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Material Transfer Into Aseptic Areas: Hierarchy Of Contamination Control

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INTRODUCTION

The presence of viable organisms and in particular bacterial spores in the Grade A environment presents a significant risk of contamination in aseptically prepared products and potential patient harm (1). In terms of risk terminology, the likelihood of product contamination is low, but the consequences are severe. This places a considerable emphasis upon transfer disinfection / decontamination (2). The method that was commonly used for this process was a standard transfer hatch (an interlocking device with no air supply) where items going in were (ideally) multi-wrapped and subject to manual disinfection (sometimes no more than ‘spray and pray,’ a slightly better process being the application of a sporicidal agent and wiping the surfaces of each item). This process has been improved by fitting localized air supplies (Grade A / ISO class 5 at rest), which is the minimum EU GMP Annex 1 requirement (as indicated in the 2020 draft)(3), and with the wider availability of decontamination chambers (which use bio-decontamination agents like hydrogen peroxide) (4). A commonality with each of these approaches is the application of a sporicide, with in aqueous or vapor form. The Technical Committee (CEN/TC 216 “Chemical disinfectants and antiseptics”) of the European Committee for Standardization has defined a sporicidal as a product which kills dormant bacterial spores of relevant test organisms under defined conditions.
However, despite some similarities there is a contamination control hierarchy with the different methods of transferring items into an aseptic area. If autoclaving is placed at the top (since it is a sterilization rather than a disinfection method), then it follows*in order of increasing contamination risk*:

1. Autoclaving / depyrogenaton tunnels
2. Automated cycle decontamination chambers.
3. ‘Dynamic’ pass-through hatches with HEPA filtered air supply.
4. ‘Static’ pass through chambers with no air supply.
5. Personnel transferring items (e.g., via changing rooms).

This paper assesses the transfer disinfection process, the available technologies and critical bio-decontamination aspects, focusing on the important criteria for device assessment and operation.

HIERARCHY OF CONTAMINATION CONTROL MEASURES

The most effective way to transfer items into an aseptic processing area is by autoclaving (or, in the case of glass bottles and vials, depyrogenation). Not only is this a sterilization method (using a penetrative agent – moist heat), but the process is also in the hands of the pharmaceutical or healthcare organization. This is followed by the use of sterile, disposable, multi-wrapped items (with sterilization typically by gamma radiation or ethylene oxide gas) which can be ‘pass through’ using a decontamination chamber, where a sporicidal disinfectant or

'decontamination agent' (such as hydrogen peroxide) is used (triple-wrapped is the general standard for multi-wrapped items) (5). Due to the inability to penetrate items, concerns around reproducibility of the vapor distribution, and with some bacterial spore enzymes being relatively insensitive to peroxide damage (such as F1ATPase and pyruvate kinase and organisms like*Bacillus megaterium*) (6), the term 'decontaminant' rather than 'sterilant' is reserved for hydrogen peroxide vapor (and at times bio-decontamination is used to indicate that chemical contaminants are not inactivated).

Next in order is the dynamic pass-through hatch equipped with HEPA filtered air (which requires a manual disinfection step, with the disinfectant being a sporicide). Lastly is the static transfer hatch without an air supply (a fourth option of personnel simply carrying items in via a changing room or transfer room will not be covered in this review). While all pass-through activities present the risk of passive particle transfer, the automated decontamination chamber presents the best chance of inactivating those particles that are microbial in nature compared with dynamic and static hatches. With the dynamic hatch, the contamination risk will be relatively lower than with the static hatch due to the ventilation of the pass-through box interior. This dynamic application of HEPA filtered air reduces the particle count within the hatch as well as the probability that particles will escape or enter the hatch. The static hatch will have some degree of control through a pressure differential, provided that cleaner air enters. However, with no active flushing the system is far weaker. Furthermore, such a process is wholly reliant upon cleaning and disinfection by operators; this is something that can theoretically be demonstrated by the use of adenosine triphosphate (ATP) bioluminescence and it often, in this author's experience, delivers variable results and this method is generally of a poor sensitivity. This can relate to disinfectants being applied using inadequate contact times; the variations with different surface types and topography; a failure to use an adequate number of wipes; the use of wipes without sufficient antimicrobial activity; poor technique that can lead to spread of contamination from one surface to another (such as via unorganized wipe patterns); a loss of disinfectant activity due to the binding of the active from a disinfectant to cloths of unsuitable material substantial amounts of cellulose may reduce the antimicrobial efficacy of the disinfectant, among other factors (7-11).

