S E C T I O N XX
AQUATIC ORGANISM C O N T A I N M E N T
F A C I L I T I E S

6.1 REQUIREMENT FOR AQUATIC ORGANISM CONTAINMENT FACILITIES

This Section sets out requirements to ensure that aquatic organisms that are infected with, or that may contain infectious microorganisms, are contained in facilities that will prevent the escape of the microorganism or the aquatic organisms at any life stage. The general principles of aquatic organism containment can also be applied to aquatic organisms that do not contain any infectious microorganisms, such as specific pathogen free (SPF) aquatic organisms and genetically modified or transgenic aquatic organisms. This Section is not intended to be used as a substitute for other regulations or guidelines that apply to these aquatic organisms, such as those issued by DAFF, OGTR or equivalent New Zealand regulatory agencies.

In Australia, aquatic organisms exposed to exotic microorganisms shall be housed in containment facilities that meet the requirements of DAFF. Aquatic organisms exposed to genetically modified microorganisms shall be housed in accordance with OGTR requirements. Disposal of such aquatic organisms, including aquatic organisms at early reproductive stages such as eggs, and including potentially contaminated water, shall be in accordance with the relevant regulations or guidelines.

In New Zealand, aquatic organisms exposed to exotic microorganisms and aquatic organisms exposed to genetically modified microorganisms shall be housed in facilities approved by MAF. Disposal of such aquatic organisms, including aquatic organisms at early reproductive stages such as eggs, and including potentially contaminated water, shall be in accordance with the relevant regulations or guidelines.

Facilities and arrangements for aquatic animal care and management shall be consistent with good aquatic animal welfare practices and in accordance with either the *Australian code of practice for the care and use of animals for scientific purposes (reference 1.34)* or the New Zealand animal *Welfare Act 1999*, as appropriate.

NOTE 1: The *Guidelines to promote the wellbeing of animals used for scientific purposes: The assessment and alleviation of pain and distress in research animals* should also be consulted (reference 1.35).

NOTE 2: Refer to "Guide for the care and use of laboratory animals", US, NRC reference 1.36 for specific information related to aquatic animal husbandry.

NOTE 3: The USDA-ARS (United States Department of Agriculture, Agricultural Research Service), national agricultural library has some excellent reference material related to the care and use of species such as molluscs, see reference 1.37.

It should be noted that the aquatic environment can contain organisms and microorganisms other than the aquatic organisms of interest. This can be intentional, such as aquatic environments that include the relationship of aquatic organisms with nominated or known microorganisms. It can also be non-intentional, such as the presence of unknown or unwanted microorganisms in the aquatic environment itself. These considerations need to be borne in mind when considering appropriate containment measures.

Aquatic organism handling can be uniquely hazardous for research, care and management personnel. Hazards can include injury from physical impact, biting, stinging, exposure to toxins, and puncture wounds from fins or spines. Some species of sea organisms are amongst the most toxic in the world. These can include aquatic animals, some marine reef organisms and some species of aquatic plants. Personnel may need to enter the water in larger aquatic containment environments, adding hazards such as drowning and requiring operators to use specialist breathing equipment.

6.2 PRINCIPLES OF AQUATIC ORGANISM CONTAINMENT

Aquatic organisms can be held in a variety of containment facilities that are designed to ensure that the aquatic organisms, and the microorganisms that may be being used in conjunction with the aquatic organisms, do not escape from containment. Aquatic organisms under experiment may be either small laboratory aquatic organisms (e.g. zebrafish, snails, marine plankton and corals) or larger animals (e.g. crocodiles, sharks). The requirements for housing and maintenance of aquatic organisms may differ in scale as a result but the overall principles that apply are the same. While some aquatic organism PC1 facilities confine larger aquatic organisms in a secure and fenced off pond or pool, other aquatic organism PC1 facilities are designed to contain smaller aquatic species such as zebrafish and Xenopus in aquatic environments such as tanks or aquaria.

Facilities may be designed and constructed in the same way as a laboratory and may be integral to, and inseparable from, the laboratory itself. At lower containment levels (aquatic organism PC1 and aquatic organism PC2), there may be little difference between the design and construction of aquatic organism containment and laboratory containment facilities.

The use of primary containment devices should be considered at all levels of aquatic organism facilities. Separate primary containment supports the prevention of contaminant spread, assists with the control of cross-contamination, and may help to reduce personnel exposure.

For the purposes of this section, primary containment for aquatic organisms shall be taken to be a level of protection such that:

-Aquatic animals are unable to escape the primary containment environment
-Lids, caps or seals are provided so that the normal behaviour of the aquatic organisms, including surface-breaking activity, jumping, spraying, does not result in liquid escaping from the primary containment device.
-If primary containers are designed to be moved or carried, measures are in place to minimise the likelihood and quantity of spillage if the container is dropped or tipped. This can include sealing lids with restraints to hold them in place.
-Routine maintenance of the aquatic organism environment can be carried out with minimal opening and closing of the primary containment devices. This includes features such as automated water circulation, automated aeration and facilities for the introduction of feed.

Where smaller aquatic organisms may be infected with, or exposed to, Risk Group 3 microorganisms, it is preferable that they are kept in primary containment devices that prevent the laboratory environment from being exposed to the Risk Group 3 micro-organims.

