PC2 Laboratory Safety Manual

Laboratory Name:

Chief Investigator:

Department:

Version 1.0

19/02/2014 12:04:00 PM

Signature: _____

Date of next review:

Table of Contents

Introduction	3
Description	3
Nature of Work	3
The Laboratory	3
Contacts	3
Access	4
Training	4
Risk Management	
Hazard Identification	6
Pathogen Risk Groups	6
Register of Micro-organisms	6
Risk Assessment	
Assignment of Risk Group	
Safety Information about Microorganisms	
Risk Control	
Engineering Controls	
Biological Safety Cabinet	
Fume Cupboards	
Pipettes	
Administrative Controls	
General Laboratory Rules	
Personal Hygiene	
Hand Washing Technique	
Working Alone	
Work Procedures	
Decontamination	
Waste Disposal	
Spills	
Personal Protective Equipment	
Vaccinations	
Clothing & Footwear Requirements	
Laboratory Gowns	
Eye Protection	
Gloves	
Instructions for the safe removal of contaminated gloves	
Risk Management	
References	
Appendix A Laboratory Arrangement	
Appendix B Induction Checklist	
Appendix C Cleaner/Maintenance Personnel Induction Checklist	
Appendix D Safety Training Plan and Assessment (Draft)	
Appendix E Register of Microorganisms	
Appendix H Record of Vaccination Form	

Introduction

is a Physical Containment level-2 (PC2) facility, certified by the University's Institutional Biosafety Committee (IBC) for handling:

- pathogens of Risk Group 2
- low-risk dealings with genetically modified organisms
- human cell cultures and body fluids.
- This manual is provided to document safe operating practices required in order to achieve level 2 containment, and is the basis for training all laboratory personnel who wish to use the facility.

This manual refers to a number of additional sources of information, including:

- An MSDS folder which provides safety information about agents in use.
- An Equipment folder which contains operating manuals, procedures and test and maintenance records.
- An Induction folder which is a record of induction and training undertaken by personnel using the laboratory.
- A Risk Management folder which contains risk assessments for procedures undertaken

These folders are located at

This manual will be reviewed regularly, and at least annually, by and the Hazard Management Officer to ensure that it remains up-to-date.

Description

Nature of Work

is used for the following types of work:

The Laboratory

Appendix A provides an illustration of the laboratory configuration, equipment arrangements and location of emergency equipment. The following description should be read in conjunction with Appendix A.

Contacts

Further information about the operation of the laboratory can be obtained from the following people:

Name

Role

Extension

Hazard Management Officer 4921 6846

Access

Routine access is only provided to persons that have:

- undertaken the General Laboratory Safety Induction
- undertaken the PC2 Laboratory Induction (Appendix B)
- been issued with the required personal protective equipment
- have been authorised as a laboratory user by

The laboratory shall be kept locked at all times when not in use. Only authorised persons shall be provided with key for the laboratory.

Where access is required for the purposes of maintenance or cleaning, this will only be provided to persons that have:

- undertaken the Cleaner/Maintenance Induction (Appendix C)
- demonstrated that they have any required personal protective equipment
- been authorised by

Names of authorised persons will be maintained by the Laboratory Supervisor, and a record of inductions kept in the Induction folder.

Working safely is a condition of access to the PC2 facility. Repeated failure to observe safe working practices and procedures will result in the withdrawal of access privileges.

Training

During their time in the laboratory, users will be required to develop and ultimately demonstrate competence in the range of practices required to work safely in the laboratory. Appendix D provides a list of these competencies.

Timing of the training program and assessment will depend on the nature of the work undertaken, the current level of experience of the person and their need to work unsupervised. These records should also be kept in the Induction folder.

Risk Management

Risk Management is the process of recognising situations that have the potential to cause harm to people or property, and doing something to prevent this from occurring. The risk management process consists of well-defined steps that lead to informed decisions about controlling the impact of risks. These steps are:

- 1. Hazard identification
- 2. Risk Assessment
- 3. Risk Control
- 4. Evaluation & Review

This manual covers each of these steps with regard to risks specific to Risk Group 2 microorganisms.

Hazard Identification

Pathogen Risk Groups

Microorganisms falling into Risk Group 2 present moderate individual risk, and limited community risk. They cause human, animal or plant disease but do not pose a serious risk because effective treatment and preventative measures are available and there is limited potential for spread.

For example, *Staphylococcus aureus* rarely causes life-threatening disease in a laboratory situation, and is a Risk Group 2 microorganism. *Human immunodeficiency virus*, although potentially lethal, is also a Risk Group 2 microorganism (when not concentrated) because in normal laboratory circumstances the risk of transmission is low [1].