Hence, we can present a hierarchy of control measures for entering a Grade B / ISO class 7 (in operation) area:

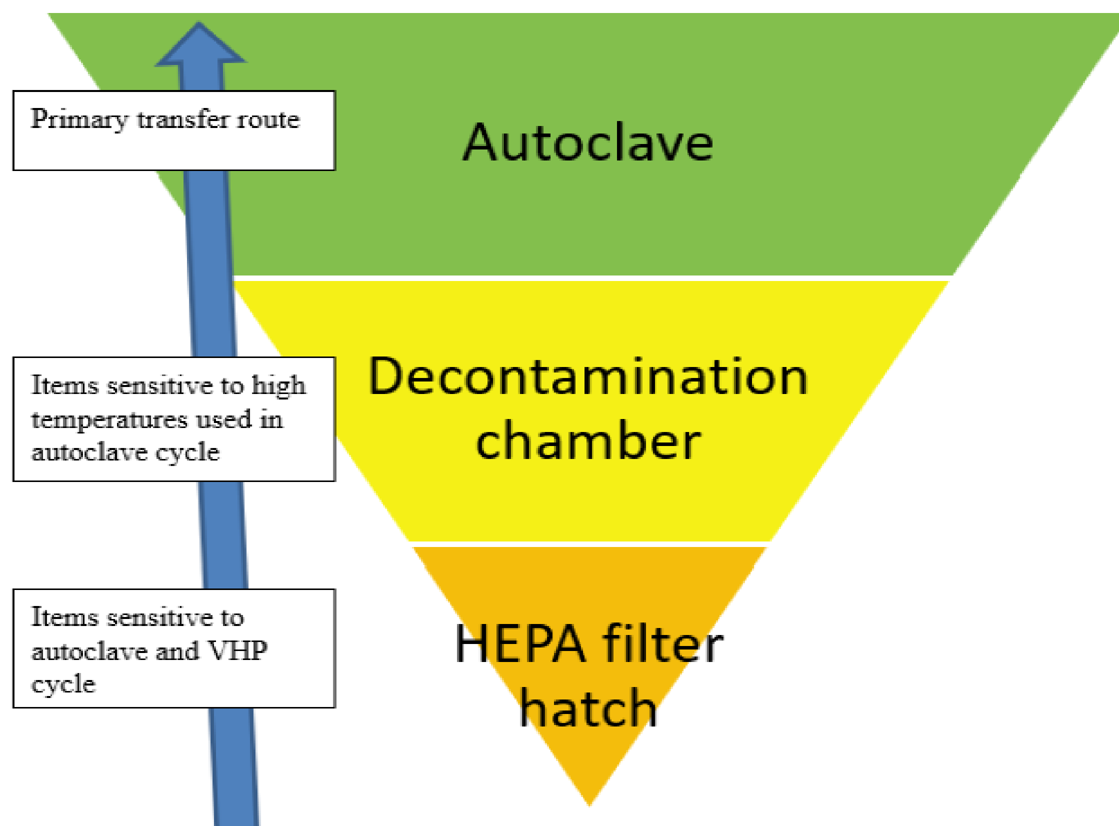

Figure 1: Hierarchy of control measures for material entry

OBJECTIVES OF MATERIAL TRANSFER BETWEEN CLEANROOM GRADES

The transfer of materials between different cleanroom grades is most critical for transfer between Grade C (ISO class 8 in operation) and Grade B (ISO class 7 in operation) (12).

Figure 2: The pass-through cascade.

As indicated above, this can be by:

1. Autoclaving / depyrogenaton tunnels
2. Automated cycle decontamination chambers.
3. 'Dynamic' pass-through hatches with HEPA filtered air supply.
4. 'Static' pass through chambers with no air supply.
5. Personnel transferring items (e.g., via changing rooms).

