SAFE USE OF MICROBIOLOGICAL SAFETY CABINETS

Microbiological Safety Cabinets are designed to protect users and the environment (which includes other people in the laboratory) from aerosol risks arising from the handling of hazardous biological material. Some types of cabinet are also designed to protect the materials being handled within them from environmental contamination. Air discharged from the exhaust of the cabinet is filtered to remove microbial contamination and is either ducted to outside or recirculated into the laboratory.

Microbiological safety cabinets are not designed to protect the user from all hazards, e.g. radioactive, toxic or corrosive hazards, and the exhaust HEPA filters will not remove these types of contaminants from the exhaust air. Particular care must be taken when using materials with such additional hazards to ensure these are not discharged into the laboratory environment from cabinets that are not externally ducted.

This document describes the main factors that should be taken into account when selecting the appropriate safety cabinet for your intended use; where it should be positioned within the laboratory and venting arrangements. As externally vented microbiological safety cabinets are deemed to be “Local Exhaust Ventilation” (LEV) equipment for controlling exposure to hazardous substances, there is a statutory requirement under the COSHH Regulations for regular maintenance, examination and testing to be carried out at least every 14 months.

The relevant British Standards covering microbiological safety cabinets are:
- BS EN 12469 Biotechnology - Performance criteria for microbiological safety cabinets 2000 (this standard supersedes BS 5726 Microbiological Safety Cabinets 1992, Parts 1 & 3), and
- BS 5726 Microbiological safety cabinets - Information to be supplied by the purchaser to the vendor and to the installer, and siting and use of cabinets - Recommendations and guidance 2005 (this standard supersedes BS 5726 Microbiological Safety Cabinets 1992, Parts 2 & 4),

The purchase, installation, commissioning, servicing and maintenance of microbiological safety cabinets should always comply with the British Standard specifications. In relation to Safety Cabinets, the terms “user”, “worker” and “operator” are synonymous. Materials handled within a Safety Cabinet are commonly described as the “work” or, less obviously, as the “product”.

1. Types of Cabinet

There are three types or “Class” of Microbiological Safety Cabinet which differ significantly in design and mode of operation. These are referred to as Class I, Class II and Class III cabinets. All provide protection to the user (operator protection), with Class II and Class III cabinets also providing a clean working environment to protect the work from contamination (termed product protection).

The British Standard defines the three types of cabinet as follows. A diagrammatic representation of the airflow patterns in the different Classes is provided in Appendix 1.

| Class I | Safety cabinet with a front aperture through which the operator can carry out manipulations inside the cabinet and which is constructed so that the worker is protected and the escape of airborne particulate contamination generated within the cabinet is controlled by means of an inward airflow through the working front aperture and filtration of the exhaust air. |
Class II Safety cabinet with a front aperture through which the operator can carry out manipulations inside the cabinet and which is constructed so that the worker is protected, the risk of product and cross contamination is low and the escape of airborne particulate contamination generated within the cabinet is controlled by means of an appropriate filtered internal airflow and filtration of the exhaust air.

*Note:* A typical way of achieving this is by means of a uni-directional downward (laminar) airflow inside the cabinet and an air-curtain at the front aperture.

Class III Safety cabinet in which the working area is totally enclosed and the operator is separated from the work by a physical barrier (i.e. gloves mechanically attached to the cabinet). Filtered air is continuously supplied to the cabinet and the exhaust air is treated to prevent release of micro-organisms.

In both Class I and II microbiological safety cabinets the inward airflow protects the user by minimising the escape of any airborne particulate contamination generated within the cabinet. In Class II microbiological safety cabinets the “downflow” of filtered air affords protection to the work minimising contamination during manipulations. In Class III cabinets the physical barrier protects the user from the work and the air going in to the cabinet is filtered to protect the work.

Some manufactures also produce a hybrid Class I/III cabinet but this is not described within the British Standard. The hybrid, as the name suggests, can be used as either a Class I or Class III cabinet by the use of a removable port that attaches to the front aperture. However the construction and testing of these cabinets is such that when used in Class III mode it is not equivalent to the specification of a standard Class III cabinet.