Register of Micro-organisms

Appendix E provides a register of the microorganisms used in this laboratory, including their nominal risk group, origin and storage location. Health Canada MSDSs for infectious organisms website: www.hc-sc.gc.ca/pphb-dgspsp/msds

Risk Assessment

Assignment of Risk Group

Microorganisms are classified according to their level of risk. This classification is based on factors such as:

- pathogenicity the resulting disease incidence and severity;
- the route of transmission aerosol, ingestion or parenteral;
- the target hosts including issues such as the required infectious dose, their immune status, etc.;
- the concentration of organisms in the media being handled;
- agent stability its ability to survive over time or under standard disinfection regimes; and,
- the availability of effective prevention and treatment measures.

There are a number of sources of categorisation information [1-4]. The primary reference is AS/NZS 2243.3 Safety in the laboratory Part 3 - Microbiology [1].

Note that Risk Groups, hence containment requirements, do not just depend on the microorganism, but may also be a function of the type of operations undertaken with it. For example, diagnostic blood samples can be handled under Level 2 containment conditions, but concentrated cultures of HIV must be handled under Level 3 conditions [1].

As a result, selection of risk control measures should be based on an assessment of the tasks to be undertaken and information about the pathogen(s) involved.

Safety Information about Microorganisms

Safety data sheets for the microorganisms are available in the MSDS folder.

In addition, there are a number of on-line sources of safety information, including:

ChemWatch	http://locust.newcastle.edu.au/chem (internal access only)
Canadian Office of Biosafety	http://www.hc-sc.gc.ca/pphb-dgspsp/msds-ftss/index.html
CDC	http://www.cdc.gov/od/ohs/biosfty/bmbl4/bmbl4toc.htm (see Section VII - Agent Summary Statements)
HSE	http://www.hse.gov.uk/hthdir/noframes/agents.htm

Risk Control

The hierarchy of controls should be applied to the management of risks stemming from microbiological hazards, namely:

Elimination - does the work with the microorganism need to be done? *Substitution* - can a microorganism in a lower Risk Group, or of lower overall risk, be used to do the work? *Modify* the work system or process - e.g. use lower concentrations or smaller quantities. *Isolation* - can the microorganism be completely contained during the work? *Engineering* controls - e.g. use a biological safety cabinet *Administrative* controls - safe work procedures and practices - e.g. laboratory rules like no mouth pipetting

Personal Protective Equipment is used - e.g. laboratory gowns, gloves.

Physical Containment of microorganisms represents a combination of engineering and administrative controls, coupled with the use of personal protective equipment. However, <u>elimination</u> and <u>substitution</u> should always be considered prior to starting work with a given pathogen.

Primary barriers to transmission of infectious agents are provided by enclosures such as biosafety cabinets and personal protective equipment. The laboratory facilities (or work area) provide a secondary barrier, which acts to contain the agent in the event of a failure of the primary barriers. The combination of primary and secondary barriers and work practices constitute the level of containment that must be used to keep the risk of exposure of laboratory workers and the outside environment to acceptable levels.

An important point that should not be overlooked is that the work practices are integral to the maintenance of these barriers. Laboratory equipment and design contribute to safety only if they are used properly by people who are trained and, where necessary, supervised. It is critical that all laboratory users are trained in the correct use of personal protective equipment, in the monitoring of the facility and its equipment, and in the practices that should be followed while working in the laboratory.

Engineering Controls

Biological Safety Cabinet

Where there is the potential of generating aerosols containing infectious microorganisms the primary barrier of choice is the Class II Biological Safety Cabinet (BSC).

There are several standards that apply to the design and use of these cabinets in Australia [5]. Copies of the BSC manuals for the laboratory can be found at in the Equipment folder.

BSCs should not be confused with other laminar flow benches or fume extractions systems. Class II BSCs are open-fronted, ventilated containment enclosures intended for work with Risk Group 2, 3 and 4 organisms that can be deactivated by a formaldehyde fumigation procedure. They are self-contained work-stations and operate independently of other air-handling systems. The cabinets incorporate High Efficiency Particulate Arresting (HEPA) filters, which are the physical containment barrier that trap sub-micron particles such as microorganisms.

In Class II BSCs an inflow of room air into a full-width grille at the base of the work opening creates an air barrier. A quantity of air, equal to that of the barrier air, (normally about 30% of the total airflow) is exhausted to the room via a HEPA filter. The rest is separately HEPA filtered, and recirculated within the work zone via vertical, downward laminar airflow, to provide product protection.

Class II cabinets therefore provide personnel, environment and product protection. However, because of the exhaust into the room, it is important to note that **the BSC is not suitable for handling materials containing volatile toxic or radioactive chemicals.**

UV lamps

The BSC is fitted with germicidal ultraviolet (UV) lamps in the work zone. UV can be a useful adjunct to surface cleaning procedures, but should not be seen as a replacement for good cleaning technique.