With the above, '1' is outside the scope of this paper, and '5' are a process of such variability and risk that careful consideration is required should this method of transfer be attempted. With '2', '3', and '4', the process involves: the hatch or chamber has interlocks, so that only one side door can be opened at a time, preventing depressurization of the cleanroom. Transferring materials into the cleanroom begins when an operator opens the pass-through door on the "dirty air" side; the interlock mechanism automatically or manually locks the cleanroom-side door shut. The operator places materials into the device on the "dirty air" side and closes and latches the door. The device will have latching doors on both sides that tightly compress against closed-cell or non-absorbent gaskets to prevent air loss from the cleanroom. This is important, for if the air pressure in the cleanroom drops, untreated dirty air at lower pressure will flow into the cleanroom. With '2'an automated bio-contamination cycle is run; with '3' the disinfected items are flushed with clean air; with '4' there is no additional decontamination. Once any decontamination cycles have been run, the interlocking mechanism releases and an operator opens the cleaner side door and transfers materials into the cleanroom. After transfer is complete, the cleanroom side door is closed. The application of a heavy-duty sealing lock ensures contaminants are kept out when the chamber is not being used. Furthermore, the use of interlocking doors prevents both doors from being opened at the same time. Ideally these should be time controlled so that the correct amount of disinfection and air flushing is achieved. There are two types of interlock: mechanical or electrical. Mechanical interlocks come in two types: manual or automatic. For manual mechanical interlocks, these must be engaged by the operator, severely reducing transfer efficiency, therefore automatic types are preferrable. With electrical interlocks these provide chambers with automatic, hands-free doors or systems equipped with access controls, occupancy sensors, building management integration or event logs. Remote data logging can provide information relating to the user, time, duration, and product(s) scanned for entry or removal (13).

The operating principle is that 'pass-through' is one way, for clean materials to enter the area. Waste should have a separate route out, and away from an areas used for aseptic processing. Each of the '2' to '4' options involve the use of a sporicidal agent (sporistatic agents or other disinfectants are not suitable). The accepted sporicidal agents for direct surface contact are chlorine, hydrogen peroxide, hypochlorous acid, and a combination of hydrogen peroxide and peracetic acid. For automated decontamination, where a vapor is generated, the options are typically conventional hydrogen peroxide vapor, ionized hydrogen peroxide, chlorine dioxide, and ozone (the latter two being, at the time of writing, emergent technologies) (14). Sporicides should be evaluated for their efficacy. For transfer disinfection, in relation to the maximum allowable numbers of microorganisms on a cleanroom surface, the minimum target is >log 2 or 3 kill within a practicable time (such as 5-10 minutes). This criterion is in the context of the bioburden on consumables and within the cleanroom being relatively low (typically less than 100 CFU per item in relation to a given unit of measurement, such as 25 cm2)

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Further similarities include the materials of construction. These include cold rolled close annealed (CRCA) powder coated, stainless steel grade 304 or 316 L (the primary material of construction should be 316 L since 316 grade stainless steel contains more nickel and molybdenum than 304 grade stainless steel, enhancing its resistance to corrosion in wet environments, and degradation from bleach-based disinfectants). The surface roughness value should be Ra ?0.8 µm. The hatches and chambers should have a coved internal work surface for ease of decontamination and pre-disinfection cleaning. Such radius corners do away with hard-to-clean

crevices or lips. In addition, stainless steel that is electropolished removes metal impurities from the surface that can damage metal. The use of gaskets will minimize air exchange between spaces and prevent air leakage.
Most devices will have glass view panels. Larger devices will often be fitted with conveyors, to enable heavier materials to be loaded and then passed through at the appropriate juncture. It is typical for devices to be fitted with audible and visual alarms, to either indicate a malfunction (such as pressure loss or the leaking of hydrogen peroxide, where applicable) or that the material is ready to be transferred through. The use load presence sensors can also be fitted so a video camera feed is activated, enabling a specific load to be assessed as part of a deviation or during an audit.
An alternative option, for fluid transfer, is wall pass-through technology for transferring liquids, such as the filtration of product or cleaning solutions. This technology is formed of single-use disposable fluid transfer and isolation barrier components and it conveys fluids without the need to physically move bins and totes, thereby removing the risk of breakage, spillage and lost pharmaceutical product associated with bin transfer.