### 2. Cabinet selection for particular applications

A risk assessment should be undertaken to determine the Class of cabinet appropriate for a particular work activity. This should take into account the nature of the potential hazards in terms of not only the micro-organisms involved and their route of infection but also the techniques to be carried out and whether protection of the work (product protection) is needed.

The Class of cabinet required is not linked to the Containment Level assigned to the work. It is a commonly made mistake to think these are connected and it can lead to inappropriate selection of cabinet. The following sets out some general guidance on selecting a cabinet.

Modern Class II microbiological safety cabinets designed to meet the current British Standard give a high degree of protection to the user. When Class II cabinets were first introduced some years ago, they did not give nearly such a high level of protection as the Class I cabinets. Their use was therefore limited and it was not acceptable to use a Class II cabinet for Hazard Group 3 pathogens. Nowadays, with the improved performance, this type of cabinet is suitable for most pathogens apart from those in Hazard Group 4. Additional guidance on the selection of cabinets for work at Containment Level 3 is provided in Appendix 2.

New Class II cabinets will probably be the cabinets of choice for most applications in the University as these provide both operator (user) and product protection (protection of the work) and so allow for flexibility in future use when the nature of the research work may change. However where an older Class II cabinet is already in situ, care should be taken to
ensure its performance is adequate for purpose.

Class I cabinets should be used if procedures within the cabinet are likely to generate a significant aerosol and/or disrupt the air flow pattern within a Class II cabinet and so compromise operator (user) protection. An example would be use of a homogeniser to break up tissues. A Class I cabinet would be preferentially selected over a Class II for work with certain pathogens that infect via the airborne route (for example Neisseria meningitidis) if there is no need for protection of the work (product protection).

Since a Class III cabinet is totally enclosed this offers the highest level of protection to both the user and the work. However, in practice this level of protection tends only to be required for the most hazardous work i.e. for certain Hazard Group 3 and Hazard Group 4 pathogens in Containment Level 3 or 4 facilities.

Some general comments on selections likely at the different containment levels:

I. At Containment Level I a cabinet is unlikely to be required for operator (user) protection as any micro-organisms involved are unlikely to cause harm (otherwise the work would be assigned to a higher containment level). Class II cabinets can be used to provide protection of the work (product protection), for example for tissue culture work; a Class I cabinet will not provide product protection. Alternatively a vertical laminar flow cabinet could be used to provide product protection.

II. At Containment Level 2 usually a Class II cabinet would be used to provide both operator (user) and product protection (protection of the work) unless the procedures are likely to generate a significant aerosol or compromise air flow pattern in which cases a Class I cabinet should be used. If a respiratory pathogen is being used then consideration should be given to using a Class I cabinet.

III. At Containment Level 3 select cabinet according to nature of work, see additional guidance in Appendix 2.

Where operator (user) protection is required for work with hazardous micro-organisms the cabinet should meet the requirements of the current British Standard relating to Microbiological Safety Cabinets (BS EN 12469). Some manufacturers sell tissue culture cabinets. These are very similar to Class II microbiological safety cabinets but do not meet the full specification of the British Standard and so are less expensive. Whilst these do offer operator protection to the user, since they do not meet the recognised standard the use of tissue culture cabinets is not recommended where operator protection is required for work with hazardous micro-organisms.

There are other types of cabinets or hoods available but these do not provide operator protection, they are designed to protect the work only. Examples of such types of cabinet include laminar flow cabinets. These types of cabinet must not be used if operator (user) protection against micro-organisms is required. Laminar flow cabinets may use either horizontal or vertical laminar flow. Since horizontal laminar flow cabinets blow air from the back of the cabinet across the work and into the face of the user it is entirely inappropriate to use this type for work handling anything other than clean, non-hazardous materials. Fume cupboards and hoods must not be used to provide operator (user) protection against airborne biological hazards. If there is any doubt as to the suitability of a particular cabinet for use then the University Biological Safety Adviser should be contacted for advice.

4. Venting arrangements

It is good practice to discharge the exhaust air from Microbiological Safety Cabinets to the outside. Within the University the following approaches should be taken:
I. In Containment Level 2 facilities, venting via a HEPA filter through a window or wall to the same level is acceptable, provided attention is paid to the position of nearby opening windows or adjacent air intakes. Depending on the building, planning restrictions may prohibit this as an option.

