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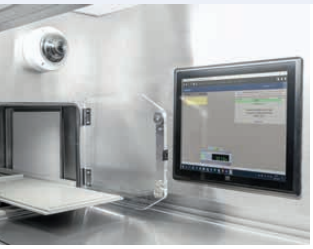

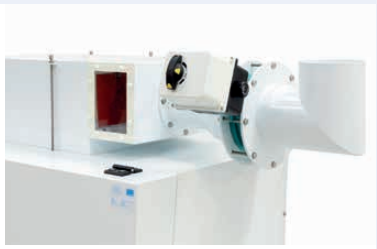

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Picture: The CDC F negative pressure isolator from Envair, with integrated medication workflow software

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Clean Air and Containment Review **is a quarterly journal aimed at users, specifiers, designers, manufacturers, installers and testers of clean air and containment equipment. It publishes articles of topical, technical and historical interest, updates on standards and regulations, news, views and information on relevant events, especially training.**

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EUROMED communications
County House, Easton Lane, Winnall,
Winchester, Hampshire SO23 7RU
T: +44 (0)1428 752222
F: +44 (0)1428 752223
e: publisher@euromedcommunications.com
www.euromedcommunications.com

Editorial

Welcome to CACR47, the first issue of 2022. The intention is to publish four issues in 2022. This issue consists primarily of three substantial and informative articles that examine three diverse and unrelated aspects of clean air and containment technology.

The first of these, from The Baker Company, in the USA looks at how containment in a biosafety cabinet is compromised by multiple users. Apart from making some strong points, the article is interesting for European readers because it casts some light on American practice. An open-fronted cabinet like the Class II BSC (biological safety cabinet) does not provide a perfect containment barrier. Therefore, it must be designed and operated with considerable care. I know, because I have seen it, that this is not always the case. I will never forget one highly qualified laboratory head, in a top hospital in a distant land, who proudly showed me her Class II BSC in use with the sash fully open!

The second main article is another sample chapter from Bill Whyte's latest book, Cleanroom Testing and Monitoring. This time it is Chapter 8: Filter installation leak testing by the photometer method. I hope, if you read this, you will get a flavour of the depth and clarity with which Bill covers his subject. The book forms the course material for the CTCB-I Cleanroom Testing courses which are run by the CTCB-I member societies in the UK, Ireland, the Nordic countries, Holland, Belgium and Turkey. For more information on the CTCB-I, please visit http://www.ctcb-i.net/index.php.

The third main article is a comparison of radiation methods for the sterilization of cleanroom items by the prolific and highly knowledgeable Tim Sandle, Head of Microbiology at the UK Bio Products Laboratory. The article describes the three main types of irradiation technologies and explains the relative merits of each. It is important that cleanroom users understand these and the implications when suppliers of sterilised items change from one method to another.

CACR47 concludes with the usual News items and Life-lines (rather more serious this time) as well as details of Events and Training.

John Neiger

Pearls of wisdom

It is generally assumed by BSC manufacturers that there shall be a single user at a time, yet BSCs are constructed in many sizes, with common widths varying from 3 to 6 feet. With that much work area, it could be assumed [wrongly] that multiple users could sit side by side and maintain the same level of protection as a single user.
Kara F. Held and Robert Thibeault, page 4

The air supply to a cleanroom should be free of significant amounts of airborne contamination. This is achieved by installing suitable air filters and, in particular, high efficiency filters (HEPA and ULPA) at the entry of the supply air to the cleanroom. However, leaks can occur in the high efficiency filter installations and allow unfiltered air to enter the cleanroom. How these leaks are located is discussed in this chapter.
W. Whyte, page 10

There is sometimes confusion over the terms 'radiation' and 'irradiation'. The former refers to different processes of transferring energy or it is a reference to the radioactive source; whereas irradiation refers to the specific process whereby an object is deliberately exposed to radiation. Irradiation does not necessarily equate to sterilisation (many food products are irradiated to extend their shelf-life, for example). It simply means a product was exposed to a radiation source. To achieve sterilisation by irradiation requires validation against the desired Sterility Assurance Level.
Tim Sandle, page 20

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EDITOR
John Neiger
T: +44 (0)1494 882094
M: +44 (0)7967 572958
e: jneiger@johnwrite.co.uk

EDITORIAL PANEL
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Gordon Farquharson (UK)
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SUBSCRIPTIONS
Jill Monk

Published by:
E C Pharma
County House,
Easton Lane,
Winnall,
Winchester,
Hampshire SO23 7RU
T: +44 (0)1428 752222
e: publisher@euromedcommunications.com
www.euromedcommunications.com

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Multiple users in a Biosafety Cabinet compromise containment

Kara F. Held and Robert Thibeault

This paper is reprinted with the kind permission of The Baker Company and is one of a series of white papers under the heading of 'Baker BSC Myth Busters.'

Abstract

Biosafety Cabinets (BSCs) are primary containment devices used to help reduce the risk of contamination to the work, the worker, and the surrounding environment. These devices have rigorous testing and certification to ensure their functional capabilities of contaminant reduction according to NSF International Standard 49 but these BSCs are always tested empty without users present in front of them for a functional baseline. However, BSCs are never used in this manner. There are known commonly performed actions within BSC operation that may compromise BSC containment. Here, we address one of these BSC myths, specifically whether two or more people can work within one BSC and still maintain their intended capacity for particulate containment as measured through visual smoke demonstration and NSF International Standard 49 Microbiological Aerosol Testing.

Introduction

Primary Containment for most biological laboratories starts with the Biosafety Cabinet (BSC). These ventilated enclosures are built and tested to rigorous specifications dictated by NSF International Standard 49 to provide containment of particulates, aerosols and biohazards through three mechanisms: Personnel protection, Product protection, and Environmental protection. This ensures that the user, the experiment or work being conducted, as well as the laboratory and building, are protected. This is achieved through the use of specifically directed and controlled velocities of air and High Efficiency Particulate Air (HEPA) filtration technology to remove particulates from the airstream.

How the air flows through a BSC will determine which class and type of cabinet it is. The three main cabinet classes are determined based on level and type of containment, or protection from biohazards, it will provide. Class I cabinets do not provide Product Protection and are generally referred to as "powder hoods" [in the USA but not Europe – Editor]. Class III BSCs are gas-tight, closed glove boxes that operate under negative pressure, designated primarily for high-risk biohazard work.

Class II cabinets make up the greatest population of BSCs found worldwide. This class includes 5 Types of BSCs: A1, A2, B1, B2, and C1. Class II Type A cabinets are recirculating, allowing for reduced energy usage and the A2 classification make up the vast majority of all BSCs (airflow flow patterning shown in Figure 1).

Type B cabinets must be hard-ducted or directly connected to a facility's exhaust system and provide fully exhausted air out of the BSC. Type C1 is a relatively new hybrid of the Type A and B cabinets allowing for flexibility of ducting outdoors or venting the cabinet to the room using dedicated zones in the work area and different installation protocols (NSF International, 2018).

The ability for a BSC to provide Containment is dependent on unobstructed airflow, as well as the velocity of the air coming in through the front access opening; however, when a BSC is in use, materials, tip boxes, pipettes, even users' arms will restrict airflow. Larger front access openings also require more air to be moved, with the blower compensating to keep the cabinet balanced.

All NSF International certified BSCs must be able to overcome at least some of these restrictions. The microbiological aerosol testing as described in NSF International Standard 49 accounts for some of this by directly testing how much aerosolized bacterial spores will exit the BSC (Personnel Protection test), enter the BSC (Product Protection test), or travel across the worksurface (Cross Contamination test) (NSF International, 2018).

The NSF Standard 49 provides testing criteria, usage recommendations for how to operate a BSC safely, but interestingly never mentions how many operators can sit at the front access opening at once.

It is generally assumed by BSC manufacturers that there shall be a single user at a time, yet BSCs are constructed in many sizes, with common widths varying from 3 to 6 feet. With that much work area, it could be assumed that multiple users could sit side by side and maintain the same level of protection as a single user. However, this had not yet been tested. Here we shall determine what level of protection multiple users would experience in a standard 6 foot Class II Type A2 BSC with both an 8 inch and 10 inch front access opening.

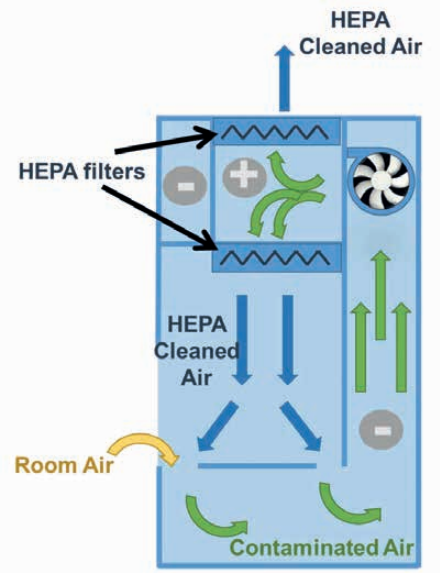

Figure 1: Sideview diagram of a Class II Type A2 BSC

Methods

Smoke visualization

To visualize airflow patterning within a Class II Type A2 6-foot wide BSC (Baker SterilGARD SG604), a Rosco Fog Machine (Model 1700) was outfitted with a 4 inch hose attached to a 6 foot PVC pipe with holes drilled every 2 inches to provide a uniform curtain of smoke.

This pipe was installed at eye level along the outside glass sash above the front access opening to visualize potential inward flowing air (Figure 2a).

Figure 2a: Construction of a smoke curtain at the front access opening with the mannequins present

Figure 2b: Placement modification of the NSF International Standard 49 Aerosol Microbiological testing set up in the presence of three users

Airflow measurements

Airflow into the cabinet was measured with a Direct Inflow Measurement (DIM) device (Shortridge Airdata Multimeter ADM-850L) on a capture hood attached to the front access opening when no mannequins were present. With mannequins at the front opening, a modified capture hood was created for the exhaust filter. Two to three mannequins were equally spaced along the front access opening to simulate multiple users at the BSC (Figure 2b). The stainless steel challenge cylinder used for NSF International Standard 49 Aerosol Microbiological testing to help break the front barrier was also included.

The exhausted volume of air could then be used to calculate the inflow velocity using the standard equation (**Equation 1**), where Q = volumetric flowrate, V = velocity, and A = cross sectional area of where the airflow is being measured, and $Q_{intake} = Q_{exhaust}$.

Equation 1. Q = V*A

The velocity of single streams of air were measured using a thermal anemometer or "hotwire" (TSI VelociCalc Plus Model 8385), such as around the mannequins or the downward flowing air in the work area.

Aerosol Microbiological Challenge testing

The containment capability of the BSC was tested using microbiological aerosols as described in NSF International Standard 49 (NSF International, 2018). Testing was split into three types: Personnel, Product and Cross Contamination testing. Both the sidewall and center Cross Contamination configurations were used (NSF International, 2016; NSF International, 2018). The collision nebulizers contained a slurry of *B. subtilis var. niger* spores (May, 1973); both nebulizers and Tryptic Soy Agar petri dishes were placed as directed in Standard 49 (NSF International, 2018), or as close to as written as possible.