- UV lamps should be used for 20 to 30 minutes at the beginning and end of work. They should not be left on for extended periods.
- Personnel exposed to UV radiation may suffer eye damage and erythema (sunburn). Work opening covers should be in place whenever UV lamps are in use.
- UV radiation degrades nitrile, plastics and rubber products and organic coatings.
- UV is ineffective in dynamic air streams, on dried organic matter, and is not penetrating. Radiation intensity reduces over time due to degradation of the lamps. Where the use of UV is a significant element of surface decontamination procedure, regular testing of lamp intensity should be conducted. This can be arranged by

<u>Use</u>

Points that should be noted about BSC operation include the following:

- Before materials are introduced to a BSC they should be (externally) decontaminated
- Keep 'clean' and 'dirty' materials separated inside the cabinet.
- Minimise rapid air movement near the cabinet opening, such as that caused by people walking past, to maintain the laminar air flow. Do not use Bunsen burners in Class II cabinets use disposable loops or electric heating instead.
- Don't use centrifuges inside a BSC.
- Keep the front intake grilles clear.

- Keep the exhaust discharge clear over at least 60 cm in order to allow free air flow and access for maintenance. Do not store items on top of top-exhaust cabinets.
- Before starting work at the BSC, adjust the laboratory stool to ensure that your:
 - forearms remain parallel to the work surface most of the time
 - lower back is supported
 - head is upright
 - feet can reach the footrests.

Operating the BSC

- 1. Remove the front metal cover and set aside.
- 2. Start the BSC blower and ensure the BSC lights are operating.
- 3. Gently open the front perspex shield using both hands.
- 4. Spray the inside of the cabinet with disinfectant and wipe down. Place required equipment in the BSC and spray any unsterilised surfaces with disinfectant.
- 5. Lower the perspex shield and replace the metal cover.
- 6. Turn the BSC blower and lights off, and switch the UV lamps on.
- 7. After 30 mins the hood is ready for use. Ensure the UV lamps are off, remove the metal cover and start the BSC blower and lights.
- 8. Always wear gloves and avoid rapid or unnecessary arm movements while working in the BSC.
- 9. When finished, spray all equipment with disinfectant before removing it from the BSC. Then spray the interior of the BSC with disinfectant and wipe down.
- 10. Replace the metal cover. Shut off the BSC blower and lights, and turn on the UV lamps for 30 minutes.

NOTES:

- Keep the front grill clear at all times.
- Check that the BSC is 'in test' before use.

Testing & Maintenance

BSCs require inspection and testing of airflow and filter performance at least annually, as well as after modification (including filter changes) or relocation. Cabinets should also be tested if there is reason to suspect they are not operating correctly.

Testing and maintenance of biological safety cabinets is the responsibility of . Users should ensure that the BSC has a current test sticker on it prior to use. Records of testing are maintained in the Equipment folder. Because maintenance may require access to the 'dirty' side of the system, decontamination using formaldehyde is usually carried out as part of the testing program.

Fume Cupboards

A fume cupboard is essentially a ventilated box with an adjustable work opening that is used to minimise exposure to chemical vapours. It provides extraction to remove any fumes produced within the box. It is designed to have laminar flow through the front opening, i.e. the flow is to be even and non-turbulent through the open face of the cupboard.

To obtain even flow through the face of the fume cupboards baffles are generally installed at the back of the cupboard. These baffles are set to extract the air from two or more heights across the back of the fume cupboard. If the openings provided by the baffles are blocked by items stored in the cupboard then the air-flow through the face of the cupboard can become uneven.

Whenever anything is placed within the fume cupboard it introduces turbulence. This means that the containment of fumes may be affected. If a fume cupboard is not used in the proper manner then there may be situations in which fumes escape out of the front of the fume cupboard towards the user instead of being drawn away from the user.

Unless the room is of sufficient size or appropriately ventilated a fume cupboard will not be able to draw sufficient air and will subsequently not function properly.

The following details need to be considered to ensure that the fume cupboard's performance is not compromised:

- Do not work within ten centimetres of the leading edge. The larger the item, the further back it needs to be within the fume cupboard to overcome the turbulence created.
- Do not place storage items behind the area you are working in. This is of particular importance where a perspex screen or lead bricks are used for radioisotopes.
- Minimise the amount of items stored within the fume cupboard.
- The amount of flammable solvent placed in a fume cupboard should be minimised (and the subject of a risk assessment [6].
- Do not put large equipment, such as ovens in the fume cupboard, as they block the baffles and produce regions of zero or low flow in the workspace.
- Always have the sash as low as possible during the work.
- Minimise traffic past the front of the fume cupboard as this can cause turbulence and result in fume escape.
- The laboratory doors near the fume cupboard should be kept closed during its use.
- The make-up air supply and room ventilation should be on whenever the fume cupboard is in use.

Fume cupboard performance should be checked every 6 months to ensure adequate face velocity (an average of > 0.5 m/s) and laminar flow. This testing is arranged by Facilities Services. The cupboard should be checked to ensure is has a current test sticker prior to it being used. Records of testing are maintained in the Equipment folder.