II. If it is not possible to vent to the outside, a recirculating cabinet fitted with double HEPA filters on the exhaust may be considered in Containment Level 2 facilities (see note below) providing there are no other hazardous contaminants in the discharged air. Consideration must be given to a safe method of fumigating the cabinet.

III. In Containment Level 3 facilities cabinets must exhaust via a HEPA filter to the outside. The output duct must be taken to roof level or alternatively it may exhaust at the same level, providing a double HEPA filter is fitted. If for some reason ducting to atmosphere is not practicable then the University Safety Services must be contacted for further advice.

Note:- in the old British Standard (BS 5726) it was specified that if air is recycled back into the laboratory that it be through two HEPA filters whereas BS EN 12469 specifies only a single HEPA in the exhaust with the caveat that risk assessment may demand additional requirements. Within the University it is recommended that all recirculating Class II cabinets be installed with double HEPA on the exhaust to ensure they are suitable for work with all types of micro-organisms. This will need to be specified when the cabinet is ordered.

5. Siting, installation and commissioning

The siting of a microbiological safety cabinet is extremely important. Air currents and movement of people in the laboratory can adversely affect the performance (operator protection) of a cabinet. Factors to be considered include the proximity of cabinets to doors, windows, ventilation ducts and to movement routes - see Appendix 3. Positioning of cabinets within laboratories should meet the guidelines set out in the current British Standard (BS5726:2005), some of which are reproduced here in Appendix 3. For new cabinets the supplier should always visit the site, undertake a site survey and advise on installation and meeting BS5726:2005 prior to contracts being placed. If the proposed sitting does not meet the recommendations set out in BS5726:2005 and there is no suitable alternative then the University Safety Services should be contacted for advice.

Cabinets must be properly installed and commissioned. Prior to use the cabinet must pass the performance tests specified in BS EN 12469. The test requirements are quite detailed, need specialist equipment and competent persons to undertake the work properly. This therefore forms part of the service offered by the supplier. Schools should note however that similar requirements apply when cabinets are moved or relocated and so a specialist contractor will need to be appointed to undertake such works.

If a School chooses to install a cabinet itself then the requirements of BS EN 12469 must be met. A specialist contractor must be appointed to undertake the operator protection (KI Discus) test prior to use. It is strongly recommended that Schools ask the contractor to comment on the installation at that time, specifically as to whether it meets the requirements of BS EN 12469. Particular points to note are in relation to siting, incorporation of anti-blow back valves and the need for additional fans if ducting is longer than two metres or bent in any way.

6. Routine Maintenance, Examination and Testing

In order to meet the British Standard specification, cabinets undergo various testing when manufactured. Within the British Standard there are also requirements for tests on
installation and regularly thereafter to demonstrate performance under conditions of use.

Most importantly, microbiological safety cabinets constitute local exhaust ventilation (LEV) systems in that they offer protection to the worker (user) from airborne hazards. As such there is a requirement for regular maintenance, examination and test under the COSHH Regulations. Therefore, all microbiological safety cabinets should be serviced on an annual basis and undergo examination and test at that time. It is a requirement of the COSHH Regulations that a record be kept for 5 years of the examinations and tests and of repairs. Health and Safety Executive Inspectors are likely to request sight of, or copies of, records during visits to the University.

The certificate should show tests results for:

1. Volumetric airflow measurements and airflow patterns

These include various measurements of face velocity (inward airflow) at the front aperture and, in Class IT cabinets, the velocity of the laminar downflow.

For Class I cabinets the measured face velocity should be between 0.7 m/s and 1.0 m/s at all points. For Class II cabinets this should be not less than 0.4 m/s.

The downflow in a Class II cabinets (not applicable in a Class I) should be between 0.25 m/s and 0.5 m/s.

2. Exhaust HEPA filter test

The HEPA filters on the exhaust are there to ensure that any contamination in the airstream is filtered prior to discharge. It is therefore important to check the integrity of the filters to ensure there are no holes and the filter is properly located so there are no leaks around the edges. The test is undertaken by introducing an aerosol challenge to the airstream upstream of the filter and testing to see if there is any penetration downstream.

Filters should have an efficiency of at least 99.995% (or penetration of <0.005%).