OBJECTIVES AND OPERATION OF DECONTAMINATION CHAMBERS

The objectives of an automated cycle decontamination chamber are to decontaminate both the chamber and the load. Some forms of contamination can be removed through the cleaning process, such as by proteins, carbohydrates and lipids. However, this would require a separate qualification and the scope of this paper is with microorganisms: viruses, bacteria, fungi, and bacterial spores (15). Within the hospital sector, these are sometimes referred to ss 'no touch' devices to indicate the absence of personnel interaction. Such systems work as outer surface decontamination devices and are designed to achieve a 6-log reduction in spore forming bacteria (and vegetative cells) (16), fungi (17), and viruses (18). The ability of the device to achieve a sufficient level of biological kill is assessed using *Geobacillus stearothermophilus* biological indicators. Alternatively,*Bacillus subtilis* , Clostridium sporogenes, and*Bacillus cereus* have been used as challenge microbes. The important factor is that the selected organism is resistant to hydrogen peroxide vapor.
Chambers come in a range of sizes, the most common are complete 'pass throughs,' where large items of equipment can be wheeled in (these are sometimes referred to as zone-to-zone or cart-to-cart devices). Smaller chambers are of a wall-mounted type. Important design features include ensuing that the chamber and the door system are manufactured to specific hourly leak rates per specific applications. Decontamination chambers will have an air filtration system that uses HEPA filters. Such systems are re-circulatory in and designed to ensure EU GMP Grade A / ISO 14644 class 5 cleanliness levels with unidirectional airflow
Important performance criteria include:

- HEPA integrity testing (six-monthly)
- Velocity measurement (automated, with alarms)
- Air-change rates
- Non-viable particle monitoring (six-monthly)
- Light intensity
- Sound levels
- The air flow pattern under simulated working conditions

Hydrogen peroxide cycles and other forms of bio-decontamination

The hydrogen peroxide and water vapor is typically generated by flash evaporation on a heated plate, which is in excess of the boiling points of the components of the solution. This ensures that the ratio of hydrogen peroxide and water are the same in the vapor phase as in the liquid that is evaporated (19). Vapor is a gas at a temperature lower than its critical point.
Hydrogen peroxide decontamination is usually performed in four phases (20):

1. Stabilizing the equipment temperature and where necessary to adjust the relative humidity inside the closed chamber to a predetermined level.
2. Flash evaporation of the hydrogen peroxide solution into a heated air stream and pass the vapors into the chamber to raise the gas concentration to the required level for decontamination.
3. Liquid continues to be evaporated into the air stream, often at a lower rate in order to maintain the required gas concentration inside the chamber. This is sometimes referred to as the dwell phase; in essence it means a sufficient time period form the vapor to contact the surfaces in order to achieve microbial kill. Kill is assessed by using biological indicators in qualification runs. Cycles are then normally increased to increase the achieved log reduction of bacterial spores to a theoretical higher log value (much like the Sterility Assurance Level of an autoclave).
4. The passing of clean air into the chamber to remove the hydrogen peroxide vapor by dilution.

Of these steps, the control of temperature and humidity are every important; while the humidity range can be relatively wide, the efficacy of hydrogen peroxide vapor is very temperature sensitive (21).
The effectiveness of hydrogen peroxide vapor depends both on the overall concentration and the even distribution of the hydrogen peroxide inside the chamber. In order to vaporize the hydrogen peroxide before dispersing it into the working space, a controlled amount of liquid peroxide is sprayed onto a heated plate

above which an air stream is flowing. During the decontamination process it is important to continuously measure the hydrogen peroxide concentration inside the working space during all the phases of the treatment. If the difference between actual and set concentration can be continuously measured, this will determine the flow rate of the liquid hydrogen peroxide released onto the evaporation plate.

It is also important to ensure good vapor distribution, which can be evidenced from chemical and biological indicators during load development. Hydrogen peroxide vapor, as indicated above, will not kill microorganisms protected by a covering or shielded through other occluded surfaces. The opportunities for the vapor not to reach all surface should be minimized and it is important that procedures specify item positioning or loading according to the qualification. Otherwise, this will make the decontamination process unreliable and non-repeatable. Hanging items within the chamber rather than laying them flat aids a more effective distribution of the vapor and hence greater vapor-to-surface contact.

A further factor of importance is material compatibility. In general, many materials are compatible. For example, a review of a range of laboratory equipment, for use in a biocontainment level III facility, showed that all items of equipment were compatible with the decontamination process (22). While materials are often compatible, many can absorb the vapor leading to extended aeration times. For instance, hydrogen peroxide can penetrate polyethylene materials that provide the primary packaging for many presterilized items. This problem increases the larger the dimension becomes. In addition, certain polymeric materials will adsorb peroxide rapidly and desorb it very slowly. Whereas materials like glass, elastomeric materials, aluminum, and stainless steel are generally impervious to peroxide (23).