Where smaller aquatic organisms may be infected with, or exposed to, Risk Group 4 microorganisms, it is preferable that they are kept in some form of primary containment device

which is sealed gas-tight and water-tight from the laboratory environment and is fitted with supply and exhaust HEPA filters. Additional special precautions may be required for these facilities to control liquid leakage or spillage hazards, especially in situations where the Risk Group 4 microorganisms may be present in the liquid.

In situations where it is not possible to keep aquatic organisms in primary containment devices (e.g. for large aquatic species such as crocodiles or sharks), or in situations where the enclosures are unable to be designed to prevent the spread of water spray or aerosols, the room itself will form the primary containment environment. In these situations additional measures shall be taken to provide containment based on the hazards associated with the microorganisms that may be present. All of these features shall be supported by good work practices and training to ensure the protection of personnel and the environment.

Measures for consideration include:

i) Bunding and other liquid escape mechanisms
ii) Additional construction standards to control spillage, leakage and ease of cleaning
iii) Aerosol escape prevention mechanisms, including HEPA exhaust filters at the higher containment levels
iv) The use of dedicated PPE that remains in the facility
v) Showering of personnel before leaving the facility
vi) The use of specialised PPE for some species
vii) The use of specialised training and work practices for some species

Liquid waste from aquatic organism facilities will often be a major containment risk. Liquid waste from low level (PC1 and PC2) aquatic organism containment facilities can often be decontaminated inside or adjacent to the facility using proprietary filtration and chemical or UV technologies. Liquid waste from higher containment level aquatic organism containment facilities shall either be pressure steam sterilized in a closed batch heat treatment system or shall be effectively decontaminated by some equivalent means to ensure that infectious microorganisms are destroyed. When liquid waste is transported for treatment at locations remote from the facility, measures shall be taken to address the risks of leakage or spillage such that effective containment is maintained until liquid waste is decontaminated.

Solid waste shall be segregated, decontaminated where necessary and disposed of according to applicable regulations. (See also Section 12.) Aquatic organism containment facilities should have access to decontamination facilities within their own areas. Waste from low level (PC1 and PC2) aquatic organism containment facilities can be decontaminated outside the facility. However, waste shall be contained to prevent dissemination of any infectious microorganisms. Waste from higher level aquatic organism containment facilities shall either be pressure steam sterilized in the facility or decontaminated in a closed system to ensure that all infectious microorganisms are destroyed.

As a general principle, the biological and physical containment recommended for working with infectious agents in vivo and in vitro are comparable. Infected aquatic organisms should only be handled by trained staff using appropriate procedures designed to protect staff and the environment from exposure to the microorganisms. When housing aquatic organisms in which microorganisms are to be used, the physical containment levels for work with microorganisms

shall follow the containment levels appropriate for the microorganism. Requirements for aquatic organism PC1, aquatic organism PC2, aquatic organism PC3 and aquatic organism PC4 facilities are set out in Clauses 6.4, 6.5, 6.6 and 6.7.

6.3 OTHER CONSIDERATIONS ASSOCIATED WITH AQUATIC ORGANISM CONTAINMENT

6.3.1 Designing facilities for different aspects of aquatic organism handling

Managers and operators shall obtain information from reliable sources to assure that the facility can be operated with good practice methods. This includes animal husbandry, inspection and monitoring for disease or illness, appropriate response to incidents involving the species being cared for, and taking care to optimise the health of animals.
Prior to designing facilities, separate areas should be considered for different activities, for example housing, experiments, post-mortem examinations, disposal of wastes and associated maintenance. This separation is dependent on species and type of aquatic organism. Applicable aquatic animal welfare guidelines should be consulted and followed.

Infected, non-infected and quarantined aquatic organisms should be separately housed, and precautions taken to prevent cross-infection.
Infected aquatic organisms, aquatic organisms suspected of being infected, quarantine aquatic organisms and diseased or sick aquatic organisms shall be held in separate containers from other aquatic organisms and shall be provided with their own water supply to prevent infection of healthy aquatic organisms.
Even aquatic organisms that have not been deliberately infected may harbour organisms that are dangerous to other aquatic organisms, and in some instances to humans.

Training staff in aquatic organism handling is the best method of preventing injury, both to staff members and to the aquatic organisms. Refer to clause 6.1 for some useful references associated with the handling of aquatic organisms.

6.3.2 The occurrence of allergic reactions in personnel handling aquatic organisms

Some aquatic animal and plant species can cause allergic reaction and this hazard should be considered when applicable. Inhalation is one of the most common ways for allergens to enter the body. To reduce the incidence of these conditions, adequate ventilation, including an increased number of air changes per hour, should be ensured and local exhaust systems provided where necessary (see Clause 6.3.3). In addition, aquatic environment handlers, technical and scientific staff should take appropriate precautions to prevent the development of allergies.

Any unusual personal reaction or allergy to the aquatic species present in the facility should be reported so that appropriate action can be taken.

It is recommended that respiratory protection is worn in situations where allergenic material may be present in the aquatic facility to prevent the development of allergies. Usually P2 particulate respirators are adequate but fit testing of the respirator is important to ensure that it is appropriate for the individual and advice from an occupational hygienist or similar should be sought.