3. Operator Protection Factor (or KI Discus) Test

As part of the inspection, a containment test for operator (user) protection should be undertaken. This is usually by the KI Discus method where an aerosol of potassium iodide is generated within the operating cabinet and sampling devices are placed in front of the cabinet to capture any aerosol escaping from the working area. The operator protection factor (OPF) is defined as the ratio of exposure to airborne contamination generated on the open bench to the exposure resulting from the same disposal of airborne contamination generated within the cabinet.

When tested in accordance with the British Standard all cabinets in use should have an operator protection factor of at least $1.0 \times 10^5$.

Within the University the following approaches should be taken:

1. All cabinets must have an operator protection (KI Discus) test included as part of the commissioning process for new or relocated cabinets;
2. All cabinets must be tested for operator protection (KI Discus test) on an annual basis, or every six months if in Containment Level 3 facilities;
3. Operator protection tests to be carried out in such a way as to ensure the cabinet and
the laboratory are as representative as possible of normal working conditions, i.e:

a. with any air conditioning units or other ventilation systems in the laboratory switched on;

b. with other safety cabinets and fume cupboards within the laboratory switched on;

c. with the cabinet loaded with a typical arrangement of equipment and samples;

d. with a person moving around the laboratory, particularly if any pedestrian traffic is near the cabinet; and

e. with doors (laboratory, nearby incubators and fridges etc) being opened and closed.

Copies of KI Discus test certificates must be kept for at least 5 years (a requirement under the COSHH Regulations).

7. Training and correct use of cabinets

The effectiveness of the microbiological safety cabinet depends on:

- good design
- suitable installation;
- ongoing maintenance; and
- correct use.

Comments on the first three items in this list have been covered in earlier sections. It is important users of microbiological safety cabinet are trained in correct use not only in order to understand how the cabinet works but also because poor technique can compromise the operator protection afforded by the cabinet.

Training in the use of Microbiological Safety Cabinets will be provided by Technical and academic staff as required and supplemented with practical training provided by local personnel dealing with the specifics of the particular equipment, location, work, etc.

Training should be provided to cover:

- principles of how the different classes of cabinets work including airflow patterns;
- suitability of different cabinets for particular types of work;
- principles of airflow, operator protection factor and filter penetration tests;
- limitations of cabinet performance;
- how to work at cabinets safely;
- operation and function of all controls and indicators;
- how to decontaminate the cabinet after use (routine cleaning); and requirements for fumigation and, where appropriate, how to do this.

The Health and Safety Executive has produced a video entitled Microbiological safety cabinets: Safe working practices which is available to buy from HSE Books ISBN 07176 2099 9 (£29.38). It is recommended that all users of safety cabinets within the University view this video (19 minutes running time) as part of their training.

Incorrect use of microbiological safety cabinets can compromise their performance and adversely affect the level of operator protection afforded by the cabinet. Some of the most common factors that users should pay attention to are:

- the user should avoid sudden and sweeping movement of their arms to minimise disturbance of the air flow patterns;
- large and bulky equipment should not be placed in Microbiological Safety Cabinets, nor should equipment be placed on air grilles as both these will disturb air flow
patterns;
- centrifuges, including microfuges, should not be placed in a Microbiological Safety Cabinets unless an operator protection factor (KI Discus) test has been carried out with it running in situ and has been proven not to compromise operator protection;
- bunsen burners should not be used in Microbiological Safety Cabinets, particularly Class II, because of the concern about the effect of the heat rising from the flame on the laminar downflow of air in the cabinet. However, if they are used, they should be placed towards the back of the cabinet and a low profile type used. If the bunsen is used in conjunction with alcohol etc for flaming, then the alcohol pot should always be placed to the far side of the bunsen in order that any drips from the item being flamed do not drop in the pot and ignite it; and
- Microbiological Safety Cabinets should always be installed in appropriate locations to ensure any traffic movement within the laboratory does not cause draughts to disturb the airflow patterns at the front of the cabinet (see Appendix 3) and affect performance. Users should be aware of this requirement and should ensure the 1 metre clear behind rule is observed when they are using the cabinet.

A checklist of “Dos” and “Don'ts” for users when working at Microbiological Safety Cabinets is provided in Appendix 4.