Alternatives to conventional hydrogen peroxide vapor ionized hydrogen peroxide, which does not require a conditioning phase, or chlorine dioxide gas. Ionized hydrogen peroxide systems produce a mist by aerosolizing a solution of 7.5% solution. The hydrogen peroxide particles are electrically charged, range in diameter from 8 to 12 ?m, and can circulate freely in the air as an aerosol decontaminant with access to all material surfaces (24).

OBJECTIVES AND OPERATION OF PASS-THROUGH HATCHES

For multi-wrapped, sterile disposable items, an alternative method for passing through items between cleanrooms of different grades is a conventional pass-through hatch. These come equipped with and without HEPA filtered air (to comply with the 2020 draft EU GMP Annex1 (3), a HEPA filtered air supply is required). The objective with both types of hatch is, after a manual disinfection step, that items placed inside the chamber are exposed to a ventilation system that provides filtered air to ensure a clean environment inside the chamber, before the opening of the door.

The so-called 'static' transfer hatch is best suited for cleanrooms of the same grade and for use outside of aseptic areas. The so-called 'dynamic' transfer hatch is suitable for passing items into aseptic areas, and it essentially operates as a mini airlock (25).

Transfer disinfection and contamination control

The most important aspect of the use of pass-through hatches is transfer disinfection. Contamination will arise from the external environment, the internal environment, surface cross-contamination (such as from cart surfaces), and via personnel, specially via fingerprints from gloved hands. Ensuing appropriate levels of education and training of staff can help to reduce the finger dab contamination levels.

With the transfer disinfection step, the essential elements here are with the incorporation of a 2-stage transfer disinfection process, ensuring that the required contact time for the disinfectant is achieved:

- Application of sporicide by spraying and then wiping (or use of a pre-saturated wipe).
- Application of an alcohol-based disinfectant by spraying and then wiping (or use of a pre-saturated wipe). This serves to address any contamination that may have been spread around and increases the drying time from the first application (26).

The use of pre-sterilized multi-wrapped procedure kits whereby successive layers of wrapping can be removed as items are transition from outside, through controlled-not-classified areas, and through the cleanroom cascade. The removal of outer packaging at the earliest stage possible represents an important consideration (it is good practice that items taken from a general storage area into the first stage cleanroom should be wiped to remove dust and bioburden using an alcohol dampened wipe (27). An aqueous-based sporicidal wipe is generally not considered appropriate at this stage as this may damage the integrity of the packaging at an early point in the process.) When specifying single-use disposable items suppliers, the items below should be considered (many of these considerations are applicable for automated decontamination as well, in reference to the section above).

Table 1: **Specifications for the presentation of single-wrapped, sterile disposable technologies**

Factor	Detail	Method of assessment
Item assembly: cleanliness	Assembly of items should be in an EU Grade C / ISO class 8 (in operation) cleanroom or at a higher grade.	Audit

Factor	Detail	Method of assessment
Terminal sterilization	The method of sterilization is either: gamma radiation or by ethylene oxide (EtO). If EtO sterilization is used it is important to assess how penetration into inaccessible parts to sterilize is assessed and with the desorption of the gas. With gamma irradiation, this is typically >25kGy.The minimum and maximum should be justified. If items can be reprocessed (especially sterilization) by the supplier, supporting data must be provided. The desirability or non-desirability of this must be stated in the technical agreement. If the receiving organization are to sterilize the items, the material impact should be assessed with supporting supplier data.	Audit Technical agreement Validation certification identity check on delivery.
Sterility Assurance Level	Details concerning the Sterility Assurance Level (SAL) assessment must be provided, and the minimum acceptable SAL is 10-6 (overkill is desirable).	Specification and audit (sterility to be verified as identity check on delivery) Technical agreement.
Wrapping and packaging	The design of the wraps should have no folds or webs which will make them difficult to decontaminate. There should be the minimum creases on the surfaces to facilitate even and effective wiping.	Audit and specification
	The packaging materials must be robust (for example, 100um PAPE, Polyclear or similar material).	Specification
	The permeability of the overwraps to moisture and contamination should be provided by the manufacturer.	Supplier review
	The seal of the bag must have been assessed, such as the dye intrusion test (or equivalent) to show packaging integrity. Key parameters are sealing pressure, temperature, and time. The tolerance should be agreed between the vendor and the purchaser. The method of sealing should be assessed (e.g., initial sealing of 3 sides is common to position the materials in the bag followed by a final seal). Overall packages should be pressure tested using pressure decay and drop tests. This these tests should be a realistic worse case specific to the product presentation	Audit (including evidence reproducibility) Technical agreement for s tolerance.
	All items are contained within a box, and within a dust cover.	Specification and identity che on receipt
	Within the dust cover, all items are triple wrapped.	Specification Technical agreement
	Items should have hole or suitable opening to permit hanging for VHP. [LP12]	Specification and supp acceptability
Opening wrappings	The wrapping should be easy tear or peel opening (with tear indent or start point). The contained components should be strategically orientated to facilitate clean removal from packaging.	Supplier assessment a specification.