The presence of the mannequins blocked some of the standard locations, and modifications were required. After the tests were conducted, all petri dishes were covered and placed in a 37˚C cell culture incubator (Baker Cultivo Ultra Plus). Results were read after 24 hours of growth, and pass/fail was determined according to the Standard (NSF International, 2018).

Results

Smoke visualization

The air patterning within a Class II Type A2 BSC has a standard airflow, where room air enters the front access opening at a minimum velocity of 100 feet per minute (fpm). This strong inward flow prevents escape of hazards from the work area creating Personnel protection. The air is then drawn up the back or side plenums by a motor/ blower system and pushed into an equalizing plenum. Here a portion of the air is exhausted through the exhaust HEPA filter, providing Environmental protection. The majority of the air is pushed through the supply HEPA filter in a unidirectional downward manner so as to completely and uniformly cover the work area. This creates Product protection. Disruption in the airflow in any kind can lead to loss of protection.

Figure 3: Smoke penetrating the work area in the presence of three users' hands indicating a loss of Product Protection

Supplemental Video 1 (click on picture): Loss of containment by smoke visualization in the presence of three users in front of a 6 foot Class II Type A2 BSC

Figure 4: Center Cross Contamination test (left) set up in the presence of three users

The mannequins were set up in front of the BSC and the changes to baseline air patterning was observed. With 2 mannequins, minimal disruption to the air flow was seen, mostly around the NSF stainless steel rod and the mannequin hands.

The further into the cabinet the hands were placed, the greater chance of some external air entering the work area. The greatest effect was seen with 3 mannequins placed at the front access opening. Great bursts of smoke were seen entering the work area which lead to the potential for contamination. Again, the most air flow disruptions were seen around the hands near the NSF stainless steel rod (Figure 3 and Supplemental Video 1).

Airflow measurements

The overall airflow through the BSC can be determined by measuring the volumetric inflow of air through the front access opening and comparing that to the total volume of air coming out the exhaust filter. The exhaust measurement is commonly greater than the inflow through the front access opening because this will include any additional air seeping in from around the viewscreen, through cable ports, and other unsealed areas as designed. All inward flowing air helps with Containment of potential biohazards.

The inward volume flow of air was shown to not be restricted by placing 2 or 3 mannequins in front of the cabinet front access opening where inflow was measured at 410 cubic feet per minute (cfm), and exhaust measured at 453 cfm at set point with no mannequins in front. This exhaust measurement was maintained with 2 mannequins at the front access opening and was only altered slightly to 450 cfm with the addition of the third mannequin (Table 1).

Aerosol Microbiological Containment testing

The 6 foot Class II Type A2 BSC (Baker SterilGARD 604) was first tested empty to the full criteria dictated by NSF International Standard 49 (NSF International, 2018) at both the 8″ and 12″ window sash opening with no mannequins in front to show a passing baseline for the cabinet. Once the BSC was determined to be properly functional, the BSC was balanced at an 8″ sash opening and tested with 2 mannequins spaced equally in the front access opening.

A wider front access opening will lead to a greater volume of air entering, which can be calculated using Equation 1. This greater volume of inflowing air can lead to potential loss of Containment if the BSC cannot properly control the air. When the 6 foot Type A2 BSC (Baker SterilGARD 604) was rebalanced with a 12″ window sash, the 2 mannequins at the front access opening caused a loss of Personnel, Product, and Center Cross Contamination protection within the BSC. Adding the third mannequin yielded the same result (Table 2). This is evidence strongly supporting the NSF International committee decision to change the Cross Contamination testing method from the sidewall to the center of the BSC work surface (Test configuration shown in Figure 4).

The BSC was able to pass the Personnel, Product and Sidewall Cross Contamination test; when the Center Cross Contamination test (NSF International, 2018) was conducted, however, there was a loss of Protection.

When a third mannequin was added to the front access opening, the Personnel Protection was maintained, but Product

Table 1: Volumetric Airflow measurements through the front access opening (Inflow) or out the exhaust HEPA filter (Exhaust) of a 6 foot Class II Type A2 BSC when user bodies are located at the front access opening

	Users in front of BSC	Inflow	Exhaust
8″ front access opening	0	410 cfm	453 cfm
	2	n/a	453 cfm
	3	n/a	450 cfm

Table 2: Aerosol Microbiological Containment testing results for multiple users in a 6-foot Class II Type A2 BSC at both an 8″ and 12″ window sash. Pass (green) and Fail (red) criteria determined by NSF International Standard 49 (NSF International, 2016)

		Personnel	Product	Sidewall Cross	Center Cross
8″ front access opening	2 users	PASS	PASS	PASS	FAIL
	3 users	PASS	FAIL	FAIL	FAIL
12″ front access opening	2 users	FAIL	FAIL	PASS	FAIL
	3 users	FAIL	FAIL	PASS	FAIL

and both Cross Contamination protection were lost (Table 2).

Conclusions

Biosafety Cabinets (BSCs) have historically been tested by certifying agencies such as NSF International to ensure they provide the intended level of Containment or protection from biohazards. The current tests performed require that the BSC be empty and free of obstructions with the intention that this will determine the BSC's peak level of performance. It is known that common laboratory practices may impede this peak BSC performance. One such practice would be the presence of multiple users at the front access opening. Here the magnitude of that impact and the subsequent consequences were determined.

While the volume of air entering the BSC was not impacted by the presence of bodies blocking the front access opening (Table 1), it was visually observed that the mannequin hands breaking the front access air barrier caused potential influxes of smoke into the work area of the BSC, which could lead to potential contamination and loss of containment (Figure 3 and Supplemental Video 1).

This was then confirmed with the microbiological aerosol testing, where failures in maintaining protection were widely observed (Table 2). When set with the most common configuration of an 8" window sash opening, 2 users at the front access opening successfully maintained Containment per the NSF International Standard 49 prior to 2018 (NSF International, 2016). However, with the addition of the new Center Cross Contamination test, a potential for spreading biohazards across the cabinet was uncovered.

This phenomenon was again observed with a 12" window sash opening, but with the addition of a loss of Personnel and Product Protection. As the window sash is raised and the front access opening area increased, the speed or velocity of air needs to be maintained at the required 100 fpm, so the volume of air entering the cabinet is increased.

With a greater volume of air comes the greater risk of contaminants entering the work surface as demonstrated with the results (Table 2).

The greatest Containment loss was observed by adding the third user at the front access opening. By having the third body blocking the directionality of air into the cabinet and the added two arms breaking the front access opening air barrier, there was a much greater risk of contaminants entering the worksurface (loss of Product Protection) as well as a greater risk of biohazards exiting the cabinet (loss of Personnel Protection, Table 2). Here again was observed the passing result in the Sidewall Cross Contamination test and a failing result in the Center Cross Contamination test supporting the NSF International committee's decision to change the test recommended in the latest edition (NSF International, 2018).

There are some laboratory practices that may require multiple people to work within the same biosafety cabinet at the same time, however the risks associated with such procedures should be appropriately known to both users and biosafety officers. It is recommended that only a single user operate a BSC at a time; if it must be done, a maximum of two users in a 6 foot BSC operating at least 14 inches away from each other is the next best option, assuming they take extra precaution to prevent Cross Contamination to each other and have conducted a Risk Assessment with the appropriate Biosafety Officer.

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Kara Held, Ph.D. brings an extensive knowledge of cell biology and pharmacology as the Science Director to The Baker Company. Prior to her coming on board, she was a Lab Manager, Safety Officer, and Researcher at the Harvard Stem Cell Institute studying Spinal Muscular Atrophy using iPS cell-derived motor neurons. She earned her Ph.D. at the University of Vermont Cell and Molecular Biology Program and completed her postdoctoral training at Yale University in the Vascular Biology and Therapeutics program studying mechanisms of blood vessel regulation. Her recent experience in academic research brings a unique perspective, familiarity and hands-on knowledge base with laboratory science and biosafety, which she now applies toward education and product testing as a part of the New Product Development team.

Bob Thibeault is the Senior Product Development Engineer of the Product Development Group at the Baker company with 20 plus years of experience. He is responsible for all aspects of new product development and testing, including mechanical and microbiological testing. Bob has had extensive experience in all avenues of manufacturing at Baker, including metal fabrication, assembly, engineering design, and test before moving into New Product Development. Bob's knowledge and history of biosafety cabinet design are an invaluable asset to Baker.

Cleanroom Testing and Monitoring, Chapter 8: Filter installation leak testing by the photometer method

W Whyte

This article is the second of a short series of extracts from Bill Whyte's new book Cleanroom Testing and Monitoring and is reproduced here with the kind permission of the author, Bill Whyte, the publisher, Euromed Communications, and the owner of the copyright, the Cleanroom Testing and Certification Board – International (CTCB-I). The objective in publishing these extracts is to give readers a flavour of the content and depth of the book which is recommended as a comprehensive textbook and an essential reference for cleanroom managers, cleanroom test engineers, cleanroom service engineers, cleanroom designers and specifiers and anybody who is concerned with cleanrooms. All too often testing and monitoring are insufficiently considered until an installation is physically complete. If you design and build an installation to achieve a certain performance, it is essential that you understand and plan at an early stage for how that performance will be verified and monitored throughout the life of the installation.
Editor

Chapter 8 Filter installation leak testing by the photometer method

The air supply to a cleanroom should be free of significant amounts of airborne contamination. This is achieved by installing suitable air filters and, in particular, high efficiency filters (HEPA and ULPA) at the entry of the supply air to the cleanroom. However, leaks can occur in the high efficiency filter installations and allow unfiltered air to enter the cleanroom. How these leaks are located is discussed in this chapter.

There are two methods used to locate leaks in high efficiency air filter installations that are described in ISO 14644-3: 2019 [ref 9]. These are by use of a photometer, or by a light scattering airborne particle counter (LSAPC). The photometer method is discussed in this chapter and the LSAPC method is discussed in Annex D.

Information on types of high efficiency air filters and their particle removal efficiency has been given in Chapter 3. In this chapter, it is assumed that the filters installed in a cleanroom have the correct particle removal efficiency and have been individually tested by the manufacturer to ensure they are free of leaks. However, owing to possible damage during transportation and installation, the high efficiency filters must be tested after installation. They should also be tested periodically, to locate any leaks that have developed over time.

8.1 Types of leak

The type of leaks found in high efficiency air filters are the type shown in Figure 8.1, and these will now be discussed.

A. filter media.

B. filter media-to-filter casing interface.

C. gasket or gel seals.

D. filter casing joints.

A – Leaks in filter media

Leaks can occur in the filter media. Many of these leaks are found where the filter media is folded to produce pleats.