8. Fumigation of cabinets

Fumigation must be carried out only by a trained responsible person with adequate knowledge of the procedure and the precautions to be followed.

Fumigation with formaldehyde vapour is the recognised and most commonly used method for this type of fumigation procedures although an alternative systems (e.g. using vapourised hydrogen peroxide) are available.

Formalin is a commercially available 40% solution of formaldehyde vapour in water. When formalin is heated formaldehyde vapour is generated in quantity. Formaldehyde is a Schedule 1 chemical under the COSHH Regulations and has a Workplace Exposure Limit (WEL) of 2 ppm (or 2.5 mg/m³). Concentrations encountered during fumigation are many hundreds of times higher than this so fumigation operations must be carried out only by trained personnel under a “Safe System of Work”. All workers using formaldehyde must be aware of safe handling procedures.

Under certain conditions formaldehyde can react with hydrochloric acid and chlorine-containing disinfectants, e.g. hypochlorites, to form bis (chlormethyl) ether, a potent lung carcinogen. Hydrochloric Acid and chlorine-containing disinfectants must therefore be removed from Microbiological Safety Cabinets before fumigation.

Formaldehyde vapour is an extremely effective biocidal agent. It acts as an alkylating agent, inactivating micro-organisms by reacting with carboxyl, amino, hydroxyl and sulphydral groups of proteins as well as amino groups of nucleic acid bases. A number of factors affect the efficiency of fumigation. For formaldehyde to act to maximum effect it must be able to penetrate (pre-cleaning is helpful if it can be done without jeopardising safety) and it must be able to dissolve at adequate concentrations in a film of moisture in the immediate vicinity of the organisms to be inactivated. Water vapour generated in the process of dispersing formaldehyde provides the essential optimum level of relative humidity and so it is important to ensure that water is added to the formalin prior to vapourisation. Too much formaldehyde results in the deposition of sticky deposits of paraformaldehyde and in cabinets may
contribute to filter blockage. The amounts of formalin and water required for fumigation are given below.

Fumigation is most effective above 20°C and relative humidity of 65%. Below 18°C formaldehyde fumigation is less effective. Below 9°C, formaldehyde sublimes and is less easy to vapourise.

There are a number of methods of generating formaldehyde vapour:
1. heating a mixture of formalin and water in a purpose-made vaporising unit;
2. using commercially available formaldehyde generating kits; or
3. mixing formalin and water with potassium permanganate crystals*.

*WARNING: the correct relative concentration of these two components is essential to avoid a violent reaction. It is therefore recommended that this method is NOT used.

If Microbiological Safety Cabinets have been used for work with hazardous micro-organisms, they must be fumigated in the following circumstances:
1. after a major spillage or a spillage where inaccessible surfaces have been contaminated;
2. before any maintenance work on the cabinet where access to potentially contaminated parts is necessary (including filter and pre-filter changes);
3. before carrying out filter penetration tests; and
4. when there are any changes in the nature of the work that result in significantly different risks.

Agents causing the Transmissible Spongiform Encephalopathies are resistant to inactivation by formalin and alternative decontamination procedures are required. Further advice is available from the University Biological Safety Adviser.

Where the cabinet has been used for hazardous micro-organisms, HEPA filters should be handled only with appropriate protective clothing (laboratory coat and heavy duty gloves) even after fumigation. After fumigation, filters must be disposed of as hazardous waste.

Microbiological Safety Cabinets must be sealed before fumigation to prevent leakage of formaldehyde into the laboratory. It should be checked to ensure the closure panel/night door has been properly and securely located and a good seal has been achieved. Where necessary, sealing tape should be used to ensure there is no leakage. With Class III or hybrid (Class I/Class III) cabinets a blanking plate should be fitted over the inlet filter.

If the cabinet is a recirculation type, there must be a safe means of venting the formaldehyde vapour safely outside the building, such as by the use of a fumigation adaptor kit (see below), or by absorption onto Activated Carbon filters.

