-inactive carbon</td>
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</tr>
<tr>
<td></td>
<td>-absorbent powders</td>
<td>&quot;</td>
</tr>
<tr>
<td></td>
<td>-coarse materials</td>
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Figure 1. Osmosis

Figure 2. Reverse Osmosis
Consequently, when there is a need for purified water, the permeate stream is of interest and importance. When the need is for concentration of dissolved salts in the original solution, e.g. to dewater without heat, to recover valuable solids or to reduce waste disposals costs, the concentrate stream is of importance.

The membrane used for reverse osmosis is a modified cellulose acetate film, approximately 100 $\mu$m in thickness. It is asymmetric and one surface has a somewhat dense layer, of approximately 0.2 $\mu$m, which serves as the rejecting surface. The remainder of the film is a relatively spongy mass. Approximately two-thirds of the weight of the membrane is made up of water. The film is generally cast from commercial grades of cellulose acetate (2.5 acetate with an acetyl content of about 39 percent).
III. FILTER TESTING AND EVALUATION

The evaluation of filters, used to sterilize liquids and gases encompasses not only the filters themselves, but the apparatus and procedures. In many cases, the filters were found adequate, but the associated equipment such as filtration assemblies, suggested for use, were found to be sources of failure, either because of inadequate seals or fragility. Procedural difficulties are also encountered in many cases because of handling problems with fragile filters and cumbersome apparatus. Reference 11 includes a detailed account of a series of tests conducted to evaluate high efficiency particulate air (HEPA) filters, membrane filters and filtration methods, filters and filtration methods for liquids under pressure, and filters and filtration methods for decontaminating gases under pressure.

There are a number of efficiency test methods which are presently used to certify satisfactory performance of the filters in actual service. The system shown in figure 3 is a typical test arrangement that permits accurate determination of the bacterial arrestment of filters, or media, prior to installation in an air filtration system. The test system is simple and can be set up quickly. The bacteria, usually B. subtilis var. niger spores, are nebulized into a chamber where the cloud of bacteria is mixed with additional air. The aerosol is then drawn into the duct, at the rated face velocity, through the filter under evaluation, and then exhausted through a blower to the
Figure 3. Test System for Determining Bacterial Arrestment of Filters
outside. Samples of the aerosol are taken before and after passing the filters.

In the dioctyl phthalate (DOP) test, a monodisperse cloud of the liquid of 0.3 μm drop diameter and 100 mg/m³ concentration is produced by condensation of DOP vapor. The smoke mixture is introduced into the filter at rated air-flow. Samples of this smoke are taken upstream and downstream of the filter. The efficiency is determined by comparing the concentration by a light scattering meter, and a photo-electric cell device known as penetrometer, which is sensitive to concentration differences of 0.001 percent. The readings are practically instantaneous, and efficiencies of the order of 99.99 percent may be measured. The test is exacting because liquid drops are more difficult to remove than slightly irregular shaped solid particles. The DOP apparatus is moderately expensive and requires careful operation and maintenance.

The methylene blue test is a British standard test. The test may be used over an efficiency range of 50 to 99.99 percent (ref. 3). A cloud of particles is continuously generated by atomizing a 1 percent aqueous solution of methylene blue, and allowing the droplets to evaporate to produce suspended solid particles. The sizes of the particles vary from 0.01 μm to 1.5 μm. This suspension is drawn continuously into the inlet duct with the air, and passed through the filter to the fan
and atmosphere. Baffles are located on each side of the filter to ensure mixing of the inlet and filtered cloud. Installed behind the mixing devices are sampling nozzles through which known volumes of air are drawn through $\frac{1}{2}$-inch esparto sampling paper. The stains are developed by exposure to steam, and their optical densities are found by means of a densitometer. The advantage of the methylene blue test is the simplicity of the required apparatus. It cannot be used, however, at high temperatures.

In general, filters can be evaluated and compared by efficiency determinations which use adequate and effective methods of microbial challenge; and by use of the velocity profile, which serves as a fingerprint of the filter characteristics.

Physical characteristics which could bias test data include dispensing methods, electrostatic charge on particles and dielectric surfaces, relative humidity, particle size and disturbance, and gravitational effects.
IV. FILTER RELIABILITY

A. GAS FILTERS

Results of tests of gas and liquid filters, primarily HEPA and membrane, have been reported in the literature (refs. 11, 12, 13 and 14). Most of the HEPA filters tested were found to be as claimed, i.e. 99.997 percent efficient for the removal of viable organisms in the size range of 0.3-1 μm. However, wide variations were generally observed in efficiency, in the effective filtration surface area, and in the velocity profile characteristics. Some of the HEPA filters have been found to be poorly constructed, resulting in edge leakage. In some designs, the effective filtration area is critically reduced by the sealing methods used, necessitating much higher velocities in the central regions to maintain the rated capacity. The latter condition increases the chances of filter matrix damage and poor air velocity distributions. This apparent lack of reliability strongly suggests the need to evaluate filters for a particular application rather than to rely entirely on manufacturers' recommendations.

B. LIQUID FILTERS

With the exception of metal membranes which, when tested, were found to be inefficient, the membrane filters were the most reliable of all liquid filters tested, provided that they were used in reliable apparatus designed specifically for this application. Membrane filter
holders were found to be poorly designed, and glass holders were least acceptable because of chipping and fragility. In general, membrane filters must be protected from sharp edges and must be fully supported to prevent damage and leakage. As is the case with HEPA filters, manufacturers' quality control is rather unreliable. This is evidenced by the great variance in filtration times which indicates a wide variation in pore size distribution in most membrane filters.
V. FILTER APPLICATIONS FOR SPACECRAFT STERILIZATION

The need to limit particulate contamination of spacecraft, either biological or inert, necessitates the use of filtration. During manufacture and assembly of a spacecraft filtration can provide an effective means of removing inert particulates which otherwise might cause malfunction of intricate components. Filters are widely used in clean rooms and areas where clean assembly is necessary. They may also be employed for the purification of air, nitrogen and other gases in spacecraft sterilization ovens, and for sterilization of gases and liquids before insertion into the spacecraft. Finally, liquid filters could provide a dependable device for the physical detection of microorganisms in potable water in space system water supplies.
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