B – Leaks at filter media-to-filter casing

Leaks can occur where the filter media pack is sealed into its filter casing and these are often in the corners. This type of leak is usually found when the filter is scanned during manufacturing, but damage can occur when the filter is installed. Uneven fitting of the filters into their housings or ceiling grids, and over-zealous tightening, can cause these leaks. Problems can also occur during use, such as damage by people walking on lightweight ceilings that causes the filter casing to flex. This can cause the sealant that holds the filter media to the casing to give way and allow leakage.

C – Leaks at gasket and gel seal

Leaks can occur at the seal between the filter and its housing or ceiling grid. The two systems used to prevent this are gaskets and gel seals.

Synthetic rubber gaskets: Gaskets are a common type of seal used between a filter and its housing, or between a filter and a ceiling grid. Show in Figure 8.2 is the arrangement when a filter is inserted into its housing.

Filter gaskets are usually made from synthetic rubber foam, such as neoprene, and are about 6 mm thick when uncompressed. They are glued onto the surface of the filter casing where it contacts the filter housing or ceiling grid. The filter is drawn down by clamps and the gasket is compressed so there should be no leakage between the filter casing and its housing or ceiling grid. Gaskets are also produced by dispensing foam onto the filter casing as a continuous gasket material. A thin film of silicon

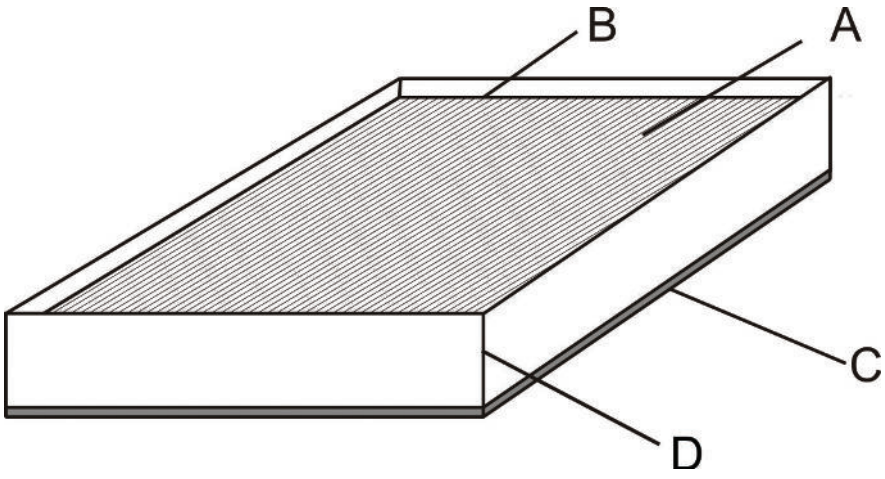

Figure 8.1: Leakage areas in a high efficiency filter

grease is often applied to the gasket surface when mounting the filter to help seal it and ensure that the filter comes away cleanly when the filter is changed. Gasket leaks often occur at corners. Leakage occurs because of poor quality or damaged gaskets, or because the mounting surface is distorted, or uneven.

Gel-seal method: In a UDAF cleanroom, filters are often installed into a suspended ceiling grid. Gaskets can be used, but an alternative approach is to use gel seals. In this approach, the ceiling grid is made from extruded aluminium channels which contain a jelly-like fluid that should not flow out of the channel. When a filter is fitted, knife-edges in its casing enter the channel and the gel flows round the knife-edge to prevent airborne contamination passing into the cleanroom. A typical arrangement is shown in Figure 8.3. Similar arrangements are available for individual filter housings used in non-UDAF cleanrooms.

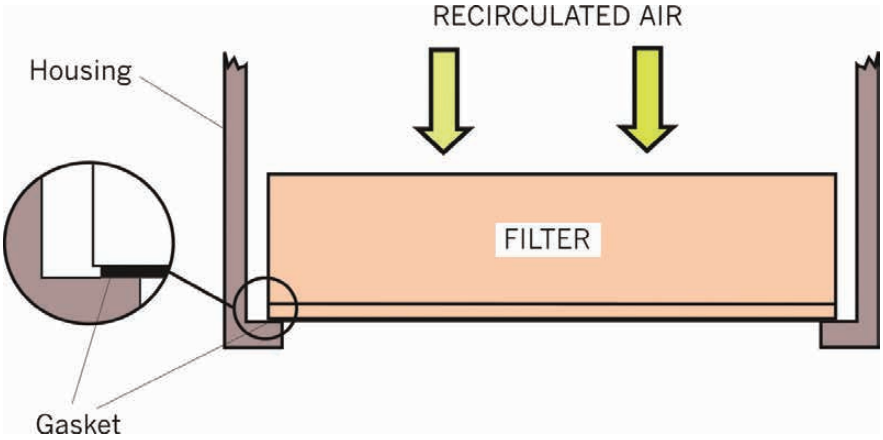

Figure 8.2: High efficiency filters with gaskets in a suspended ceiling grid. Filters inserted from above the ceiling grid

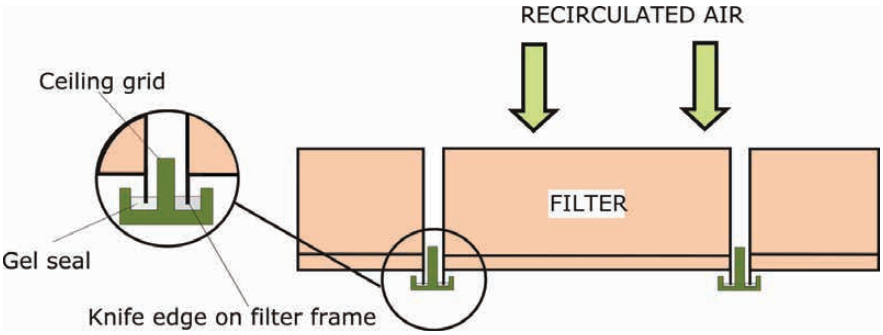

Figure 8.3: Filter housing gel-seal method

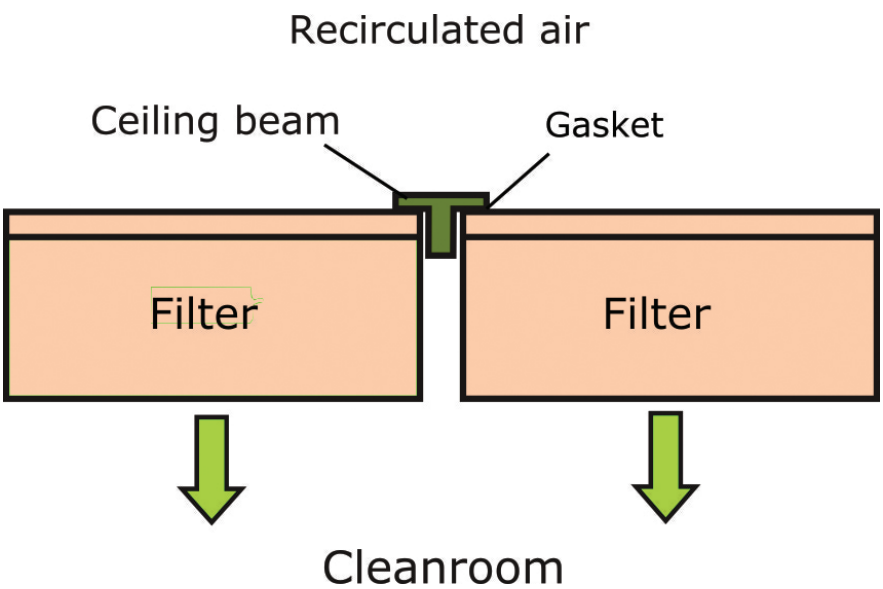

Figure 8.4: Ceiling (and gasket) leaks from a cleanroom inserted filter

D – Leaks from filter casings

Filter casings are made from a variety of materials but aluminium is a common material for high efficiency filters. If the casing is poorly manufactured, or insufficient care taken during transportation or installation, leaks can occur, often at joints. If the filter is inserted from above the ceiling, then, as can be understood from inspection of Figure 8.2 and Figure 8.3 that casing leaks are unimportant, as any contamination coming from leaks in the casing will have to pass through the filter before entering the cleanroom. However, if the filter is inserted upwards from the cleanroom (see Figure 8.4), then leaks of contaminated air from the casing can directly enter the cleanroom.

Testing high efficiency filter installations for leaks is usually carried out by challenging the filter with an artificial test aerosol. This test aerosol is introduced upstream of the filter at a suitable and even concentration. Any leaks are found by scanning downstream of the filter installation with a photometer probe to locate and measure any penetration of the test particles.

8.2 Requirements of leak testing

Type of aerosol challenge

There are two methods of generating artificial aerosols for testing filter installations with a photometer. These are either cold or hot (thermal) aerosols generated from specially selected liquids. Di-octyl phthalate (DOP) was the original material used to produce aerosols to test filters. However, because of reported toxic effects, it is no longer used in many countries, and oils with similar properties, such as poly-alpha olefin (PAO), di-ethyl hexylsebacate (DEHS) which is also known as dioctyl sebacate (DOS), and pharmaceutical and food grade oil such as Shell Ondina oil, are used.

In certain cleanrooms, such as these used for semiconductor manufacturing, inert test particles are specified. This is to ensure that no 'outgassing' of chemical products that are harmful to the product or process can come from test aerosol deposited on filters or air ducts. Microspheres, made from polystyrene latex are often used with an LSAPC. They are not discussed in this chapter but in

Annex D of this book, where the LSAPC method of locating and measuring filter installation leaks is described.

Aerosol generators

Laskin nozzle generators: To create a cold-generated test aerosol, a Laskin nozzle is used with an oil of the type previously discussed and compressed air at a pressure in the range of 140kPa to 170kPa (20-25psig). Figure 8.5 shows an aerosol generator with one Laskin nozzle, although generators are available with up to six nozzles. It should be noted that the oil level should be kept above the top of the nozzle.

The output of one Laskin nozzle is less than 0.5g/min when using an air pressure of 170kPa (25psig). This provides a concentration of approximately 10 mg/m^3 (10µg/L) in 0.8m^3/s of airflow, which can only be used to test a small air supply system like that found in an isolator, RABS, or UDAF workstation. To test a larger system, multiple nozzles are needed.

Thermal generator: The thermal generator uses an inert gas as a propellant, and oil of the type previously discussed is injected as a mist or aerosol into a heated evaporation chamber operating at an appropriate temperature. In this chamber, the oil mist is vaporised in the inert gas. When this mixture exits the generator and meets the cooling effect of the surrounding air, the oil condenses into an aerosol with particles of a size that is suitable for testing high efficiency filters. Particle size will be discussed in the next section.

Thermal generators are able to generate the large quantities of test challenge needed for high volume air supply systems and, for that reason, they are often used in preference to Laskin nozzles. A thermal generator does not need an air compressor to generate the high pressure required by a Laskin nozzle, although it needs a cylinder of inert gas, usually carbon dioxide or nitrogen, as the propellant. The inert gas is used to remove the flammability risk in the heated chamber where the heater block operating temperature is above the flashpoint of the oil. If the challenge aerosol from a thermal generator is to be introduced into a positive pressure duct or plenum, then a separate fan or blower is required. The generator is not connected directly to the intake of the duct or plenum but an air blower draws in condensed aerosol from the generator and mixes it with ambient air and pushes it into the duct or plenum. This arrangement is shown in Figure 8.6.