Pipettes

- Mechanical or electronic pipettors are to be used for all pipetting tasks; never pipette by mouth.
- Because pipette tips can pierce a biohazard bag, they should be treated as sharps
 - do not remove them from the pipettor by hand
 - dispose of them in a sharps container at the bench
- The action of pipetting can form aerosols
 - Pipette slowly, particularly when using pipettes for mixing, to avoid aspirating aerosol or liquid into the pipette body.
 - Where aerosol transmission is a risk, carry out pipetting operations in a BSC.
 - Filtered tips or filter plugs may be required to avoid sample crosscontamination.
- Avoid bringing the body of the pipette into contact with the vessel you're pipetting from.
- Spray or wipe the body of the pipette over with disinfectant after use and store it upright.

• If infectious liquids are aspirated into the pipettor, do not continue to use the unit. Disassemble the unit in a BSC (wearing gloves) and decontaminate the components by soaking in disinfectant solution.

Continuous use of pipettes has the potential to result in forms of occupational overuse syndrome. The following points should be observed in order to minimise the risk of this occurring:

- Make sure the laboratory stool height is adjusted so the pipette can reach the work with the forearm and wrist held in a straight line, parallel with the work surface.
- Arrange the work to ensure that it can be reached without stretching; minimise the amount of pipette travel.
- Break from pipetting regularly at least 5 minutes every 30 minutes.
- When carrying out large numbers of transfers, use an electronic pipette in favour of a mechanical one.

Administrative Controls

General Laboratory Rules

- 1. No eating, drinking, smoking, handling contact lenses or applying cosmetics in the laboratory at any time.
- 2. Only self-adhesive labels shall be used; this prevents moistening of labels with the tongue.
- 3. Storage of food or drink in the laboratory is prohibited.
- 4. Long hair must be tied back.
- 5. Pipetting by mouth is strictly prohibited; always use the pipettors provided.
- 6. Only specified personnel can access the laboratory.
- 7. The laboratory doors must be kept closed when work is in progress.
- 8. Smelling/sniffing of bacterial plates is prohibited.
- 9. All cultures must be clearly identified and dated.
- 10. Decontaminate benches before and after working at them.
- 11. Laboratory gowns and gloves MUST be removed before leaving the laboratory.

Additional rules include the following:

Personal Hygiene

It is important that hands are washed correctly [8]. A suggested technique is as follows:

- Rinse your hands in warm running water.
- Apply about 5 mls (a squirt) of antibacterial product to the palm of the hand and rub the palms together to work up a lather.
- Using the method shown overleaf, wash both hands.
- Rinse under running water. Have the hands pointing down so that water drains from the fingers into the sink. This will remove the water and foam but the antibacterial soap will still be resident on the hands. Then pat your hands dry and turn the tap off with the paper towel.

Hands should be washed:

- Before leaving the laboratory (and *after* removing laboratory gown).
- Between glove changes.
- Whenever you suspect contamination.

Hand Washing Technique

1. Move from the palms to the inside surfaces of the thumb changing from left to right hand



3. Move over to the backs of the hands and then to the wrists giving it a few twists around the wrist.



5. Intertwine the fingers of both hands again to cover the webs of the fingers.



2. Intertwine fingers of both hands and work them back and forth to full length of fingers on each side.



4. From the wrist, move the hand on top over the backs of the fingers, including the thumb on the hand below.



6. Rub the nails and fingertips back and forth over the palm of the opposite hand.



Working Alone

In general, working alone in a PC2 laboratory does not in itself increase the risk of being injured. In addition, Risk Group 2 microorganisms do not pose an immediate threat to the health of anyone exposed to them. For these reasons it is only work with Risk Group 3 or higher organisms that is considered of sufficient risk to preclude working alone [9].

The primary concerns with people working alone are:

- The possibility that they undertake activities without appropriate training or approvals, or without using standard practices.
- The increase in risk associated with persons actually being alone, particularly with regard to personal safety.
- The lack of access to the emergency response support, particularly first aid, in the event that the person is injured or requires medical attention.

The following issues therefore need to be considered when contemplating work after hours:

- The nature of the work .
- The capacity of the person to conduct it (that is, their experience and training).
- Additional risk factors, such as any medical conditions.
- The means of communication available. On this point, Security should be notified so that they are aware of persons present in the building.

Where after hours work is necessary, this should be authorised by

Work Procedures

Innoculating Loops

These should be sterile before and after use. To avoid spatter and aerosol generation whilst sterilising loops in a Bunsen flame, slowly draw the wire through the tip of the blue cone, starting at the base of the wire, and ending with the loop. The loop should be completely closed.

Note there is the potential to contaminate loop handles when taking samples from deep tubes. If this is likely to occur, handles should be decontaminated by standing in disinfectant solution. This can be avoided by using sample tubes and loop wires of appropriate lengths where possible.

<u>Sharps</u>

Sharps are essentially anything that have the potential to penetrate the skin, but are typically needles, scalpels, Pasteur pipettes and broken glassware. The main risk with using sharps is self-inoculation.

- Keep the use of sharps to a minimum.
- Do not bend needles or try to recap them after use.
- Use blunt cannulas where possible.
- Discard sharps into sharp containers.