The fumigation procedure should ensure inactivation of any micro-organisms that have penetrated the HEPA filter by adequate exposure of the downstream side of the HEPA filter and the ductwork to formaldehyde. In the absence of specific recommendations by the cabinet manufacturer the cabinet fans should be turned on for 10 - 15 seconds after about half the formalin has been evaporated and again after evaporation is complete. Passive migration of the fumigant through the filter can occur but this is not ideal. Some Microbiological Safety Cabinets have automatic fumigation cycles programmed into the controls and in these instances the manufacturers' instructions should be closely followed.

Formaldehyde should be left to disperse within the cabinet for at least six hours (or preferably overnight) after which time the fumigant should be exhausted to atmosphere by
switching on the fan and allowing air from the room to enter the cabinet. Before venting the formaldehyde in this way it is essential to ensure that no-one is in the vicinity of the exhaust outlet and that the exhaust air does not enter nearby windows or ventilation air intakes.

Following decontamination, the Microbiological Safety Cabinet must be purged of all residual formaldehyde. With recirculation type cabinets a fumigation adaptor kit should be fixed over the cabinet exhaust, prior to fumigation, to allow discharge of fumigant via the flexible trunking into either a ducted cabinet, fume cupboard, a fumigation port or out of a window. Care must be taken to ensure no formaldehyde is discharged back into the laboratory. Under no circumstances should recirculating type cabinets be fumigated unless there is a safe means for discharging the formaldehyde to the outside atmosphere in such a way that no person can be affected by the gas discharge.

Formaldehyde must not be recirculated into the laboratory, this would constitute an uncontrolled release of a hazardous chemical and be a reportable incident to the Health and Safety Executive. In the event of investigating such an incident, or if during a visit, Health and Safety Executive Inspectors became aware that such an unsafe practice was in use, it is very likely that enforcement action would be taken and a successful prosecution be brought against the University.

Typical quantities of formalin required for fumigation of the different types of cabinets are:

<table>
<thead>
<tr>
<th>Cabinet Type</th>
<th>Amount of BP Formalin</th>
<th>Amount of Water</th>
</tr>
</thead>
<tbody>
<tr>
<td>Class II (1800mm wide)</td>
<td>30 ml</td>
<td>30 ml</td>
</tr>
<tr>
<td>Class II (1200mm wide)</td>
<td>25 ml</td>
<td>25 ml</td>
</tr>
<tr>
<td>Class II (900mm wide)</td>
<td>20 ml</td>
<td>20 ml</td>
</tr>
<tr>
<td>Class I</td>
<td>20 ml</td>
<td>20 ml</td>
</tr>
<tr>
<td>Class III</td>
<td>20 ml</td>
<td>20 ml</td>
</tr>
<tr>
<td>Class III Hybrid</td>
<td>20 ml</td>
<td>20 ml</td>
</tr>
</tbody>
</table>

Whilst fumigation is in progress a large notice must be posted on the front of the cabinet to warn that the cabinet is being fumigated. An example of a suitable sign for posting on the cabinet when fumigation is taking place is provided in Appendix 5.

Manufacturers of Microbiological Safety Cabinets should provide detailed instructions for fumigation of their particular cabinets and these should be followed. This is particularly important when the cabinet has an automatic fumigation cycle. An outline of the main principles of fumigation has been provided above and manufacturers' instructions should be consistent with these (if there are serious discrepancies please contact the your BSO or University Safety Services for advice).

Within Schools and Departments there must be a written procedure in place for cabinet fumigations. A summary of the main steps is provided in Appendix 5. This summary is a model only and should be modified as appropriate to tailor it to the particular cabinets in a laboratory. The written procedure must identify those individuals competent and authorised to carry out the fumigation process.
Appendix 1

MICROBIOLOGICAL SAFETY CABINETS - Summary of Types

Operations where there is a risk of airborne infection must be performed in microbiological safety cabinets or under equivalent containment. Three types of cabinet are specified by the current British Standard:

Class I
An open fronted cabinet through which air is drawn at a sufficient rate to minimise aerosol escape. The air is filtered by a high efficiency particulate absorption (HEPA) filter and is discharged to the exterior. The worker but not the work is protected. Suitable for work with Hazard Group 2 pathogens and most Hazard Group 3 pathogens.

Class II
An open fronted cabinet where the working space is flushed with a downflow of sterile air which is HEPA filtered and recirculated. Some air is drawn in through the front of the cabinet and a corresponding amount discharged to the outside through a HEPA filter. Both work and worker are protected. Suitable for Hazard Group 2 pathogens and, in some circumstances, for Hazard Group 3 pathogens.