Thermal generators can typically produce from about 1g/min to around 20g/min of aerosol. If a test challenge of about 10mg/m^3 is required for use with a photometer, then sufficient aerosol will be generated to test a ventilation system with an air volume supply rate of about 30m^3/s.

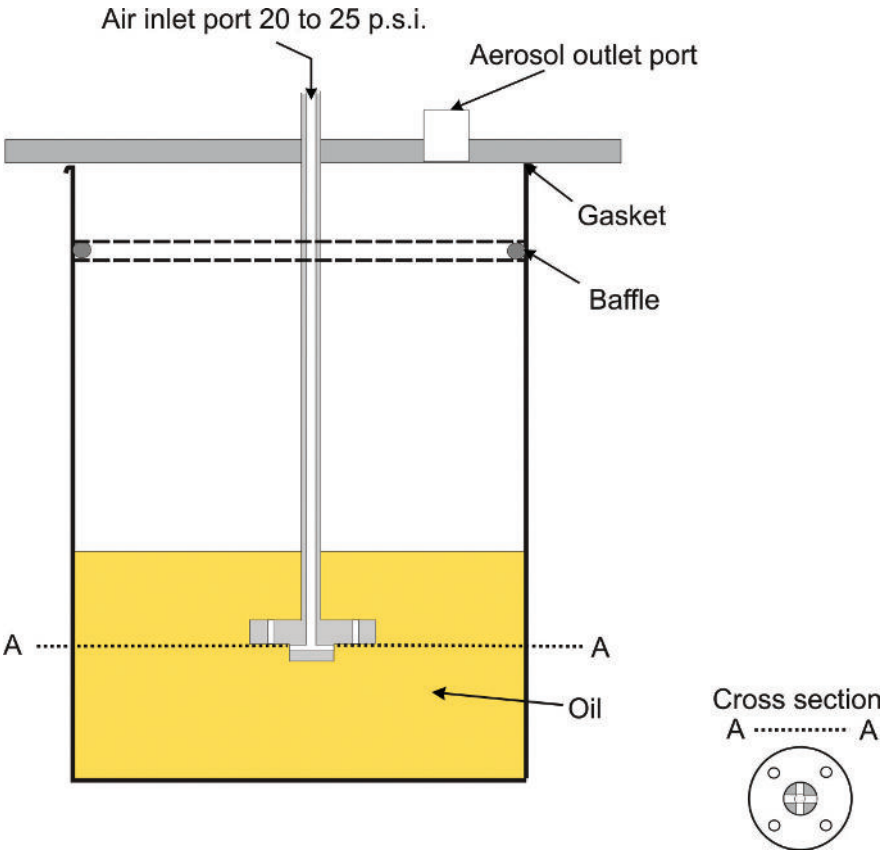

Figure 8.5: Cold aerosol generator with Laskin nozzle

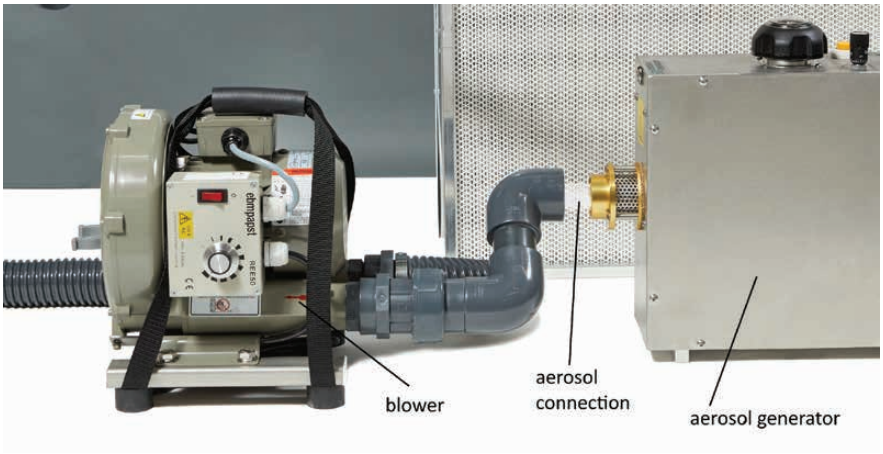

Figure 8.6: Thermal generator passing smoke to an air blower

Size distribution of the test challenge particles

ISO 14644-3: 2019 suggests that the mass median diameter of the aerosol test particles used to test high efficiency air filters will typically be between 0.3 µm and 0.7 µm with a geometric standard deviation of up to 1.7 µm. The FDA Guidance [ref 14] suggests that *'the challenge involves use of a polydispersed aerosol usually composed of particles with a light-scattering mean droplet diameter in the submicron size range, including a sufficient number of particles at approximately 0.3µm. Although the mean is normally less than one micron, it is greater than 0.3µm'.*

The above specifications are achieved by both a Laskin nozzle and a thermal generator, although thermal

generators usually produce a slightly smaller size of particle.

Photometers

A photometer is used in conjunction with a sample probe to locate any high concentrations of test particles that indicate a leak. An air sample is drawn from the probe into the photometer (see Figure 8.7) and as the airborne particles pass through a beam of light in the photometer, they scatter light. The amount of forward-scattered light is measured by a photomultiplier tube and converted into an electrical signal. This signal is shown on the display panel of the photometer in the required units of measurement (mg/m^3 or µg/L); the more particles, the higher the signal and displayed value. It is worth noting that the value of mg/m^3 is identical to µg/L but ISO 14644-3; 2019 uses mg/m^3.

A photometer usually measures a test particle concentration of between 0.0001mg/m^3 and 100mg/m^3. It measures the total amount of light reflected by the particles and is, therefore, different from an LSAPC which sizes and counts each single particle.

The general method of using a photometer to locate leaks is as follows:

1. Ensure all switches and connections on the instrument are in their default positions. Switch on the instrument, and ensure the sample selection switch or valve is in the required position,
2. Set up the instrument according to the manufacturer's instructions,
3. Measure the upstream challenge using the valve selector switch,
4. Set the instrument to measure the percentage filter penetration and the concentration of the upstream challenge aerosol (mg/m^3) as the 100% reference,
5. Scan the surface of the filter for leaks. When a leak is located, the penetration is calculated as follows by the instrument and shown on the display as a percentage of the challenge aerosol.

$$Leak\ penetration\ (\%) = \frac{Y}{X} \times 100$$

where:
Y is the measured leak in mg/m^3,
X is the average upstream challenge concentration in mg/m^3.

6. If the penetration reading is over the agreed value, which is usually 0.01%, a leak has been found. Its location should be noted.

Probe type

ISO 14644-3: 2019 recommends two standard types of probe. The most common is known as a 'fish tail' probe, with intake dimensions of 1cm x 8cm. This type of probe is shown in Figures 8.7 and 8.8. The other type of probe is a circular probe with an intake diameter of 3.6cm. These probes are used with photometers that have an air sample flow rate of 28.3L/min (1ft^3/min).

Scanning with probe

The scanning speed of the probe across the filter face is important. If the scanning speed is too quick, a leak can be missed. ISO 14644-3: 2019 suggests a scanning speed of approximately 5cm/s. The scanning should be carried out by means of overlapping passes over the filter face. The overlap should be about 1cm and the probe held 3cm, or less, from the filter face or filter installation.

What is a filter leak?

It is necessary to decide what particle penetration of the filter installation is considered a leak. ISO 14664-3:2019 suggests a penetration exceeding 0.01% of the upstream concentration should be used for filters with overall efficiencies of ≥99.995% (as classified by the ISO 29463 [ref 1] and EN 1822 [ref 2] method). However, if the overall efficiency of the filters is ≥99.95% and <99.995%, then the acceptable penetration is 0.1%. Where the overall efficiency is <99.95% then the penetration that defines a leak should be decided between the customer and supplier.

Figure 8.7: Photometer being used with probe and printer

8.3 Method of testing filter installations

Preliminaries

Before starting any testing with an aerosol generator, it will be necessary to consider whether smoke alarms can be set off by spillage of smoke. If this is likely to be a problem, it is best to turn off any smoke alarms rather than suffer the embarrassment of the arrival of the fire brigade. Sealing the alarms (temporarily) with plastic film and tape, so that they cannot detect the aerosol particles, is another possibility. Alarms that are set off by a temperature increase can be ignored.

Release of test challenge particles

The test particles that challenge a filter should have an even concentration across the back of the filter. If this is not done, uneven concentrations may cause the filter to incorrectly pass or fail the leak test. ISO 14644-3: 2019 suggests that the variation in the aerosol concentration over time should not exceed +/–15 %. However, the same standard provides no information about the allowable variation in the challenge concentration across the back of the filter, although it is often considered that this should also not exceed +/–15%.

In some situations, the test aerosol can be introduced just after the air conditioning plant. If this is done, then by the time the aerosol reaches the terminal high efficiency filters, it should be well mixed and the concentration of test particles across the back of the terminal filter will be even. However, if the aerosol is injected into the ductwork leading to the filter, it should be introduced at a distance no closer than 15 to 20 duct diameters before the filter. To ensure good mixing, the test aerosol should be injected into the centre of the duct but a good method of introducing the test aerosol is to use a pipe across the duct that has a series of holes along its length. This pipe is known as a 'sparge' pipe and is shown later in Figure 8.13. The evenness of the filter challenge should be measured at multiple points before the filter before testing starts.

Prior to scanning a filter, it is necessary to set the concentration of test particles in the air immediately upstream of the filter. A concentration of between 1mg/m^3 and 100mg/m^3 should be used. To reduce the potential for blockage of the filters with test particles, it is best to use a concentration at the lower end of this range but this should be consistent with the capability of the photometer, as not all photometers can work at lower concentrations.

The length of tubing between the aerosol generator and its entry point to the ventilation system should be as short as possible. This will minimise particle deposition in the tubing and changes in the particle size distribution.

Scanning for leaks

Once it has been established that the particle concentration behind the filter is even, the concentration should be measured by the photometer and its reading is set as the 100% challenge. The filter face can then be scanned for leaks which will show as >0.01%, or other agreed penetration.

The normal scanning method is to use a probe to scan over the whole filter installation for leaks. It will be necessary to decide where to start scanning. It is usually best to start at the gasket area, rather than the filter face, so that any spillages of test particles from the gasket area do not show up as false leaks in the filter media. After checking the gasket, it is best to move on to check the sealing between the filter media pack and its casing, and then finish with the filter face.

Scanning for gasket leaks: There are different types of gasket leaks to be considered. These are gasket leaks from filters inserted from above the ceiling of the type shown in Figure 8.2, and gasket leaks from cleanroom-inserted filters of the type shown in Figure 8.4. Scanning for gasket leaks in a filter inserted from above the ceiling is a relatively simple procedure as the gasket interfaces are visible and accessible. Scanning a cleanroom-inserted filter for gasket leaks is a more difficult procedure and this is now discussed.