Decontamination

For the safety of all laboratory users items used in conjunction with infectious material must be decontaminated when they are finished with. This is achieved by:

- wiping with a disinfectant
- soaking in a disinfectant/sterilant/bactericide/viricide, or
- autoclaving (pressure steam sterilising)

The choice of decontamination procedure will depend on the microorganisms involved, the presence of other materials (chemicals, radioisotopes, organic material, etc.) and the equipment to be cleaned.

Chemical disinfectants are used for routine decontamination and spills. It is important that a decontaminant is effective against the microorganisms being handled is selected and available before work commences. Table 1 is provided to assist with the selection of disinfectants.

Table 1 Disinfectant selection (a bold tick indicates that the disinfectant is preferred for the application/agent).

Hypochlorite	Alcohols	Formaldehyde (gas)	QUATS	lodophors				
--------------	----------	-----------------------	-------	-----------	--	--	--	--

Use Parameters

Concentration	1-5%	70- 85%	5 g/m ³	0.1- 2%	0.5%		
Contact time (mins)	10 -	10 -	600 -	10-	10 -		
	20	30	900	30	30		

Effective against							
Vegetative bacteria	✓	✓	✓	~	~		
Bacterial spores	×	×	~	×	×		
Lipophillic viruses	✓	✓	~	~	~		
Hydrophillic viruses	✓		✓		×		
Fungi	✓		✓		~		
HIV	✓		✓	~	~		
HBV	✓		✓		×		
Applications		•	•	•	•	•	
Liquid discard	✓		×				

Liquid discard	v		~				
Glassware	✓		×	✓	✓		
Instruments	✓	~	×				
Equipment total decon.			✓				
Benchtops		~	×				

Important things to wipe daily include:

- door handles
- sink taps
- pipettors

Waste Disposal

Non-infectious material, such as waste paper and plastic products, is collected in the waste bins lined with plastic bags.

Infectious waste generated in the laboratory consists of several types, each of which have different disposal routes

<u>Sharps</u>

Are placed in the sharps containers. When these reach 75% full they should be replaced. For replacement and removal of sharps containers contact

Solid Waste

Items such as samples, culture plates and bottles, used gloves, etc., should be placed in the bio-hazard bins lined with an autoclave bag. When full, the contents of these bins should be . Operation of the autoclave should only be carried out by .

Note that containers placed in the autoclave should NOT be sealed - they should remain open to ensure the penetration of steam and to prevent them exploding. In addition, chemical or radioactive waste should not be placed in autoclave bags.

After autoclaving, bags should be placed

Liquid wastes

Small quantities of liquid waste can be sterilised by autoclaving.

Chemical disinfection may be used for liquid waste containing radioactive material.

Spills

Liquid spills generally have three components:

- the bulk liquid that puddles on the surface
- small splashes of liquid that are distributed around the spill area
- even smaller droplets that form airborne particles (aerosols)

If a spill of infectious liquid occurs:

- 1. Immediately evacuate the room and take steps to keep others out.
- 2. Remove all contaminated clothing and place in a bag for autoclaving.
- 3. Wash exposed areas of skin with copious amounts of soap and water; a shower may be necessary.
- 4. Allow sufficient time (30 mins) for the aerosol created to settle before re-entering the room.
- 5. Assemble a clean-up team of at least three people, one of whom should act as an observer.
- 6. Make sure sufficient disinfectant is available; freshly made bleach solution is the agent of choice.
- 7. Put on all necessary protective clothing required for dealing with a spill before reentering.
- 8. Cover the spill with absorbent material such as paper towels or a spill pillow.
- 9. Pour disinfectant around and on the absorbed spill; do not pour it directly on the spill, as this can generate more aerosol.
- 10. Collect the wet towelling or spill pillows into a biohazard bag for disposal.
- 11. Autoclave all items used to clean the area, including the protective clothing. Do not autoclave material containing hypochlorite, since chlorine gas can be produced.

All major spills must be reported to . A major spill is one in which:

- 1. hazardous materials contact skin, eyes, etc.,
- 2. a break in the skin occurs,
- 3. the spill splashes over an area larger than 30 cm in diameter,
- 4. the extent of the spill is undetermined, or
- 5. the spill involves an agent transmitted by aerosol.

Personal Protective Equipment

Vaccinations

The table below provides a list of Risk Group 2 pathogens where prophylactic vaccination is available and may be indicated [1].

Table 2 Vaccinations that are available for infectious agents; those shown in bold type are recommended for health care laboratory workers [7].

Bacteria	Viruses
Clostridium tetani	Hepatitis B
Corynebacterium diphtheriae	Influenza (recent isolates)
· · ·	Measles, Mumps, Rubella
Neisseria meningitidis	Varicella
Mycobacterium tuberculosis*	
Coxiella burnetii [*]	Vaccinia
Salmonella typhi [*]	Hepatitis A
	Polio ^{**}

These should only be considered Risk Group 2 when present in clinical or food samples Work with wild polio virus is subject to additional restrictions imposed by the World Health Organisation.