Class III
A totally enclosed cabinet where the operator is separated from the work by gloves attached to ports and the incoming and outgoing air is HEPA filtered. Gives a high degree of protection to work and worker. Suitable for Hazard Group 3 and Hazard Group 4 pathogens.
Appendix 2

SELECTION OF MICROBIOLOGICAL SAFETY CABINETS FOR CONTAINMENT LEVEL 3 FACILITIES

Current guidance from ACDP is that Class I and Class II cabinets are suitable for work with all Categories of biological agent, except those in Hazard Group 4 and the use of Class III cabinets is usually confined to work with biological agents in Hazard Group 4. This is a broad generalisation and as always is subject to risk assessment of the particular work activity. Therefore, a risk assessment must be undertaken to determine the type of cabinet that should be used for the different types of work that may be undertaken within a Containment Level 3 laboratory. The following provides additional guidance for users in the University

a) A Class I or Class III MSC must be used:
   1. where there is an airborne route of transmission for the Hazard Group 3 agent (e.g. *Mycobacterium tuberculosis*); or
   2. if a procedure is used which generates a significant aerosol (e.g. homogenisation).

   If in the above cases protection of the work is essential then either a Class III cabinet should be used or other engineering controls (e.g. HEPA filtered room supply air) introduced to clean up the room air prior to it entering the front of the cabinet.

b) A Class III MSC must be used:
   1. where certain strains of dangerous pathogens are handled (e.g. multiple-drug resistant); or
   2. where there is an airborne route of transmission for the Hazard Group 3 agent (e.g. *Mycobacterium tuberculosis*) and a procedure is used which generates a significant aerosol.

c) A Class II MSC may be used for work with:
   1. agents in Hazard Group 3 with derogation such that use of a MSC is not essential (e.g. parasites);
   2. non-Hazard Group 3 pathogens that are assigned to Containment Level 3 under DEFRA restrictions (subject to DEFRA licence conditions);
   3. materials known or suspected of containing Hazard Group 3 agents but there is no cultivation or concentration of those agents (e.g. bloods from endemic regions, liver samples from hepatitis patients);
   4. other pathogens in Hazard Group 3 which are unlikely to be infectious via the airborne route (including the blood borne viruses such as hepatitis, HIV etc);

   - providing the cabinet is tested for operator protection (KI Discus) every 6 months; all KI discus testing must be in accordance with the British Standard (4 head test) and no individual value of the protection factor shall be less than 1.0 x 10^5; and

   in the case of the blood borne viruses (4 above), if the work involves handling high titres of infectious virus the operator protection test must be shown to be satisfactory under the conditions of use. The in-use test must be carried out with a person moving around in the laboratory and opening and closing the laboratory door simulating normal working practices. All ventilation and other cabinets in the room must be on for the duration of the test.

*Note:* If risk assessment determines a Class III cabinet is required for a particular activity, the use of a hybrid Class III in Class III mode is not acceptable since the specification for these types of cabinets is not as high as that for a Class III cabinet built to the British Standard criteria.
**CORRECT SITING OF MICROBIOLOGICAL SAFETY CABINETS**

Poor siting of a unit can adversely affect its performance. Specialist engineers should advise on correct positioning of cabinet at installation. Cabinets shouldn’t be sited on thoroughfares or in line with doorways or opening windows. Disturbances in airflow e.g. caused by air conditioning units, should be taken into account when siting a cabinet. The following requirements should be taken into account:

<table>
<thead>
<tr>
<th>Pedestrian traffic routes should be at least 1000 mm from the front of the microbiological safety cabinet.</th>
<th>A microbiological safety cabinet should not be positioned with either side closer than 300 mm from a wall or similar obstruction e.g. structural columns</th>
</tr>
</thead>
<tbody>
<tr>
<td>Doorways should not be situated within 1500mm of the front of a microbiological safety cabinet or within 1000 mm of the side of the cabinet. The only exception is when a door includes air transfer gills. Testing should be carried out to ascertain a suitable distance. Opposing benches should not be situated within 1500mm from the front of a working cabinet.</td>
<td>There should be no opposing wall within 2000mm of the front of a microbiological safety cabinet</td>
</tr>
<tr>
<td>A bench at right angles to a microbiological safety cabinet may keep traffic away from the front of the cabinet, but any other person working at that bench will cause disturbances in airflow.</td>
<td>A projecting bench will help to keep traffic clear of the microbiological safety cabinet and work at the bench will have little effect on airflow if sited at a minimum of 1000mm from the side of the cabinet.</td>
</tr>
<tr>
<td>The distance from the front of an opposing microbiological safety cabinet or fume cupboard should be at least 3000mm.</td>
<td></td>
</tr>
</tbody>
</table>