A filter that is inserted from the cleanroom into a ceiling grid or housing will have a space between the filter and adjacent filters where the air is stagnant. To locate a gasket leak in these situations can be difficult, and Figure 8.9 illustrates the problem in a cleanroom-inserted filter in its housing. The particles from a gasket leak will spread out and fill the space between the filter and its housing and give a high concentration of particles that can cause

Figure 8.8: Scanning a filter

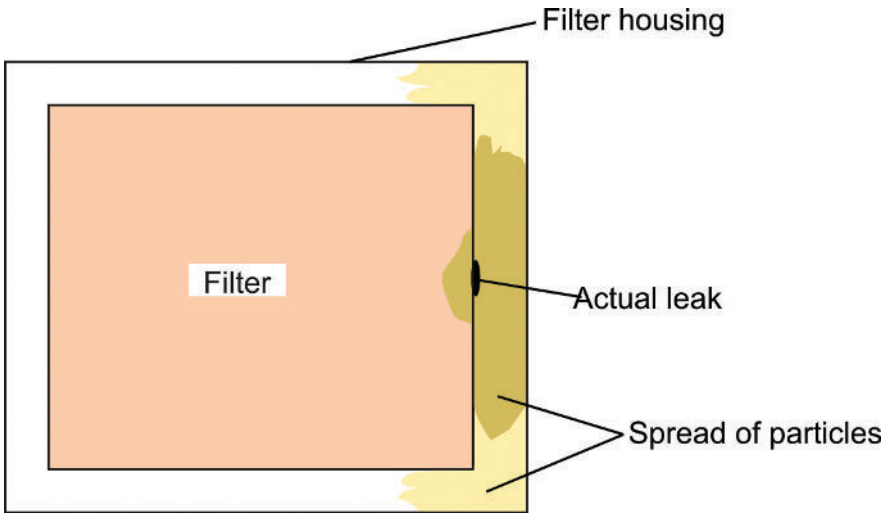

Figure 8.9: Spread of particles from a gasket leak in room inserted filter

the position of the leak to be thought to be away from its actual position. Ascertaining the exact position of the leak is time consuming and may be considered unnecessary but, if required, the probe from the sample tube can be removed and the tube used to locate the leak, although this will give a penetration value greater than had a probe been fitted. To assist in the location of the gasket leak, a particle-free jet of air can be used to clean out particles but a few wafts of air from a baffle plate may be sufficient.

If a gasket leak is found, the filter should be removed, and the seating area of the housing or ceiling grid inspected for distortions or unevenness. If the seating area is satisfactory, the filter can be re-seated using a thin layer of silicon grease. If that does not succeed in stopping the leak, the filter gasket should be replaced; this could be the first option. If that does not succeed, mastic could be carefully used between the gasket and the housing or ceiling grid. If none of these suggestions work, the filter, filter housing, or section of the ceiling grid may need to be replaced.

Checking for leaks from filters that use the gel seal method may also be required. The expectation is that gel seals will be free of leaks, but that may be incorrect.

Distinguishing between gasket and casing leaks: A difficulty with cleanroom-inserted filters is that, if there is a leak in the casing, it can be difficult to distinguish from a gasket leak. This has been illustrated in Figure 8.4. Access to the casing to check for leaks is also a problem. A method that can be used to locate leaks in the filter casing is to use a test rig similar to that shown in Figure 8.10. This allows access for scanning for leaks round the casing when the filter is challenged by a test aerosol.

The test rig shown in Figure 8.10 can also be used to speed up the fitting of filters when a cleanroom has a large area of filters to be tested. Each filter is tested on the rig for leaks in both the filter casing and the filter media. The filter is then carefully fitted into its housing or ceiling grid and, when all filters have been fitted and the ventilation switched on, the filters can be checked for gasket or gel seal leaks.

It should be noted that if the test rig is used in a cleanroom during construction and the ventilation has not been switched on, the cleanroom air will have a high particle concentration that might interfere with the recognition of leaks. In that situation, the test rig should be contained in a temporary clean enclosure which can be assembled from 25mm square box steel sections that is covered with plastic film or sheet. There is no need to ventilate the enclosure, as air coming from the filter being tested will produce a low particle concentration.

Scanning the filter face: When scanning the filter face, the probe should be held 3 cm from the filter face and the filter scanned with overlapping strokes at 5 cm/s. If a leak is found, it will be necessary to return to the leak, and pass slowly over it, so the exact position can be ascertained. If a fish tail probe is used, it is common to scan in one direction, turn the probe and scan again at 90° to the original direction so as to locate the position of the leak. It may be necessary to pass slowly over the leak several times from different directions before the exact position is ascertained. Removing the probe and using only the tube is another way of accurately locating the position of the leak. However, this method is likely to give a higher value of the leak penetration and the fish tail probe should be used to obtain the true value.

If the filter face is scanned when there is also a leak at the gasket then, as previously discussed, particles from the gasket leak can spill over onto the filter face. This can occur in the right hand

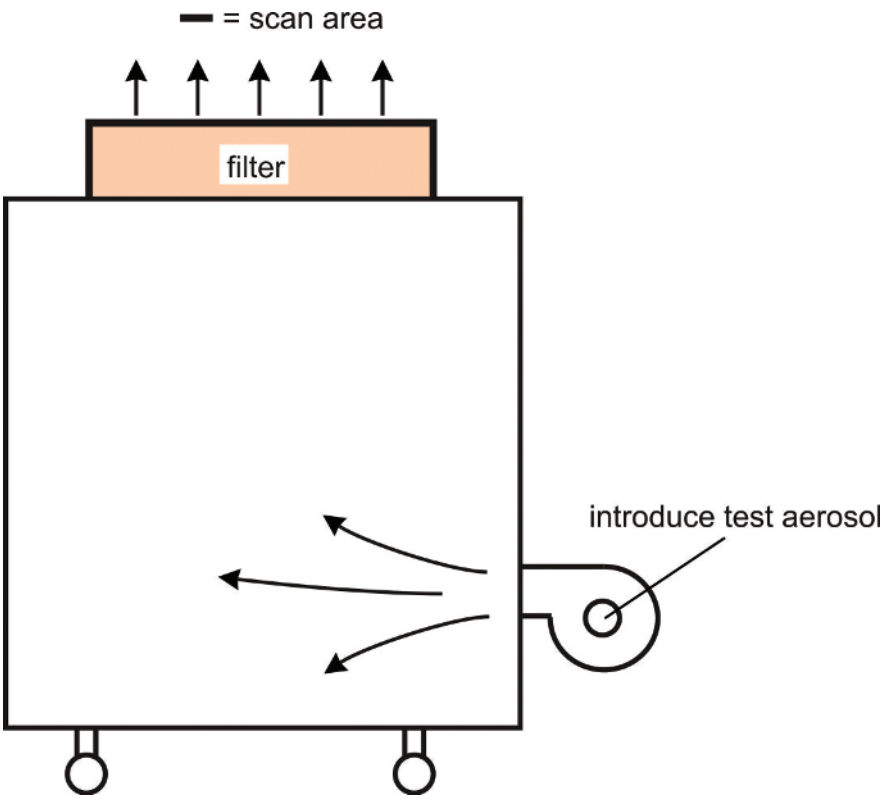

Figure 8.10: Test rig for distinguishing leaks

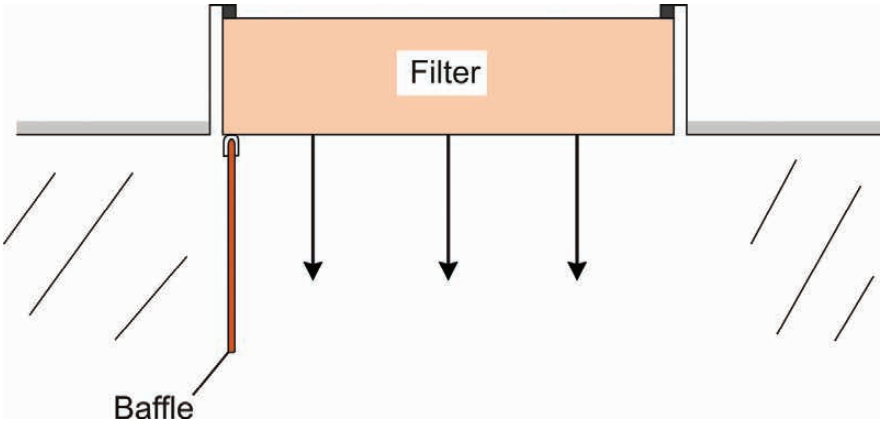

Figure 8.11: A baffle plate used when scanning a filter with leakage coming from the gasket

side of the drawing in Figure 8.11. Leaks can then be erroneously reported as being from the filter face. This problem can be largely overcome by use of a baffle plate held on the filter casing as the adjacent filter face is scanned. This approach is shown in the left hand side of Figure 8.11.

8.4 Influence of filter face velocity on particle penetration

The chance of a leak being found in a high efficiency filter is influenced by the filter face velocity. If the filters are used at a higher velocity than when tested during manufacture, the filter may unexpectedly fail the leak test. The reason for this is shown in Figure 8.12.

Figure 8.12 shows the removal efficiency of different particle sizes at different air velocities through filter media used in a HEPA filter. It should be noted that the velocities are through the filter media and not from the complete filter. Figure 8.12 shows that doubling of the velocity can reduce the filter removal efficiency by almost 10 fold. This information may be an explanation of an unusual failure of the leak test.

8.5 Additional considerations when testing filters

Testing filters in non-UDAF cleanrooms

If a ceiling diffuser is used to distribute the air supply in a non-UDAF cleanroom, it should be removed. If this is not done, it will be impossible to satisfactorily scan the filter installation for leaks. Scanning should then be carried out as previously described.

Testing filters in UDAF cleanrooms

A problem encountered when checking a complete filter ceiling of a vertical UDAF cleanroom is that the total area of the filter face is so large that leak testing can take several days. This testing may be carried out just prior to the cleanroom being opened for manufacturing and the testers can be under pressure to quickly complete the testing. To reduce the testing time, it is possible to scan by use of several photometers placed on a trolley, with their inlet probes 3cm from the filter face, and carrying out scanning by moving the trolley, which can be motorised, about the room.

It is also possible to reduce the testing time by scanning each filter on a rig of the type shown in Figure 8.10 to demonstrate that the filter face and casing is free of leaks. The scanned filter is then carefully placed into the ceiling grid. When the whole filter ceiling is in place and the ventilation system switched on, the gaskets or gel seals can be checked for leaks.

Testing filters in UDAF workstations, RABS and isolators

Isolators, RABs and UDAF workstation filters can be tested using the methods previously described. However, because of the short distance between the location where test aerosol is introduced and the filter, it may be difficult to obtain an even challenge across the back of the filter. Tapings into the plenum behind the filter in at least two positions that are well spaced apart should be provided by the manufacturer of the clean air device so that the evenness of the aerosol challenge can be checked. If they are not provided, they can be installed while on site.