Factor	Detail	Method of assessment
Physicochemical properties	Items should be assessed for low binding properties, adsorption, and leaching (when applicable),	Supplier data, with R assessment as part of supp acceptance. Verification in audit.
Particulates	Items and packaging must have low particle loading.	Supplier data, review as part audit.
Identity check	All items must be labelled with: • Batch Number and expiry date • A statement 'for single use only' • Statement of sterility	Technical agreement Identity check for release.

For the disinfection transfer step, simply spraying without wiping is not recommended, and disinfectant efficacy studies have shown wiping to be more effective in terms of ensuring complete contact (28). In addition, aqueous based disinfectants are prone to droplet coalescence on certain surfaces compared with the spreading and wettability of an alcoholic medium. Furthermore, health and safety issues often restrict the use of sprays unless local exhaust ventilation is available.

Hence, with the method of disinfectant application, the use of pre-saturated wipes (with a sporicidal agent like 6% hydrogen peroxide) arguably demonstrates a greater reduction in bioburden than dry wipes which are wetted*in situ* (29). For this reason, such wipes are often preferred for aseptic disinfection transfer. This is because dry wipes are sometimes not wetted enough to readily release sufficient alcohol onto the surface. In addition, the undulating and micro-structures of surfaces being disinfected does not always facilitate the effective delivery of disinfectant by the wipe process. Wipes additionally entrap particles and absorb residues more readily. Care needs to be taken with the selection of the wipe, in terms of particulate generation. Effective wipes function to:

1. To physically remove the bioburden from the surface
2. To ensure the presence of sufficient disinfectant for long enough to kill vegetative and where possible, spore forming microorganisms
3. To facilitate the destruction and removal of contaminants by the application of pressure against microbial cell walls during the wiping process.

A fresh surface should be exposed for each wipe for each wiping stroke. This is beset achieved by folding a wipe twice and using the four-fold wipe technique.

To assess the effectiveness of the transfer disinfection method, the bioburden of materials and within the chamber should be assessed periodically or with each operation (the latter being a recommendation for transfer into aseptic areas).

UV-C light options

Both types of hatches are sometimes equipped with ultra-violet light, which enables the surfaces of the hatch to be disinfected when not in use. Of the different forms of UV light, UV-C is the most effective (200-280 nm). UV light inactivates microbial cells by damaging their DNA or RNA through photo-chemical reactions. Here, the light initiates a reaction between two molecules of thymine. Thymine is one of the bases that forms DNA. Cells will attempt to repair of form of DNA damage, by excising or removing the two bases and filling in the gaps with new nucleotides (30). If the cellular damage is extensive, then this mechanism breaks down. Hence, the longer cells are exposed to UV light for, then the more thymine dimers are formed in the DNA and the greater the risk of an incorrect repair and the cell ceases to be able to perform its normal functions. Therefore, UV kills cells because of the accumulation of DNA damage.

The most common methods for producing UV to achieve surface disinfection are:

- **Low-pressure mercury lamp:** This has its main (>90%) emission at 254 nm.
- **Excimer lamp or Far-UVC lamp:** Type of lamp, called an “excimer lamp,” with a peak emission of around 222 nm.
- **Pulsed xenon lamps:** These lamps, which emit a short pulse of broad spectrum (including UV, visible and infrared) light have been filtered to emit mainly UVC radiation.
- **Light-emitting diodes (LEDs):** Light-emitting diodes (LEDs) that produce UV radiation emit a very narrow wavelength band of radiation, such as peak wavelengths at 265 nm, 273 nm, and 280 nm.

An alternative to UV-C is high-intensity narrow-spectrum (405 nm) light, which targets intracellular porphyrins that absorb the light and produce reactive oxygen species. However, the antimicrobial efficacy is lower than UV-C light (31).
Important factors affecting the efficacy of UV-C are:

- UVC radiation can only inactivate microorganisms if the cells are directly exposed to the radiation. Therefore, the inactivation of microorganisms on surfaces may not be effective due to blocking of the UV radiation by soil, such as dust, or other.
- The dose must be sufficient and the lower the dose, the longer the exposure time required for a given surface area.