6.3.3 Air change rates for aquatic organism containment facilities

Aquatic organism containment facilities require rates of fresh air ventilation to control moisture build-up, odours and contaminants from materials such as detritus and uneaten food, and to dilute aerosols. In consideration of the long term exposure of personnel, the fresh air ventilation rate shall be sufficient to keep moisture, odour and contaminant levels below acceptable threshold limits.

NOTES:

1 The required fresh air ventilation rate depends on a number of factors, including—

(a) the type of aquatic organism containers;

(b) the nature of aquatic organisms to be accommodated;

(c) the density of aquatic organisms and the liquid surface area in relation to room volume;

(d) the organism husbandry, particularly the rate of water circulation and the frequency of replenishment;

(e) feed and quantity of waste material that may be present;

(f) temperature, humidity and air movement; and

(g) the ventilation effectiveness.

2 Guideline minimum fresh-air ventilation rates for organism containment facilities are as follows:

Organism housing	**Fresh airflow (air changes per hour)**
Open tanks, i.e. the room forms the primary containment barrier	10-15, species dependent
Aquaria tanks or containers fitted with lids	8-12, species dependent
Isolation devices where all exhaust air is completely removed from the aquatic organism environment by capture hood or direct-ducting. These include HEPA filtered isolators at higher levels of containment	8

3 The rate may need to be increased for animal welfare reasons or to reduce concentrations of airborne substances such as pheromones. This can be important in facilities housing multiple animal species.

4 Note that many aquatic facilities can involve significant areas of exposed water surface. The evaporation rate from these surfaces can vary markedly depending on the temperature and humidity of the air, and the nature and directional flow of any ventilation systems. These evaporation rates can, in turn, affect the temperature profiles, heat loss and water loss from tanks. These factors should be considered carefully when designing these systems.

6.3.4 Decontamination and disposal of aquatic organism waste

Potentially contaminated containers from small aquatic organisms shall be decontaminated prior to disposal or reuse as described in Section 12. Infected carcasses shall be decontaminated prior to disposal. This may be achieved by methods such as alkali digestion, autoclaving, incineration or rendering. All instruments and containers that have been used in procedures with infectious microorganisms should be decontaminated before cleaning. Any special precautions that are needed, such as decay of radioisotopes, should be taken.

NOTE: Decontamination requirements apply for aquatic organism PC2 and higher facilities. See Clauses 6.5 to 6.7.

6.3.5 Transport of aquatic organisms and organism tissues between facilities

Where it is necessary to transport aquatic organisms or aquatic organism tissues from the containment facility, the appropriate precautions shall be determined. Tissues fixed to inactivate infectious materials may be removed from the facility. Live aquatic organisms and potentially infected aquatic organism tissues shall not be moved to a facility of a lower level of containment, e.g. from PC3 to PC2. (See Clause 3.5.)

6.3.6 Dissection and post-mortem examinations

Post-mortem examinations of aquatic animals that are infected or suspected to be infected with pathogenic microorganisms shall be carried out under physical containment conditions equivalent to the risk group of the microorganism present or suspected to be present.

During post-mortems, appropriate PPE such as gloves, aprons and eye protection should be worn. Where there is a risk of infection by the respiratory route, respiratory protection shall be used.

6.4 REQUIREMENTS FOR AQUATIC ORGANISM PC1 FACILITIES

6.4.1 General

An aquatic organism PC1 facility is suitable for work with microorganisms in Risk Group 1 and uninfected aquatic organisms. Microbiological containment is generally addressed by good work practices.

A sign complying with Appendix D showing the level of containment, together with hazard symbols as appropriate and any access restrictions should be prominently displayed at the entrance.

6.4.2 Construction

Aquatic organism PC1 facilities shall comply with the following:

(a) Facilities for different activities shall be appropriately segregated, considering containment, cross-contamination, animal welfare aspects. Separate areas shall be provided for:
(i) Quarantine
(ii) Breeding and production
(iii) Experimental work and procedures
In some cases separate rooms should be provided for different activities. However these requirements are species dependent.

(b) Facilities shall be constructed to prevent the escape of the aquatic organism species being contained.
NOTES:
1 The facility should be secure against incursions by feral or predatory organisms.
2 Where fencing is required, electric fencing and buried fencing should be used where appropriate. (Applicable to large amphibious facilities, such as for reptiles)

(c) Facilities shall be designed to prevent the access of unauthorized personnel.
(d) Facilities shall be designed and constructed to prevent infestation by undesirable vermin.

(e) Facilities shall be constructed in a manner that allows regular cleaning and, if appropriate, decontamination.

(f) Backflow prevention for water supplies shall comply with Appendix E.

(g) Gas supplies in the facility shall comply with the general requirements specified in Paragraph E3.1 of Appendix E.

(h) Dissection tables shall be impermeable to liquids and be covered with a washable material.