The test aerosol is often introduced into the air intake of a clean air device with the hope that the fan will mix the aerosol so it is uniform across the back of the filter. This will often not work, and it may be best to introduce the test aerosol by means of a manifold or sparge pipe that will spread the test aerosol across the intake (Figure 8.13). Sparge pipes have small holes of about 2mm diameter drilled at regular intervals along the length of the pipe in order to spread the test challenge across the rear of the filter, or filters. A manifold or sparge pipe that is permanently fitted in an isolator, RABS, or UDAF workstation is a useful design feature.

8.6 Repair of leaks

The ISO 14644-3: 2019 standard accepts that repairs can be made to any part of the filter as long as this is acceptable to the customer. The FDA guidance suggests replacement of the HEPA filter or, when appropriate, a repair of a limited area of the filter. ISO 29463 suggests that 5% of the filter face area can be repaired, although the maximum length of repair should not be greater than 3cm.

An effective repair can often be achieved between the media pack and the filter casing, and between the filter casing and the housing or ceiling grid. However, the repair of filter media leaks may be difficult to implement and, because of blockage, can have adverse effects on the uniformity of airflow. In a non-UDAF cleanroom, the air supply will quickly mix with room air and a repair may be satisfactory. However, in UDAF workstations, the unidirectional flow of air may take airborne contamination from the leak directly to a critical area and, in this situation, it may be best to replace the filter.

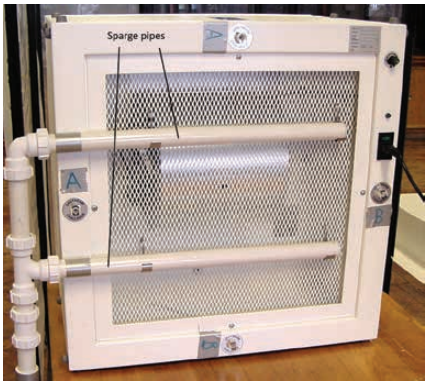

Figure 8.13: Sparge pipes used to spread the test challenge in an air intake

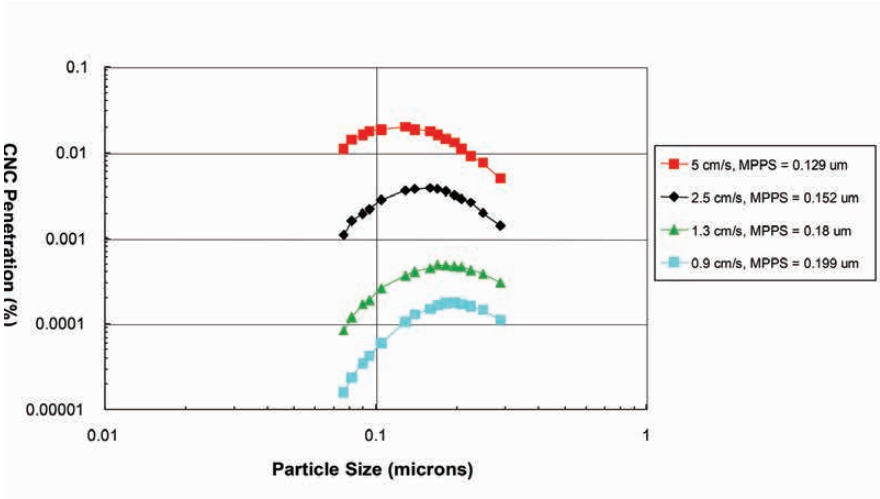

Figure 8.12: Filter penetration with respect to particle size at different velocities

Acknowledgement

Figures 8.6 and 8.7 are reproduced by permission of Air Techniques International.

CTCB-I (Cleanroom Testing and Certification Board – International) is an association which promotes, prepares and accredits internationally recognised educational courses for people who design, construct, test, monitor, operate and work in cleanrooms. Only societies set up for the education and promotion of contamination control techniques in cleanrooms can apply for membership of the CTCB-I. They must run or wish to run CTCB-I courses. Current members of the CTCB-I are Belgian Cleanroom Workgroup (BCW), Contamination Control Network (CCN), Cleanroom Technologies Society of Turkey (TTD), Irish Cleanroom Society (ICS), Cleanrooms and Contamination Control Association for Denmark, Finland, Norway and Sweden (R3 Nordic), Scottish Society for Contamination Control (S2C2) and Netherlands CC Society (VCCN). The CTCB-I is run by a Board of Delegates comprising delegates nominated by each member society. The Board of Delegates monitors the written and practical content of the cleanroom courses and the standard of examinations to ensure the maintenance of a common and high standard across the courses, and evaluates the course structure and teaching material from each new submission from a cleanroom society. The aim of the CTCB-I is to help foster the development of cleanroom practitioners in its member societies so that they practice to a very high standard. For further information please visit http://www.ctcb-i.net

Dr William (Bill) Whyte is an Honorary Research Fellow at Glasgow University and has the useful dual qualifications of a BSc in microbiology and a DSc in mechanical engineering. He has been involved in the design, testing, and operation, of cleanrooms and hospital operating rooms for over 50 years.

Bill Whyte has published over 140 journal articles on the design of cleanrooms and operating theatres, and the control of the transmission of contamination within them. He has written two books titled 'Cleanroom Technology – Fundamentals of Design, Testing and Operation' and 'Advances in Cleanroom Technology', and edited the book 'Cleanroom Design'.

He was founder and former chair of both the Scottish Society of Contamination Control and the Cleanroom Testing and Certification Board – International. He is a member of BSI and ISO working groups that are writing, or have written, cleanroom standards. He has extensive experience as an industrial consultant and presenter of educational courses about cleanrooms.

He has received the following awards for his work in Cleanroom Technology: Fellowship of the IEST, Honorary Life Member of S2C2, James R Mildon Award from the IEST, Michael S Korczyneski Grant from the PDA, Parenteral Society Annual Award, and Special Commendation Award from the British Standards Institution.

Comparison of radiation methods for the sterilisation of cleanroom items

Tim Sandle

Astract

Many items entering cleanrooms, especially aseptic processing facilities, are required to be sterile. Sterilisation by an ionising radiation source is the most common way of achieving sterility and is used for cleanroom garments as well as for plastic items intended for single-use, such as sampling containers, aseptic connectors, biocontainer bags, and mixers. For several decades, Gamma radiation was the standard method for the irradiation of cleanroom consumables. In more recent years, both electron beam and X-rays have been added as ionising radiation methods for the processing of cleanroom items.[1] The increase in use of these latter technologies is evidenced by change control notifications from cleanroom product companies indicating a move away from Gamma radiation to these alternative radiation sterilisation methods (primarily electron beam but sometimes X-rays). The shift away from Gamma is because the process of radioactive decay utilises costly isotopes which require periodic replacement (typically five years) and because international transport for radioactive materials has become far more difficult.

This article looks at the similarities and differences between the three types of irradiation sterilisation technologies commonly used to irradiate items going into the cleanroom: Gamma, E-beam, and X-ray.

Irradiation technology

There is sometimes confusion over the terms 'radiation' and 'irradiation'. The former refers to different processes of transferring energy or it is a reference to the radioactive source; whereas irradiation refers to the specific process whereby an object is deliberately exposed to radiation. Irradiation does not necessarily equate to sterilisation (many food products are irradiated to extend their shelf-life, for example). It simply means a product was exposed to a radiation source. To achieve sterilisation by irradiation requires validation against the desired Sterility Assurance Level (as discussed below).

The advantage that irradiation technologies have, for each of the three radiation sources, is a relatively fast sterilisation time compared with other sterilisation processes such as heat. In addition, the technologies, once qualified, provide a reproducible level of sterility assurance. There is also a safety element in that no radiation remains on or within the treated product (the sterilisation stops as soon as the radioactive source is removed, and the item does not remain radioactive due to insufficient energy having been passed through the object).[2] In addition, no chemicals are used for the sterilisation which contrasts with gaseous ethylene oxide sterilisation or hydrogen peroxide vapour decontamination where it is necessary to remove potentially harmful chemical residues at the end of the sterilisation process. Finally, the irradiation technologies are 'cold' sterilisation technologies in that any heat imparted to the product to be sterilised is minimal and this reduces the risk of physical damage to the product being sterilised.

The general requirements for sterilisation using radiation are provided in ISO 11137-1: 2006[3] (this standard is currently under revision), which most compendia and regulatory guidances reference. To achieve sterilisation using radiation, most compendia specify a Sterility Assurance Level of 10^{-6} or greater. The Sterility Assurance Level is a probabilistic concept (since sterility cannot be tested, only predicted).[4] A Sterility Assurance Level of 10^{-6} indicates that the possibility of microbial survival is no greater than one organism in one million (or one item in one million not being sterile). This target level of sterilisation is achieved through the radiation dose. The radiation dose is the quantity of radiation energy absorbed by the product as it passes through the radiation field during processing. The radiation dosage is not absolute and must be optimised given the properties of the sample and the level of sterility required. The unit used for the radiation dose is the 'Gray' (a dose of 1 Gy means 1 joule of radiation energy has been exposed to in each kilogram of material).[1] Radiation doses for cleanroom consumables and medical devices are often around 25 kGy. However, other doses may be required in relation to the complexity of the product and the microbial bioburden. Different microorganisms have different levels of resistance to radiation, and the same microbe may react differently depending on age, temperature of growth and ability to produce spores. Furthermore, the more organisms there are then the less effective a given dose will be and consequently the dose may need to be increased (which means bioburden determinations are an important part of the validation process). The microbial challenges notwithstanding, in practical terms 25 kGy of Gamma radiation is the same as 25 kGy delivered by an electron beam accelerator or by x-rays.

The dose is set by calculating both the minimum and maximum dose and then setting the standard dose above the minimum and below the maximum. To assess this range requires an understanding of the number of items (and their size and volume) required to be sterilised; the minimum dose to achieve sterilisation; and the dose above which there is a risk of product damage – the maximum dose.

To demonstrate this, the dose at which the product is irradiated must be established and validated (as per ISO11137-2: 2012).[5] There are different approaches to validation, the most robust being the assessment of three batches of product and characterising the microbial bioburden on the product in each batch pre-sterilisation. Characterising the bioburden is a combination of assessing the microbial numbers on the product and the resistance (decimal reduction value) of the microbial species recovered against the intended sterilisation method. This

"Gamma radiation is the oldest established irradiation method, and it is a form of electromagnetic radiation (the same as X-rays, although Gamma is more energetic)."

requires thirty samples to be tested (ten from each of three different production batches). The method to demonstrate bioburden recovery must be qualified to demonstrate whether or not there are any inhibitory substances leading to an underestimation or complete suppression of microbial growth (a stasis test), as per ISO 11737-1:2018/ Amendment 1:2021.[6] Post-sterilisation, a sterility test is required on each of the samples, as described in ISO 11737-2:2019.[7] The reason for a sterility test being part of the assessment is because of the exponential relationship between the numbers of microorganisms surviving and the extent of treatment with the sterilising agent, which means that, theoretically, a microorganism might survive regardless of the extent of treatment applied. To pass, each of the samples must record no growth of any microorganisms.