UV rarely achieves ‘sterilization’, and it should be classed as a method of disinfection, due to the potential for survivor cells. It is also possible to underestimate the numbers of bacterial survivors due to damaged cells requiring longer incubation times, when assessing recovery in validation experiments (32). Of the different forms of UV processes, pulsed UV, which has a rapid energy delivery rate, appears to be more effective than continuous UV (33). UV irradiation and hydrogen peroxide can act synergistically to kill bacteria, both vegetative cells and spores (34), but the use of peroxide does not fit with the conventional transfer hatch model.
UV light is not always practically suitable for the transfer of items since the contact times required may be too long for this process to be operationally feasible in most cases. Even for ‘at rest’ periods, many facilities operating 24/7 do not have long periods of time when the hatch is not in use. The minimum acceptable irradiance in a biosafety cabinet is regarded as 40 W/cm^2, according to the U.S. Department of Health and Human Services (35). Therefore, it takes 12.5 minutes to reach 30,000 J/cm^2 (1 W= 1 J/sec), which is taken to be germicidal for spore forming organisms (36). The application of UV light in excess of an hour or overnight presents massive overkill. However, this assumes the UV can reach the microorganisms. If there is either a film or shadowing object, the organisms in the shadowed (protected) areas will not be hit with sufficient UV energy. The use of reflective material is an option, although aluminum is the only material that has a high reflectivity for ultraviolet rays in the wavelength range of 250 nm to 400 nm and most hatches will be fashioned from stainless steel. It is also common for the bulb/lamp to become fouled and occluded by dust or other deposits, reducing its emissions unless there is regular cleaning of the UV source.

Operation of dynamic pass-through hatches

To ensure that items are flushed with HEPA filtered air for a sufficient period of time (this can be based on the calculated air-changes per hour from an understanding of hatch volume and air velocity), the transfer hatch system should keep the double-ended doors blocked so that sufficient air passes can take place (a typical time range is 3 to 5 minutes). With air velocity, this is typically within the range 0.36 to 0.54 m/s (drawing upon the target ranges for unidirectional airflow described in FDA and EU regulations) at working height, although other airflow velocities can be justified. In terms of particulate monitoring, the typically practice is to assess particulate levels at rest and to periodically demonstrate EU GMP Grade A / ISO 14644 class 5 air supply (37).
It is also important, as with any system interfacing two cleanroom areas of different grades (such as a conventional airlock) that both does cannot be opened at the same time. This principle is especially important to facilitate the removal of the material from the door connected to the cleanroom of the highest class, with the aim to not contaminate the clean area during the ventilation cycle.

CONCLUSION

The revised EU GMP Annex 1 and its requirement for a contamination control strategy includes a focal point around the transfer of materials, with an emphasis upon robust disinfection practices where the use of double-ended autoclaves is not possible (36). While there have been many advances with single-use, sterile, disposable technology, a risk remains with external contamination and the type of contamination inevitably includes bacterial endospores and fungal spores. This paper has assessed three methods of transfer: automated decontamination chambers, chambers which require a manual disinfection step, but which are equipped with HEPA filtered air supply, and static air boxes. This order presents an increasing cascade of risk, in terms of microbial contamination remaining (and where poor technique is in place, of spreading contamination further). Nevertheless, it is recognized that different facilities will have different levels of technology and hence the three methods were discussed and the important factors in relation to the qualification and operation of each presented.
Nevertheless, qualified, automated systems will always confer an advantage. This is because the use of operator dependent wipe-spray transfer disinfection techniques is widely recognized as one of the weakest links in the aseptic process. Many objects are too bulky, uneven with their surface topography, or there are simply too many items for an operator to process consistently. However, automated decontamination systems are not without their weaknesses and they require reproducible qualification. The main concern stemming from bio-validation data is with ensuring that the decontamination agent is capable to circulate and contact all affected surfaces, as evidenced by chemical indicators (distribution) and biological indicators (sporicidal kill). Where there are occluded surfaces, a risk assessment is required (such as pre- and post-manual sporicidal application). Across the different measures of material transfer, the concept of removing successive layers of wrapping remains important, as a further contamination mitigation step.

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