Where any of these pathogens is be used in the laboratory, **all** personnel using the facility should be considered as candidates for vaccination.

Advice and vaccinations can be provided through the University Health Service (UHS), and will be paid for from project funds. The UHS will maintain records of vaccination. Many users will have received vaccinations for a number of the agents listed above. A vaccination record for each user (Appendix H) will be maintained by

The UHS may also recommend health surveillance, such as chest X-rays, serum sampling, etc.

Work with pathogens may pose elevated risk to certain classes of people. Whilst the risks of exposure is the same for all persons who adopt the specified control measures, people who are more vulnerable to infection, such as those who are pregnant or diabetic or otherwise immuno-compromised, should notify and seek advice from the UHS if necessary.

Women of child bearing age need to be aware of the risks to an unborn child or themselves of exposure to certain microorganisms, such as:

Toxoplasma gondii Listeria monocytogenes Coxiella burnetii Rubella virus

Clothing & Footwear Requirements

It is a University requirement that enclosed footwear must be worn at all times in the laboratory - no bare feet, thongs, or sandals at any time. The main door of the laboratory indicates this requirement, which applies to any person entering.

Laboratory Gowns

Each person carrying out work with microorganisms will be issued with a wrap-around laboratory gown. Gowns shall:

- Be worn at all times whilst handling the microorganisms.
- Be removed and hung on the coat hooks **before** leaving the laboratory.
- Be laundered regularly, and when contamination is suspected. Laundry arrangements for the laboratory are as follows:

Visitors to the laboratory will be provided gowns when there is work being undertaken.

Eye Protection

Safety glasses shall be worn at all times when work is conducted in the laboratory. Over-glasses will be provided to those who wear prescription spectacles.

Certain operations, such as handling cryogenic liquids and samples, and unloading autoclaves, require the use of a protective face shield.

Gloves

Gloves shall be worn:

- When working in a BSC
- When handling human blood and body fluids
- When handling any of the following:
 - Clostridium botulinum
 - Mycobacterium spp.
 - Salmonella typhi
 - Shigella dysenteriae
 - Treponema spp.

(refer to [1] for RG2 parasites, fungi and viruses where gloves are also recommended).

Gloves do not provide automatic protection. This is due to the fact that even new gloves may have their integrity compromised. Consider double-gloving.

Gloves shall be changed regularly, washing hands between changes.

The standard glove available in the laboratory is a latex examination gloves. Latex gloves have been found to have the best integrity, greatest flexibility and sensitivity (allowing for dexterity) and least permeability. Allergic reactions to latex gloves, which range from mild skin irritation to anaphylactic shock, may be reduced by the use of non-powdered and low protein latex gloves. Alternatively, nitrile gloves can be used (which can also provide improved chemical protection).

Gloves shall be removed before leaving the laboratory or answering the telephone.

Instructions for the safe removal of contaminated gloves

1. Pull one glove near your wrist towards your fingertips until the glove folds over.



3. Pull the fold until the glove is almost off.



5. Slide your finger from your glove free hand under the remaining glove. Continue to slide your finger towards your finger tips until almost half of your finger is under the glove.



2. Carefully grab the fold and pull towards your fingertips. As you pull you are turning the inside of the glove outwards.



4. To avoid contamination, continue to hold the removed glove. Completely remove your hand from the glove.



6. Turn you finger 180° and pull the glove outwards and towards your finger tips. As you do this, the first glove will be encased in the second glove. The inside of the second glove will also be turned outwards.



7. Grab the gloves firmly, by the uncontaminated surface (the side that was originally touching your hand). Release your grasp of the first glove you removed. Pull your second hand free from its glove. Dispose of the gloves into a bio-hazard bag.



Risk Management

The preceding sections have covered three of the four steps in managing risks to health & safety:

For each project you intend undertaking you must demonstrate that you are managing its associated risks by completing a safety analysis.

This is conducted by conducting the hazard identification step using the Risk Management form in Appendix F. This information also needs to be provided to the Institutional Biosafety Committee when project approval is required.

This should be followed by a procedure safety analysis, which considers all hazards (chemical, biological, radiation, noise, manual handling, etc.) the tasks introduce. An example form is provided in Appendix G. Note that in many cases this analysis can be integrated with method documentation in laboratory notebooks.

Completed safety analyses must be:

- signed off by or prior to the work commencing
- included in laboratory workbooks
- stored in the Risk Management folder.