(i) Where no recognised human pathogens are present and workers are at risk of dehydration due to circumstances such as heat stress, heavy work or long shifts in response to emergencies, drinking facilities may be provided to the facility. This shall be subject to approval by the BC, following a risk assessment of the particular situation. If drinking facilities are provided, they shall be via a hands-free operation drinking fountain in a designated area.
NOTE: This option is only for aquatic organism PC1 and aquatic organism PC2 facilities.

(j) Equipment rooms containing potentially exposed contaminated material, such as liquid from aquatic storage tanks, shall be considered part of the containment facility and shall be constructed and operated at the same containment standard.
NOTE: Fully sealed treatment processes may be considered in equipment rooms of a lower containment standard provided that suitable measures are in place to safely and securely manage a failure, leak, or breach of containment.

6.4.3 Work practices

The work practices for aquatic organism PC1 facilities shall be as follows:

(a) Access to the facility shall be restricted to authorized personnel.

(b) All means of access to the facility shall be locked when aquatic organisms are not under direct supervision.

(c) For containment of open pond or amphibious habitats, the external perimeter fence shall be checked at least every three months and after storms for any breaks or holes in the fence. Any breach shall be repaired immediately.

(d) Other provisions such as feed and water supplies and regular inspections shall meet requirements for aquatic organism husbandry and welfare purposes.

(e) Aquatic organisms shall be prevented from escaping, with reasonable contingencies in place for accidents such as during handling.
NOTE: The doors should be kept closed when experimental aquatic organisms are present, and for those periods when work is being carried out within the facility.

(f) PPE appropriate for the work being carried out shall be worn. See also Clause 10.2 for detailed information on PPE. For all work with aquatic organisms, either dedicated facility clothing shall be worn or personal clothing shall be covered by suitable overalls, laboratory coat or gown. Closed footwear shall be worn, preferably separate shoes or boots that remain within the aquatic organism containment facility. Often in aquatic facilities PPE must be water resistant and especially appropriate for the activities being carried out.
NOTES:

1 Gloves, reinforced if necessary, should be considered when working with aquatic organisms and when working in a BSC.
2 Eye protection should be considered when working with aquatic organisms.
3 Protection against stings, scratches and bites should be considered.
4 P rotection against inhalation of aerosols or skin exposure to toxins should be considered if applicable.
5 Facility clothing should be laundered at appropriate intervals.

(g) Staff handling aquatic animals shall be trained in fundamental aspects of good animal husbandry. Staff shall be familiar with safe handling procedures for the aquatic organism species involved, including appropriate restraint procedures; staff shall understand the nature and hazards of any infectious agent involved and how it can be transmitted, the inoculation method to be used, how subsequent sampling is to be done, safe disposal of liquid effluents and organism waste, and emergency procedures.

(h) Staff shall be competent in inoculation procedures designed to prevent self- inoculation and to minimize aerosol formation.
NOTE: When handling or inoculating animals, the introduction of microorganisms through the skin either by accidental self-inoculation or by contact with ecto-parasites is a real risk.

(i) Aquatic animals shall be appropriately restrained during handling and procedures.

(j) Aquatic organisms shall be properly identified (e.g. by tattooing, microchip, tags, permanent branding or labels on aquaria) and accounting procedures shall be established.
NOTE: A record should be maintained to provide an up-to-date inventory of the aquatic organisms present and a chronological record of procedures performed.

(k) During post-mortem examinations, spillage trays and containers for used instruments shall be used. Procedures shall be followed to avoid cuts with the instruments used.

(l) Eating, smoking and the storage of food for human use shall not be permitted inthe facility.

(m) Drinking and the storage of drink for human use shall only be permitted in the facility in accordance with Item 6.4.2(i). If drinking facilities are provided, work practices shall be instituted to ensure gloves are removed and hands are decontaminated prior to drinking.

(n) Chemicals shall be stored in the facility in accordance with AS/NZS 2243.10.

(o) PPE shall be removed and hands shall be decontaminated before leaving theaquatic organism containment facility.

(p) Segregate wastes (e.g. broken glassware, biological and radioactive substances) and dispose of according to applicable regulations, using the most appropriate and effective method for the materials concerned. See also Section 12.

6.5 REQUIREMENTS FOR AQUATIC ORGANISM PC2 FACILITIES

6.5.1 General

An aquatic organism PC2 facility is suitable for work with infectious microorganisms in Risk Group 2 and incorporates all the requirements of an aquatic organism PC1 facility with additional requirements of construction, access, safety equipment and staff training.

6.5.2 Construction

In addition to the construction requirements specified for aquatic organism PC1 facilities in Clause 6.4.2, the following shall apply:

(a) Floors, ceilings, walls, internal furnishings such as blinds and curtains of the facility shall be smooth, easy to clean, and resistant to commonly used reagents and disinfectants. Finished surfaces shall be impervious and shall support regular and vigorous cleaning procedures that can be associated with wet facilities. Floor to wall joints shall be provided with continuous waterproof coving to facilitate cleaning. Where this coving forms part of the bunded facility volume, there shall be mechanisms in place to ensure the bunding requirement is not compromised at openings such as doors. Some aquatic facilities include aquatic environments containing salt water or chemically treated water. Facility construction, surfaces, finishes and ducts and pipes carrying air and water shall be suitable for the required environments.
NOTE: The doorway and room structure should be rodent-proof.