*Source: the above layouts are based on the recommendations for avoiding disturbances given in BS5726: 2005.*
Appendix 4

THE DO'S AND DON'T'S WHEN WORKING AT A MICROBIOLOGICAL SAFETY CABINET

DO'S
• Make sure the particular cabinet is suitable for your work (risk assessment)
• Organise and plan your work in advance
• Keep the inside of the cabinet free of clutter
• Always wear a lab coat
• Check the indicators! dials show if it's safe to use
• Sit comfortably at the cabinet centre
• Use good aseptic techniques
• Allow to purge before switching off
• Always clean up after use

DON'TS
• Do not obstruct the air intake grilles in Class II cabinets
• Do not use bunsen burners or centrifuges in Class II cabinets
• Do not use until the cabinet has warmed up
• Do not work with the UV light on
• Do not let others in the lab intrude in your space - keep 1 metre clear behind
• Do not put any paperwork in the cabinet
• Do not use if in any doubt about cabinet performance
• Do not rely on the cabinet to cover up poor technique

REMEMBER
• A cabinet only offers protection against infectious aerosol hazards
• The level of protection offered by a cabinet relies heavily on good working practices
Appendix 5

SUMMARY OF MAIN STEPS FOR FUMIGATING MICROBIOLOGICAL SAFETY CABINET

Manufacturers of Microbiological Safety Cabinets should provide detailed instructions for fumigation of their particular cabinets and these should be followed. A written procedure must be in place for cabinet fumigations. The following summary is a model only and is provided as a typical example of what to include. It should be modified as appropriate to tailor it to the particular cabinets in a laboratory.

Fumigation must be carried out only by a trained responsible person with adequate knowledge of the procedure and the precautions to be followed. The written procedure must identify those individuals competent and authorised to carry out the fumigation process.

1. Switch off the cabinet fans.
2. If the cabinet is a recirculation type, fit the fumigation adaptor kit to the discharge and position the other end to vent to atmosphere. Close the manual shut-off damper.
3. Fill the vaporiser with the correct amount of formalin and water and screw on the aluminium cap - finger tight, having checked the gasket in the cap is undamaged. Place the vaporiser in the cabinet.
4. Fit the closure panel/night door and fully seal the front screen and closure panel with sealing tape to ensure there are no leaks.
5. Post a notice on the front of the cabinet indicating fumigation is in progress.
6. Switch the vaporiser on.
7. After approximately 10 minutes switch the cabinet fans on for 10 - 15 seconds.
8. After a further 20 - 30 minutes switch the cabinet fans on again for 10 - 15 seconds.
9. Leave the cabinet in this condition for a minimum of 6 hours, preferably overnight.
10. If the cabinet is a recirculation type check the exhaust of the fumigation adaptor kit is in a position to discharge safely and open the manual shut-off damper.
11. Before venting the formaldehyde check no-one is in the vicinity of the exhaust outlet and that gas will not enter any open windows nearby.
12. Exhaust the formaldehyde from the cabinet by switching on the fans and opening the closure panel/night door slightly (remove bung if fitted or crack open) until the majority of the formaldehyde has been exhausted. After about 10 minutes the night door may be removed completely.
13. Any poly-formaldehyde residue in the vaporiser may be removed by heating with water containing a little mild detergent.
14. Run the cabinet for at least a further 15-20 minutes to remove the last traces of formaldehyde.
15. If the cabinet is a recirculation type the fumigation adaptor kit must be removed before the cabinet is used again.
WARNING

DO NOT ENTER

- FORMALDEHYDE

FUMIGATION IN PROGRESS