Once the sterilisation process has been established, quarterly 'dose audits' should be performed to assess the bioburden on samples of product to assess whether the bioburden levels remain comparable to the validated product. Changes to product design or to the manufacturing process typically require full revalidation. For routine sterilisation, irradiation is assessed by dosimetry where dosimeters measure the absorbed radiation dose delivered to the product and the dose distribution patterns throughout the product package.[1]

Achieving microbial kill is relatively straightforward. Arguably, the complexities of radiation sterilisation rest with assessing material compatibility. Considerations here include:[1, 8]

1. Is the density of the product so great as to affect radiation penetration?
2. Is the process time too long for the radiation time to be commercially viable?
3. Will the radiation process lead to oxidation? (Oxidative damage, such as arising from chain scission or molecular disruption within a polymer, can affect the bonds of plastic additives, weakening them).
4. Will the radiation process increase the likelihood of vulnerability to leachables and extractables if the product comes into contact with a pharmaceutical liquid product?
5. Will the process make the product brittle? (Embrittlement requires assessment across the shelf-life of the treated product).
6. Will the radiation process change the colour of the treated product? (Such as changing a plastic from white to yellow, which can arise through the cross-linking of polymers).
7. Does the outer packaging remain intact following irradiation?

Furthermore, some product types present greater challenges due to their size, their requirement to be disassembled, their antibacterial nature, or their adhesive properties.

Gamma rays

Gamma radiation is the oldest established irradiation method, and it is a form of electromagnetic radiation (the same as X-rays, although Gamma is more energetic). Radioactive isotopes such as cobalt-60 and cesium-137 are used as the sources of energy (with cobalt-60 being the most commonly used - a synthetic radioactive isotope made by neutron activation of cobalt-59). Radiation is produced as the isotopes undergo radioactive decay over time. For example, cobalt-60 radioactively decays to Nickel-60 and as it does so the isotope emits two gamma rays of different energies (as measured by MeV).[1]

The isotope is provided by the nuclear industry as a solid radioactive metal moulded into a cylindrical slug. The slugs are loaded into tubes of stainless steel which are then sealed. The stainless steel allows the gamma photons (the radiation) to pass through. The tubes are arranged into flat panel arrays and radiation is continuously emitted. When not in use, the arrays are stored in concrete and held within a deep-water pool, which prevents gamma radiation from escaping.[9]

Electron beam

The electron beam (or E-beam) technology utilises high-energy electrons and these are orientated towards the product to be sterilised. The product is bombarded with high-energy electrons, leading to a cascade of electrons penetrating through the product. The electrons are produced by an electrical current passing through a tungsten or tantalum filament. Through the use of an accelerator, the electrons increase in velocity to a speed close to the speed of light. To achieve sterilisation, the E-beam irradiation is operated at or less than 10 MeV (million electron volts, as a measure of energy).[1] There are two types of accelerator in common use: DC accelerators and accelerators based on radio frequency power technology.[10]
An accelerator consists of:

- Voltage generator
- Acceleration tube and electron gun
- Scan chamber and scan horn
- A control system

"The electron beam (or E-beam) technology utilises high-energy electrons and these are orientated towards the product to be sterilised."

The most important part of this sterilisation process is with the orientation of the electron beam. Here the beam needs to be of a defined size and generated in a sweeping motion to produce a curtain of electrons (the E-beam emerges through a thin window and is swept from side to side by a changing magnetic field, which transforms the tight beam cone into a wide curtain of electrons). The product to be sterilised is conveyed through the electron curtain at a pre-determined speed and duration, calculated to achieve the required level of sterility assurance. The accelerator is contained within a concrete structure to contain the emitted radiation. The main operational weakness is with the dissipation of electrons from the beam, leading to

wasted energy. Unlike Gamma radiation, the process is switched on and off through the activation and deactivation of the accelerator.

As well as sterilising products, E-beam applications include both the strengthening of polymers (creating cross-linking) and the breaking down of polymers.

X-rays

X-rays are photons, similar to Gamma rays. However, X-rays require generating and this is done using an electron beam accelerator (as with E-beam). The difference from E-beam is the conversion of the electron beam into X-rays. The X-rays are generated by interposing a metal target (such as tungsten and tantalum) between the electron beam and the product to be treated. The sudden slowing down or deflection of the charged particles creates what are termed bremsstrahlung (braking radiation) X-rays.[11]

X-ray sterilisation has emerged as a practical alternative irradiation method as a result of technological advances allowing for increased beam power ratings from industrial electron accelerators. This greater power enables the generation of X-ray beams of sufficient intensity to sterilise an object. One reason why X-ray may be preferred to E-beam is because X-rays are more penetrating than E-beam, allowing for the sterilisation of denser products.[12] To achieve sterilisation, the X-ray irradiation is operated at or less than 5 MeV. There is a greater loss of energy during the conversion process each time the process is run, compared with E-beam or Gamma making it more expensive to operate, therefore X-rays sterilisation is better suited to larger volume processing where processing on a larger scale means more items can be sterilised for each individual conversion process.

Comparison of irradiation methods

The irradiation technologies differ in terms of how they are produced, the extent of penetration achieved, the time taken to achieve the required level of sterilisation, and the effect on different materials. These differences are outlined in Table 1. With the generation of radioactivity, the isotope used in Gamma irradiation emits radiation in all directions and it cannot be turned off. In contrast, with electron accelerators no radioactive materials are involved (ionizing energy is produced electronically within the vacuum tube).[13] Even though the ionizing radiations come from different sources, they achieve the same biological effects. Microbial inactivation or kill is achieved through pushing enough energy into the microbial cell in order to break down cellular chemical bonds, including DNA. Other damaging effects upon microorganisms include rupturing cell walls and damaging proteins.[14]

The differences presented in Table 1 can assist purchasers of cleanroom materials in weighing up the advantages and disadvantages of the three irradiation methods, particularly when a producer notifies of a change in sterilisation method. In terms of the principal disadvantages, these are:[1]

- Gamma rays: low dose rate leads to slower productivity.
- E-beam: limitation in penetration.
- X-ray: less efficient for energy utilisation.

A generalised comparator for process time is that for a product processed by Gamma rays that takes 2.5 hours to sterilise, the same level of sterilisation could be achieved in 45 minutes using X-rays, and in under 10 seconds using E-beam. While the rapidity E-beams stands out, the process selected needs to be balanced with cost and compatibility of the product to the sterilisation process (both in terms of density and potential damage arising from the sterilisation). Users of low-density plastics might wish to switch to E-beam, but it should be noted that a switch from Gamma to X-ray and vice versa is easier than a switch to E-beam. This is because similar product loading patterns can be used for both technologies and the minimal dose and maximal dose tend to be similar.[15]

Conclusion

This article has provided a description of many of the factors that need to be considered when selecting between irradiation technologies as well as providing a summary of the three types of irradiation and the main differences between them.

The three different irradiation technologies are each effective at achieving the required Sterility Assurance Level under ideal conditions and within a relatively rapid timeframe (as compared

Table 1: Comparison of the three methods of irradiation

Irradiation technology	Dose rate	Exposure time	Processing rate	Penetrative ability (relative)	Field distribution	Homogeneity of source
Gamma	Low	Longest exposure time (hours).	Multiple product loads on complex conveyor system	High (most suitable for very dense materials).	Gamma rays are produced in all directions.	High
E-beam	High	Shortest exposure time (minutes).	Single product loads via simple conveyor system.	Low (the weakest penetration of the three technologies at 38 mm.	Unidirectional beam that needs to be targeted at the product.	Low
X-rays	Low	Medium exposure time (minutes to hours; longer times required compared with E-beam)	Sequence of single product loads, via complex conveyor system.	Medium (good penetration for dense products).	Forwards direction (but less narrow than with E-beam).	Medium

to sterilisation by dry heat, for example). The complexity is with selecting the most appropriate irradiation technology. This requires an assessment of the bioburden of the product as part of validation, an assessment of the effect of the sterilisation process upon the material properties of the product, and an understanding of the absorbed dose, material thickness, processing rate, and cost.

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Risk of material damage	Material compatibility	Process variables
Medium (some plastics are incompatible due to embrittlement, discoloration, or change in viscosity).	Suitable for most materials.	Time Load configuration Product configuration
Low (very few materials are damaged).	Suitable for most materials.	Beam orientation Beam energy Beam power Conveyor speed Product configuration
Medium (while some plastics are affected as with Gamma - embrittlement, discoloration, or change in viscosity, they are affected less severely).	Suitable for most materials.	Processing time Number of passes required to sterilise a product Product configuration

Tim Sandle, PhD, CBiol, RSB, FIScT is Head of Microbiology at the UK Bio Products Laboratory. He is a visiting tutor at both the University of Manchester and UCL. In addition he sits on the Pharmig committee, is Editor of GMP Review and runs a blog: Pharmaceutical Microbiology: www.pharmamicroresources.com

Environmental Monitoring Systems from ATI

Air Techniques International (more commonly known as ATI) is proud to report recent successes in winning projects for Environmental Monitoring Systems to support critical life sciences applications at Hammersmith Medicines Research, Catalent Pharma Solutions, Advent Bioservices and ThermoFisher Scientific. The order book is healthy and prospects good.

ATI supports the client right through from initial enquiry through completion using the well-established validation sequence of URS, FS/DS, DQ and FAT/SAT after which we configure the sensors and graphics and prove GMP compliance during installation and operational qualification (IQ/OQ).

Our systems use the GAMP qualified LMS Pharma real time monitoring system from Lighthouse Worldwide Solutions and then include an attached network of sensors for continuous monitoring of airborne particles, temperatures (from -80degC freezers to +50degC incubators), room humidity, pressure differences and other special parameters such as microbiological sampling or CO2 measurement.

We are proud to work closely with everyone involved to bring each project to a successful handover and then continue to support with training programmes, and mobile service and calibration throughout the UK.

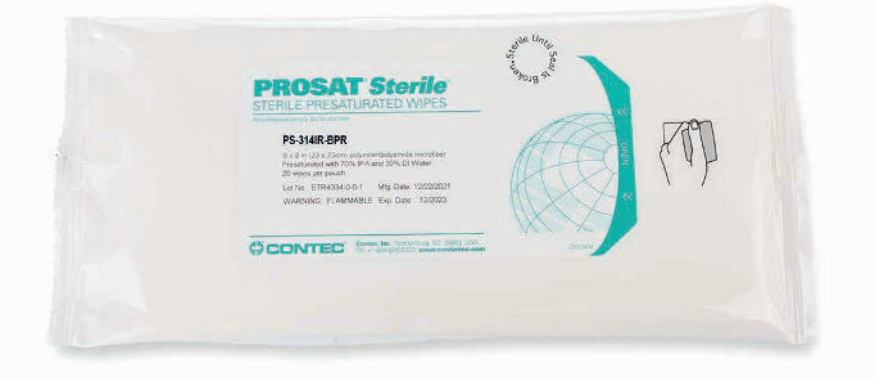
Contact us or come and see our own facility in Letchworth which has a LMS Pharma system monitoring our less critical operations that is easy to access and demonstrate. Email salesuk@atitest.com or call 01462 676446, our team can help you establish your new or next Environmental Monitoring System.