(b) Structural joints, where required, shall be durable, impermeable, easy to clean and shall resist deterioration due to commonly used cleaning agents and, where applicable, exposure to ultraviolet radiation.
NOTE: Structural joints should be minimized in containment facilities.

(c) Access doors shall be self-closing and be designed and installed to minimize the possibility of aquatic animals escaping.

(d) Windows shall be closed and sealed.

(e) Facilities shall be in place to deter the entry and exit of invertebrates. This will depend on the types of invertebrates attracted to the facility and the location. Facilities that should be considered include electric/UV insect killers, air curtains, foot baths, sticky strips, ante room, light-controlled ante room, seals and drop-down seals to doors.

(f) Any openings in the walls, roof or ceiling, such as vents and air-conditioning or ventilation inlets and outlets, shall be screened at the containment boundary with fine mesh screens having apertures of sufficiently small gauge to prevent entry or egress of invertebrates. The mesh shall be—

(i) stainless steel; or

(ii) a suitable material with regards to its—

(A) mechanical strength under the airflow load;

(B) ability to remain undamaged with regular vigorous cleaning;

(C) corrosion resistance; and

(D) resistance to attack by insects from either inside the containment facility or from the local environment outside the facility.

NOTES:
1 For aquatic facilities the requirements should be based on a suitable risk assessment and there is no specific recommendation. Conventional fly screening, with filters to ventilation supplies and exhausts, may be adequate in many facilities.

2 For applications where hazards are unknown, or for future flexibility, the recommended maximum aperture size is 0.25 mm (250 µm). Standard stainless steel mesh with an aperture of 0.25 mm and wire gauge of 0.16 mm satisfies this requirement. Smaller aperture sizes of 0.10 mm (100 µm) may be required for work which involves some arthropod varieties such as mites and thrips.
3 In locations where dust and debris can be generated, the use of roughing filters upstream of the mesh screens can result in safer and easier cleaning.

(g) Bunds or containers shall be provided to capture any potential spillage or overflow from aquatic organism containers. The bunding shall be of sufficient capacity to contain the largest single aquatic organism container, or group of containers where interconnection could result in leakage from multiple containers. The potential for leakage to occur from pipes, filters, pumps and other liquid handling equipment shall be considered when assessing bunding requirements. If the floor is used as the bunding zone, any waste openings located at or below the bund fill level, such as floor wastes or low level tundishes, shall be sealed closed or otherwise protected during normal facility operation. This is to prevent unintentional release of potentially contaminated material. Seals are not required if the liquid waste is connected to an effective liquid decontamination system during normal operation.
(h) Liquid drainage exits shall be protected against entry or exit of rodents and invertebrates by the use of adequately replenished traps or by an equivalent effective method.
(i) Provision shall be made for decontamination of liquid effluents in a manner appropriate to the type of waste. The method of decontamination and disposal shall be determined using the results of a risk assessment based on the likely composition and volume of the waste and in accordance with applicable regulations. See also Section 12.
Liquid waste treatment, where provided, shall be located as close to the aquatic containment facility as possible to minimise the length of potentially contaminated pipes.
Provisions shall be in place to manage any potential backup and to prevent discharge of untreated liquid in event of a failure. This includes any failure or leakage within the room housing the liquid waste treatment equipment. Any such failure shall be alarmed to warn the facility staff.
Liquid waste treatment systems, where provided, shall incorporate sampling ports and monitoring capability to permit testing of system efficacy with logging and recording of treatment processes.
Any pipes carrying potentially contaminated liquids outside the aquatic facility boundary shall have provisions for leakage testing, inspection and maintenance.
All liquid waste components outside the aquatic facility shall be secure against unauthorised entry or access and shall be labelled to minimise the risk of the accidental breach of containment.

(j) Containers in accordance with Section 12 shall be provided for collection, storage or disposal of infectious materials.
(k) A pressure steam sterilizer shall be available where steam sterilizing of facility wastes is required. See also Clause 10.6 and Section 12.
NOTE: The pressure steam sterilizer should be as close to the facility as possible.

(l) A PC2 facility which includes aquatic organisms shall be equipped with a hand basin with hands-free mixing taps near the personnel exit. The hand basin may be shared with other PC2 areas if a risk assessment deems this to be appropriate. Note that hand washing in a hand basin is considered essential prior to exiting the PC2 environment. Disinfectant hand rubs are not considered appropriate substitutes because of the potential requirement to remove contaminants such as dirt, detritus and other solids.
NOTES:

1 Shower facilities should be provided within the same building as the facility.
2 Consideration should be given to the provision of a hand basin or a hands-free dispenser providing appropriate disinfectant hand rub in individual aquatic organism holding rooms to reduce cross-contamination risk. See Reference 1.9. Note that this option can be appropriate in individual rooms for situations where water borne microorganisms may be present.

(m) Where required, storage space, e.g. shelves, shall be provided for reference documents and papers within the facility and separate from the work surface.

(n) A sign complying with Appendix D showing the biological hazard symbol and the level of containment, together with hazard symbols as appropriate, emergency contacts and any access restrictions shall be posted near the entrance to the facility.