It is important that this safety analysis is reviewed whenever:

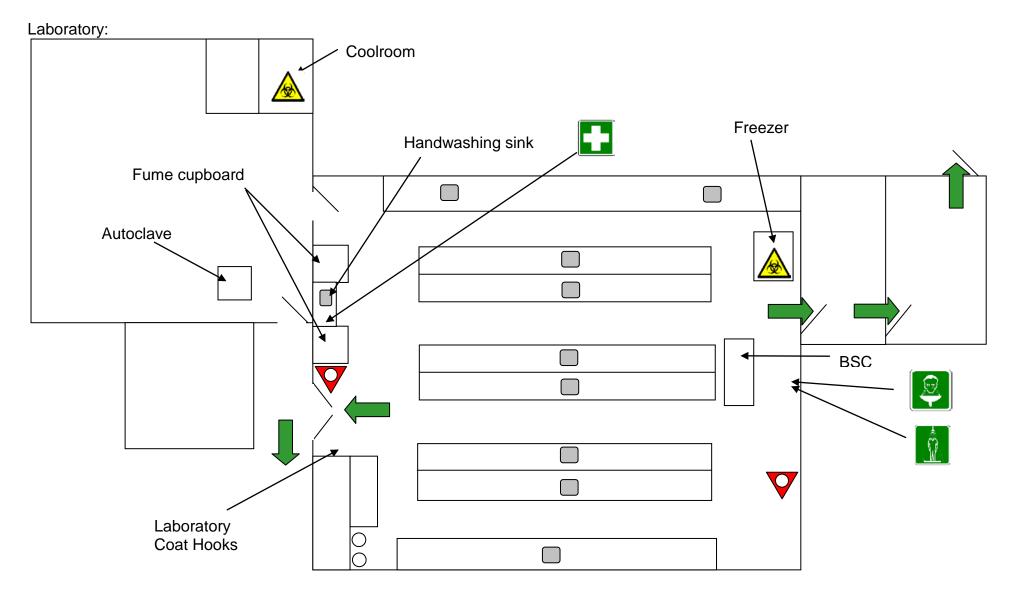
- The microorganisms used in the tasks are changed.
- An incident or accident resulting in potential exposure occurs.
- New information about a microorganism becomes available.

References

- 1. AS/NZS 2243.3:1995 Safety in laboratories. Part 3 microbiology, Standards Australia
- Second supplement to: Categorisation of biological agents according to hazard and categories of containment (4th Edition), 1995, Advisory Committee on Dangerous Pathogens, <u>http://www.hse.gov.uk/hthdir/noframes/agents.htm</u>
- 3. NIH Guidelines for research involving recombinant DNA molecules, 2001, National Institutes of Health, <u>http://www4.od.nih.gov/oba/rac/guidelines/guidelines.html</u>
- 4. Biosafety in Microbiological and Biomedical Laboratories (BMBL), 4th Edition, 1999, Center for Disease Control and Prevention & National Institutes of Health, <u>http://www.cdc.gov/od/ohs/biosfty/bmbl4/bmbl4toc.htm</u>
- AS 2647:2000 Biological Safety Cabinets Installation and use; AS 2252.1: 1994 Biological Safety Cabinets. Part 1: Biological Safety Cabinets (Class I) for personnel and environment protection; AS 2252.2: 1994 Biological Safety Cabinets. Part 2: Laminar flow biological safety cabinets (Class II) for personnel, environment and product protection, Standards Australia
- 6. AS 2243.8:2001 Safety in laboratories Part 8 Fume Cupboards
- 7. The Australian Immunisation Handbook, 7th Edition, NHMRC, 2000
- 8. http://www.newcastle.edu.au/department/fmhs/mrs/projects/handwash/How/how.html
- 9. AS 2243.1 Safety in laboratories Part 1 General

Contact Health & Safety to obtain further copies of these references.





Appendix B Induction Checklist

This is designed as an aid to inducting new personnel into the safety practices to be followed in laboratory.

All personnel should have access to the PC2 Laboratory Safety Manual, and be provided with explanations and demonstrations of its content. This shall occur before the person commences work in the laboratory.

Laboratory personnel are asked to verify they have received this information, and may be asked to confirm that they have understood it by way of questioning and demonstration. This checklist represents a record of induction, and should be kept in the Induction folder.

Laboratory: Supervisor: Department:

Item		Check
Review the contents of the PC2 Labo	pratory Safety Manual	
Laboratory Access requirements		
Location of fire fighting equipment		
Location of first aid kit		
Evacuation procedures		
Emergency eye wash and safety sho		
Location of safety documents:		
Laboratory Safety Manuals		
MSDS Folder		
Equipment Folder		
Risk Management Folder		
Issue personal protective equipment:	:	
Gown		
Safety Glasses		
Location of other personal protective	equipment	
Gloves		
Respirator		
Hand washing procedures		
Fume cupboard operation		
BSC operation		
Other items		
User's Name:	Signature:	Date:
Inducted by:		
Supervisor Acknowledgment:		

Appendix C Cleaner/Maintenance Personnel Induction Checklist

This is designed as an aid to inducting personnel who require access to the laboratory for the purpose of cleaning or facility maintenance (including maintenance of BSCs) to ensure they are aware of existing hazards.

All personnel should be provided with explanations and demonstrations of laboratory hazards. This shall occur before the person commences work in the laboratory.

Personnel are asked to verify they have received this information, and may be asked to confirm that they have understood it by way of questioning and demonstration. This checklist represents a record of induction, and should be kept in the Induction folder.