PROSAT Pi – A new presaturated microfibre wipe from Contec

Contec's new PROSAT Pi wipes are manufactured from 80g/m2 hydroentangled microfibre and presaturated with 70% IPA and deionised water. The wipes are made from a polyester and nylon split microfibe, enhancing their cleaning effectiveness and ability to pick up contamination.

The microfibre fabric is highly sorbent and also offers a metered release of the alcohol to the surface. This metered release ensures the surface coverage per wipe is greater than other comparable weight, presaturated wipes. These wipes can help a manufacturer control contamination whilst consuming less resources.

When used as a disinfectant, the IPA wipes are efficacious against bacteria in 1 min and yeasts in 3 mins. PROSAT Pi wipes are authorised for sale in the EU and United Kingdom under the EU and GB Biocidal Products RegulationFor more information or to request a sample, go to www.contecinc.com/eu/products/prosat-pi-microfibre-wipes/



SAS Daily Heads from Cherwell can help reduce risk in Environmental Monitoring

Cherwell Laboratories is highlighting the SAS Daily Heads as a viable way to help reduce risk and ensure compliance to the new stringent risk management and contamination control strategy within the revised EU GMP Annex 1.

The new Annex 1 states Grade A limits for biocontamination to now be zero, making equipment suitability a decisive factor in supporting this new tighter limit. The sterility of the sampling head is a critical factor in the effectiveness of EM devices. Daily Heads remove the need for a validated sterilisation process and substitute this with a certificated sterile product which provides a viable alternative to reduce risk of false positives.

SAS Daily Heads are certified sterile disposable air sampler heads designed for use with SAS Super, SAS Duo 360 and SAS isolator active air samplers. They are ideal where autoclave turnaround times or sterilisation facility validation can prove challenging.

Many users deploy Daily Heads for sessional monitoring, where the sampler remains in the controlled space. Reducing not only the possibility of any false positive events but reducing transfers in and out of the controlled space.

For more information about Cherwell Laboratories, please visit www.cherwell-labs.co.uk.

PMT (GB) Ltd introduce the portable air sampler with a d50 value of less than 1.0µm and a higher Biological Efficiency

The ISO90M Air Sampler is based on slit-to-agar technology which has very low d50 values of around 0.5µm with exceptional biological efficiency and the ability to count real viable events in CFU.

It is designed for use for continuous monitoring in high-grade areas, where in-process monitoring of viable particles is critical and where ISO-14698, CEN-17141 and GMP Annex 1 compliance is specified.

The d50 value, which is the minimum diameter of the particle at which a sampler is 50% efficient, represents only the physical efficiency. The biological efficiency, which expresses the rate at which the organisms are kept alive is equally important.

GMP Annex 1 now states you must have zero growth in Grade A areas and that puts even more emphasis on having the best biological efficiency, so you need to make sure your instrument can detect down to the right level.

Tests have shown that the biological efficiency of a Slit-to-Agar instrument can be up to 2.5 times higher than that for a standard Sieve Sampler.

For more information on our range of Slit-to-Agar air samplers please contact PMT (GB) Ltd at info@pmtgb.com

CCN runs successful CTCB-I Cleanroom Testing Course

March saw the first of three CTCB-I Cleanroom Testing courses scheduled for 2022 take place at the Mercure Letchworth Hall Hotel, Letchworth, UK.

As usual, the course covered filter integrity testing, measuring air velocities and volumes, and particle testing and was run at two levels, Associate and Professional. Both levels covered the same ground including a written examination, but those registered for the Professional course stayed on for additional practical examinations and reports. The primary reading material for both levels of the course was Bill Whyte's recent comprehensive book Cleanroom Testing and Monitoring.

Three identical test rigs were used with filters and housings very kindly donated by Camfil, and fans, ductwork and controllers purchased by CCN. Presenters and examiners were Tim Triggs from ATI, Kevin Beauchamp from CHTS, Billy Bailey from Camfil and Steve Ward and Mike Thomas from Validair. A total of 15 delegates including resits completed the Professional course and five completed the Associate course including one remote virtual candidate from Lithuania.

The initial feedback from all involved was that the course was a success on all counts.

The next courses will be run in April (fully booked) and November. For more details see https://www.theccnetwork.org/pages/61-events

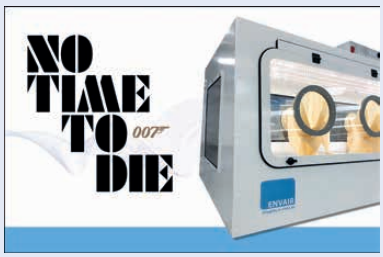
Film set consultant? An Oddjob for Envair

Design consultant to the latest Bond film is one of the more unexpected roles you'll find on Gary Bagshaw's CV. But back in May 2019, a phone call out of the blue turned into an exciting project for the MD of Envair.

A Set Decorator working on "No Time To Die" wanted to get hold of an isolator she'd seen in images of Porton Down. By coincidence, the isolator she had on her storyboard was one that Envair had installed 20 years ago. To recreate the look, Gary and Product Specialist Paul Rigby supplied a batch of original engineering drawings, and the team set to work to build a high-tech cleanroom on the sound stage at Pinewood.

Although they'd been widely consulted, and even met Daniel Craig, Gary and Paul only saw the outcome of their work when the film was released late last year. As Gary commented, "It's good to know that when you're manufacturing killer nanobots, we're the containment specialists of choice!"

For more information about Envair, please visit www.envairtechnology.com



Study of single use device media dehydration and biological recovery from PMS

The BioCapt® Single-Use (BCSU) manufactured by Particle Measuring Systems is an inertial microbial impactor with elliptic cuts (slits) used to perform microbiological analysis and cleanroom air monitoring, under active air flow. It is made of transparent polystyrene, with visible culture media. A vacuum source connects to the side adapter to allow a constant flow of air into the device. After sampling and incubation, the lid can be removed for additional colony testing. The BCSU offers diverse media formulations available in ready-to-use (RTU) culture media in packaging sterilized by irradiation.

The BCSU was recently tested to verify the effectiveness of its microbial contamination collection and media dehydration under conditions of continuous three-hour sampling. The microbial collection efficacy of the BCSU was found to be maintained after 3 hours with a level of dehydration not found to impact careful use or growth results. With the backing of this study, BCSU can be used for three-hour continuous sampling with no change to its monitoring effectiveness or capabilities.

This product is an appropriate choice for monitoring environments that have been disinfected, and where contamination by beta-lactam antibiotics may be present, especially where beta-lactam antibiotics are produced.

Learn more by reading the full study here https://www.pmeasuring.com/wp-content/uploads/2022/02/303-Study-of-Single-Use-Device-Efficiency-1.pdf

EECO2 Expanding energy efficiency to cleanroom design & build services

Translating expertise in efficient AHU design to the energy intensive environment of a cleanroom poses a unique opportunity to reduce operational expenditure, while also decreasing the environmental impact of a facility. Indeed, the energy consumption of cleanrooms is often so high due to the high airflow rates required for specific ISO classifications, with HVAC systems accounting for up to 50-75% of electricity usage. With this in mind, and as energy intensive practices come under increasing scrutiny due to public and private pressure to improve long-term environmental sustainability, EECO2 are now offering full cleanroom design & build services to support the transition to more sustainable facilities in the pharmaceutical and high-tech industries.

For more information please e-mail info@eeco2.com or visit www.eeco2.com

Life-lines

War quotes

War does not determine who is right – only who is left.
(Bertrand Russell)

Older men declare war. But it is the youth that must fight and die.
(Herbert Hoover)

The object of war is not to die for your country but to make the other bastard die for his.
(George S Patton)

Mankind must put an end to war before war puts an end to mankind.
(John F Kennedy)

I know not with what weapons World War III will be fought, but World War IV will be fought with sticks and stones.
(Albert Einstein)

If we don't end war, war will end us.
(H G Wells)

I am tired and sick of war. Its glory is all moonshine. It is only those who have neither fired a shot nor heard the shrieks and groans of the wounded who cry aloud for blood, for vengeance, for desolation. War is hell.
(William Tecumseh Sherman)

Know thy self, know thy enemy. A thousand battles, a thousand victories.
(Sun Tzu)

When the rich wage war, it's the poor who die.
(Jean-Paul Sartre)

To be prepared for war is one of the most effective means of preserving peace.
(George Washington)

Events

2022	Event	Location
April 4-8	Achema	Frankfurt, Germany
April 26-27	Making Pharmaceuticals. Exhibition, Conference and Awards	Coventry, UK
May 2-5	IEST ESTECH 2022	Virtual
May 9-11	r3nordic Symposium & Exhibition 2022	Naantali, Finland
June 14	PHSS Sterile Medicinal and ATMP product manufacturing Conference 2022	Knutsford, Cheshire, UK
October 11-13	25th International Symposium on Contamination Control, ICCCS'20	Antalya, Turkey
October 27	A3P Aseptic Technologies (Isolators & RABS) Forum	Presencial, Spain
November 14-17	IEST EDUCON	Schaumburg, Illinois
November 23-24	Cleanzone	Frankfurt, Germany

Training courses

IEST (Institute of Environmental Sciences and Technology) www.iest.org		
2022	**Event**	**Location**
May 2	The Foundations of Contamination Control using Essential Cleanroom Standards ISO 14644-1 and ISO 14644-2	Virtual
May 3	Basic Information and Implementation of the New ISO 14644-3:2019 Test Methods	Virtual
May 4	Universal Cleanroom Operations Guidelines with ISO 14644-5	Virtual
May 5	Basics of Cleanroom Design	Virtual
For a complete list of courses, please see https://www.iest.org/Training-Certs/IEST-Contamination-Control-Learning-Path		

CCN (Contamination Control Network) www.theccnetwork.org		
2022	**Event**	**Location**
April 12-14	CTCB-I Cleanroom Testing Course	Fully booked
November 8-10	CTCB-I Cleanroom Testing Course	Letchworth, UK

Other training courses including CTCB/I* training courses are provided by:		
BCW	Belgium	www.bcw.be/
ICS	Ireland	www.cleanrooms-ireland.ie/training/
R3Nordic	Nordic Countries	www.r3nordic.org/
VCCN	Netherlands	www.vccn.nl/cursusaanbod
TTD	Turkey	www.temizoda.org.tr/en/trainings

*CTCB-I Certification: Cleanroom Testing and Certification Board International Certification, see CTCB-1 website: www.ctcb-i.net/index.php

Assistant editor required for CACR to help with and gradually take over the commissioning and editing of articles. Would suit a retired contamination control expert or someone active in the field with time and energy for a small 'job on the side'. Good understanding of the subject required and an ability to write clear English. Contact jneiger@johnwrite.co.uk

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