(o) An area in which protective clothing and footwear can be stored shall be provided. If aquatic organisms are not in primary containment devices, this area shall be in an anteroom situated within the facility.

(p) Where the room itself forms the primary containment measure, and if determined necessary on the basis of a risk assessment, the following shall be provided:

(i) an anteroom, to allow changing of clothes on entry and exit and the storage of specialized PPE; and
(ii) a shower facility for staff exiting the room.

(q) Where the room itself forms the primary containment measure, and if determined necessary on the basis of a risk assessment, the following shall also be provided:

(i) an inner and outer change room on each side of the shower facility to allow staff to fully change clothing on entry and exit to the room.

6.5.3 Ventilation

An inward flow of air shall be maintained by forced extraction of air to minimize the spread of aerosols in the event of an inadvertent spill. Air may be recirculated for aquatic plant facilities and for aquatic animal facilities where aquatic organisms are kept in primary containment devices that are separately exhausted. If air is recirculated, it shall not be supplied to areas outside the aquatic organism PC2 facility.
Where an aquatic PC2 animal facility forms the primary containment measure, air shall not be recirculated.

Ventilation air shall not be directed towards doors or located in positions that can disturb air flow at a BSC.

Ventilation systems shall take account of the increased moisture burden that can occur in aquatic containment facilities.

6.5.4 Containment equipment

6.5.4.1 *Biological safety cabinets*

A Class I or II biological safety cabinet (see Clause 10.7) shall be provided if work with microorganisms transmissible by the respiratory route or work producing a significant risk from aerosol production is anticipated.

Installation and use, including the decontamination of the biological safety cabinet, shall be performed in accordance with the requirements of AS 2252 part 4.

6.5.5 Work practices

In addition to the work practices described in Clause 6.4.3 for aquatic organism PC1 facilities, the following work practices shall be observed:

(a) Protective clothing and footwear shall be worn in the facility. Gloves and eye protection shall be worn when handling aquatic organisms or material containing Risk Group 2 microorganisms. Where splashing of potentially contaminated material may extend to operator's clothing, precautions shall be taken to ensure this does not contaminate personal clothing or skin.

(b) Maintenance personnel shall be advised of potential hazards before entering the facility. Areas or equipment being maintained shall be decontaminated before the maintenance is carried out. Equipment shall be decontaminated prior to removal from the facility.
NOTE: Appendix F provides information on disinfectants.

(c) All clinical and diagnostic specimens shall be regarded as potentially hazardous. Leaking containers of potentially contaminated material shall be handled in a biological safety cabinet and the outside of the container decontaminated (see Table F1). Where a replacement sample is readily obtained, the leaking specimen shall be decontaminated and discarded in accordance with Section 12.

(d) For manipulations with aquatic organisms that could result in an aerosol containing viable microorganisms, a biological safety cabinet or other equipment designed to contain the aerosol shall be used. For larger aquatic environments, the room becomes the primary containment measure. This may require specialist personnel protective equipment.

(e) Care shall be taken in the use of syringes, needles and other sharps. Sharps containers shall be provided at each point of use. Precautions shall always be taken with any contaminated sharp items, including needles and syringes, slides, pipettes, capillary tubes and scalpels. Needles and syringes or other sharp instruments shall be restricted for use only when there is no alternative, such as for parenteral injection, phlebotomy or aspiration of fluids from aquatic organisms and diaphragm bottles.
NOTES:
1 Sharps use should be eliminated wherever possible.
2 Staff should be aware of the potential for glass equipment such as pipettes and tissue grinders to become sharps during an accident. Plastic ware should be substituted for glassware whenever possible.
3 Where infectious material is being injected under high pressure, Luer-lock fittings should be used.

(f) Viable microorganisms or aquatic organism tissues being transported out of the facility shall be double-contained. The second container shall be closed and unbreakable and the surface shall be decontaminated before removal. If taking live aquatic organisms out of the facility, they shall be contained in a manner that prevents dissemination of the microorganism and prevents escape of the aquatic organism.

(g) Work surfaces shall be decontaminated after use, after any spill of viable material, and before maintenance is carried out in the area.
NOTE: Appendix F provides information on disinfectants.

(h) Personnel shall decontaminate their hands after handling cultures and aquatic organisms.

(i) PPE shall be removed and hands decontaminated in an appropriate, pre-determined order before leaving the aquatic organism containment facility.

(j) Gowns or other protective clothing shall be laundered at appropriate intervals. If infectious materials have been spilled on gowns or protective clothing, these items shall be decontaminated, preferably by steam pressure sterilization prior to laundering or disposal. (See Section 9).
NOTES:
1 The order is usually removal of gloves followed by hand decontamination, then eye protection and laboratory gown followed by a second hand decontamination.
2 Appropriate protocols for laundering or decontaminating PPE should be implemented.

(k) Potentially contaminated re-usable laboratory ware shall be collected and disinfected or decontaminated in accordance with Section 12 prior to washing and reuse.
NOTES:
1 For chemical disinfection, pipettes placed vertically in an appropriate disinfectant solution, tip-first and fully immersed, minimize production of aerosols.
2 Thermal decontamination of pipettes that are not fully immersed in a liquid, can only be achieved in a pre-vacuum steam sterilizer.