Laboratory: Supervisor: Department:

Item	Check
Demonstrate facilities	
Laboratory Access requirements	
Location of fire fighting equipment	
Location of first aid kit	
Evacuation procedures	
Emergency eye wash and safety shower	
Location of safety documents	
Personal Protective Equipment requirements	
Spill procedures	
Decontamination requirements	
Other items	

User's Name:	Signature:	Date:
Inducted by:		
Supervisor Acknowledgment:		

Appendix DSafety Training Plan and Assessment (Draft)

Laboratory:
Supervisor:
Department:

Laboratory User's Name:

Supervisor Acknowledgment:

Element	Date Assessed	Initials
I. DISINFECTION, DECONTAMINATION, STERILIZATION		
Select decontamination protocols (chemicals, steam, UV radiation, etc.) to kill or inactivate microorganisms		
II. WORK PRACTICES AND PROCEDURES		
Demonstrate safe use and disposal of sharps		
Select and explain the use of personal protective equipment (glasses, gowns, gloves)		
Explain the use of other safety equipment & devices (pipettes, loops, etc.)		
Demonstrate procedures for managing biohazardous spills and releases		
Explain the incident reporting procedure		
III. RISK ASSESSMENT/HAZARD IDENTIFICATION - INFECTIOUS AGENTS & GMOs		
Determine the hazard category of a microorganism using routes of exposure, modes of transmission and other criteria		
Explain the hazard of exposure of service personnel to biological materials		
List factors that may affect susceptibility, resistance or consequences of infection		

Outline the risks associated with recombinant DNA technology	
IV. REGULATORY ASPECTS, STANDARDS & GUIDELINES	
Interpret and apply the OGTR guidelines for research involving GMOs	
Interpret and apply NSW Bloodborne Pathogens Code of Practice	
Interpret and apply guidelines that classify biohazardous agents according to risk	
Interpret and apply regulations for packing, labeling, shipping of infectious materials, diagnostic specimens, and medical waste	
Interpret and apply import and export requirements associated with biological materials	
Interpret and apply OHS law, standards and directives as they relate to biohazards	
Interpret and apply guidelines and regulations relating to infectious and medical waste	
V. PROGRAM MANAGEMENT/DEVELOPMENT	
Describe the role and function of the Institutional Biosafety Committee	
Interpret and help maintain a biosafety manual	
Participate in occupational health programs for persons working with biological materials	
Interpret biosafety resource/reference information	
Participate in the infectious/medical waste management program	
VI. EQUIPMENT OPERATION AND CERTIFICATION	
Demonstrate the use of a Class II biosafety cabinet	
Describe the limitations in the use of equipment for work with biohazardous materials such as fume hoods and clean benches	
VII. FACILITY DESIGN	
Outline the functions of primary and secondary barriers	

Appendix E Register of Microorganisms

Laboratory:

Microorganism	Risk Group	Location	Box No.	Culture No.	Source	Strain/Type	MSDS (Y/N)	Import Permit No.	Comment
L									

Date:	
Project:	Chief Investigator:
Persons carrying out the work:	

Person conducting the risk assessment:

Microorganism Summary - Hazard Identification & Risk Assessment

This should be completed for each disease-causing microorganism with reference to available safety data

Pathogen	Disease(s)	Transmission Routes	Standard Risk Controls (circle)	Infectious Dose	Disinfectant	Vaccination required?	Vaccinations completed?
		(circle)				•	•
		Aerosol	Gloves				
		Ingestion	BSC				
		Inoculation	Mask				
			Sharps precautions				
		Aerosol	Gloves				
		Ingestion	BSC				
		Inoculation	Mask				
			Sharps precautions				
		Aerosol	Gloves				
		Ingestion	BSC				
		Inoculation	Mask				
			Sharps precautions				
		Aerosol	Gloves				
		Ingestion	BSC				
		Inoculation	Mask				
			Sharps precautions				
		Aerosol	Gloves				
		Ingestion	BSC				
		Inoculation	Mask				
			Sharps precautions				

Faculty/Division:		School/Unit:			
Procedure/Practical:		Persons Undertaking the Activity:			
Date:					
Prepared by:					
Signature(s):					
Step Describe each step of the procedure	Potential Hazard What can hurt people undertaking each step?	Controls and Standards What defines "safe", and what is in place to make each step safe?			

Reviewed by: _

Date

Appendix H Record of Vaccination Form

Name: _____

Signature:

Date: _____

	Past Illness (circle)	Year of last childhood vaccination	Date of last vaccination (other)	Comments
Diphtheria				
Tetanus				
Polio				
Measles	Yes / No			
Pertussis	Yes / No			
Mumps	Yes / No			
Rubella	Yes / No			
Hepatitis A	Yes / No			
Hepatitis B	Yes / No			
Tuberculosis Mantoux Test	+/-	Date of test		
BCG Vaccine (only if Mantoux -)				