(l) Microbiological wastes, aquatic organism environmental enrichment material, aquatic organism cages and aquatic organism carcasses shall be decontaminated. Waste should be disposed of in accordance with Section 12.

(m) Facility rooms shall be cleaned and decontaminated after use.

(n) Report writing and long-term write up shall occur outside the facility.
NOTE: Worksheets may be used on the bench.

6.6 REQUIREMENTS FOR AQUATIC ORGANISM PC3 FACILITIES

6.6.1 General

An aquatic organism PC3 facility is suitable for work with infectious microorganisms in Risk Group 3 and incorporates equipment and practices for aquatic organism PC1 facilities except Item6.4.2(i) of Clause 6.4 and all equipment and practices for aquatic organism PC2 facilities (Clause 6.5); however, additional requirements for construction, conditions of access, safety equipment and staff training apply.

For aquatic microorganisms, the escape risks are not likely to be the same as for PC3 laboratories or PC3 animal facilities. This is because aerosol risks are less likely to be of significance for aquatic species, but liquid waste risks are likely to be greater. Although the capability of gaseous decontamination is considered essential in event of an accident or spill, the requirement for HEPA exhaust air filtration for aquatic PC3 microorganisms shall be based on an appropriate risk assessment.

When pathogenic microorganisms of Risk Group 3 are being used in association with small aquatic animals and plants, primary containment devices should be used wherever practicable. Where primary containment devices cannot be used, the facility forms the primary containment measure.

NOTE: The design of a PC3 facility is complex and those planning its construction should seek specialized advice. See also Appendix G for examples of recommended layouts for PC3 facilities showing the design principles involved and Appendix H for airtightness considerations.

6.6.2 Construction

Construction requirements applying to aquatic organism PC1 and aquatic organism PC2 facilities also apply to aquatic organism PC3 facilities, but with the removal of the option to provide drinking water. In addition to construction requirements described for aquatic organism PC1 and aquatic organism PC2 facilities in Clauses 6.4.2 and 6.5.2, the following shall apply:

(a) The facility shall be physically separated from non-PC3 areas, including offices used by facility personnel, and areas accessible by the general public. This separation shall be achieved by a double-door system where entry to the facility is gained only through an airlock or shower airlock. For an airlock, the doors shall open outwards and the outer door shall be lockable. For a shower airlock, the outer door of the shower airlock shall open outward. Either the door to the outer change room or the outer door of the shower airlock shall be lockable. Airlock and shower airlock doors shall be self-closing and fitted with seals to limit air leakage.

Where the facility forms the primary containment measure, an outer and inner change room, separated by a shower airlock shall be provided. The outer shower door shall form the limit of the aquatic organism PC3 containment for decontamination purposes.

NOTES:

1 Where separate aquatic organism rooms are contained within a PC3 facility, consideration should be given during the design of the air handling system to the use of a common system or the need for individual air exhaust systems and duct isolation valves to facilitate gaseous decontamination of all or part of the facility.

2 The airlock is provided to ensure the maintenance of the negative pressure within the facility and prevent airflow between the facility and areas external to the facility. It should not be used for any work, nor should it contain any equipment, washing facilities (apart from the shower where it is used as a shower airlock) or PPE worn in the facility.

3 Consideration should be given to the provision of a gaseous decontamination chamber for facilities that require removal and installation of equipment that cannot be steam sterilized. This facilitates removal of such items without the need to decontaminate the complete facility.

4 Depending on the need, removable panels to allow for entry and exit of large items of equipment should be considered. These should be readily resealable to an airtight condition following use.

5 Building regulations may require alternative egress in certain facility configurations. These are required to be accessible and easily usable and should not compromise facility seal integrity. Lockable doors should permit emergency egress in accordance with building regulations.

(b) Means shall be provided for minimizing disturbance of pressure differentials by reducing the likelihood of both airlock doors being opened at the same time.

NOTE: Examples of suitable mechanisms include entry and egress ‘traffic light’ alarm systems, door interlock control systems or viewing panels if suitable.

(c) The allocation of task zones and layout within the facility shall promote the movement of ventilation air from near the entry and towards the more contaminated zones such as biological safety cabinets and steam sterilizer loading trolleys.

(d) As much fresh water handling and pre- treatment equipment, valve and control equipment as possible shall be located outside the facility boundary to minimize the need for service personnel to enter the facility.

(e) The facility shall have a high level of physical security including control of access (e.g. electronic access card) and shall not open onto a public thoroughfare.

(f) Doors, apart from those to areas used for showering and changing, shall contain viewing panels to minimize entry and exit incidents.

(g) Adequate arrangements for observation of occupants shall be provided.

NOTE: Examples of suitable arrangements are the viewing panels in doors required in Item (f) if they allow adequate viewing of occupants, viewing panels in walls or electronic visual monitoring facilities (e.g. viewing cameras or closed circuit television).

(h) Two independent communications systems shall be provided. Two way communication shall be able to be conducted on at least one system, the other shall allow a person in the facility to draw the attention of persons outside.

(i) The facility shall include provisions to change aquatic organism cages, bedding, feed and water without compromising microbiological containment.

(j) A pressure steam sterilizer for decontamination of wastes shall be provided in the facility. The pressure steam sterilizer shall not be located in the airlock.

NOTES:

1 A double-ended type of pressure steam sterilizer is preferred, positioned through the barrier wall of the facility (see Note 2) and allowing adequate access for loading and unloading of the steam sterilizer and for maintenance of the associated plant from outside the facility.

2 The barrier wall of the PC3 facility should be sealed airtight to a purpose-constructed chamber barrier flange with all penetrations sealed airtight such that the steam sterilizer installation maintains the integrity of the seal of the containment facility.

3 See also Clause 10.6.8.

(k) All liquid effluent from the facility shall be decontaminated to ensure that no viable microorganisms leave the facility. For aquatic organisms, liquid effluent treatment is a complex process. The design and installation of these systems require the use of suitably qualified professionals.

The liquids to be decontaminated include all liquids potentially exposed to aquatic microorganisms, equipment discharges, tundishes, floor wastes, facility sink effluent, hand basin effluent and shower effluent.

See also Section 12.

(l) Provision shall be made for decontamination of the facility. The facility shall be constructed to contain any aerosols or gases used for decontamination. The surface finishes in the facility shall be compatible with the decontamination agents used.

NOTE: The design of the facility should avoid inaccessible spaces. See Appendix H for recommendations on design for airtightness and periodical retesting.

(m) All room penetrations shall be sealed to ensure they are airtight.
NOTE: See Appendix H.

6.6.3 Ventilation

A ventilation system that establishes a negative pressure in the facility shall be provided so that there is a directional airflow into the working area. Where facilities have supply air systems, the supply air and exhaust air systems shall be interlocked, to ensure inward airflow at all times. The proper directional airflow into the facility shall be verified by airflow tests. The facility (including the airlock) shall be structurally designed to take account of the operation under negative pressures.

Failure of a single component, such as an exhaust fan or a supply fan, can result in extremely high positive or negative pressures in the facility. Alarms and failure mode operations of ventilation systems shall address this risk to ensure that interlocks operate rapidly to stop systems. The facility shall be constructed to withstand, without cracking or deterioration, the maximum positive and negative pressures that can be generated until failure mode safeguards operate. Automatic and manual failure mode sequences shall be independent of any automated control system that may, itself, be the primary cause of a failure situation.

All air that leaves the facility shall be exhausted in accordance with the requirements of this Clause.

Air may be recirculated within each facility. If air is recirculated, this shall be achieved utilizing internally-mounted air conditioning equipment such as fan coil units and split system air conditioning units. Any internally-mounted equipment shall be provided with removable panels as required to ensure the complete penetration of gas or vapour during room decontamination.
NOTES:
1 Ventilation equipment should be located to ensure a flow of incoming air from the vicinity of the entry door towards the highest risk microbiological work areas.
2 The quantity of outside air supplied to the facility should comply with relevant health quality standards or regulations (see AS 1668.2) and should be adequate to dilute airborne contaminants.

Ventilation equipment and outlets shall be located to minimize the disturbance to the open faces of Class I and Class II biological safety cabinets.

The facility ventilation shall incorporate the following features:

(a) The facility shall be maintained at an air pressure of at least 50 Pa below the pressure of areas outside the PC3 containment barrier when both doors of the airlock are closed. When either door is open, the facility pressure shall remain at least 25 Pa below that of areas outside the PC3 containment barrier. The 0 Pa reference pressure shall be measured to minimize the effects of fluctuations due to wind and other building ventilation systems.
NOTE: Additional precautions should be considered when one or more surfaces are external and exposed to fluctuations of pressure due to wind effect. Suitable precautions include the use of an interstitial space that can be maintained at the zero reference pressure and the use of solid walls such as concrete with minimal joints, which are unlikely to be perforated or leak.

(b) The pressure differential shall be achieved by means of an independent room exhaust fan discharging to the open air through a filter.

NOTE: A variable speed drive on the exhaust fan is preferred to facilitate room pressure control adjustments.

(c) Supply or replacement air to the room shall be filtered using Type 1 Class A or Class B filters complying with AS 1324.1 and having a minimum arrestance efficiency of 90% when tested in accordance with AS 1324.2 with Test Dust No. 4. If adjustable dampers are provided in transfer apertures to assist in setting up the reduced room pressure, these devices shall not be mounted in the door.

(d) Exhaust air shall be filtered and discharged to the outside atmosphere in such a manner that it is dispersed away from occupied buildings and outside air intakes.

(e) Filtration of exhaust air to a HEPA standard is not normally required for PC3 aquatic microorganisms. For normal applications, the exhaust filter shall be Type 1 Class A or Class B filters complying with AS 1324.1 and having a minimum arrestance efficiency of 90% when tested in accordance with AS 1324.2 with Test Dust No. 4

(f) Where there is a special risk associated with the work of the aquatic animal facility due to the possibility of hazardous aerosol generation, the exhaust filter shall be a HEPA type as specified in Clause 10.9.1. An exhaust prefilter of the same standard as the supply filter shall be provided and mounted upstream of the HEPA filter. Filters shall be selected to meet the expected quantity and type of aquatic organism debris.

NOTES:

1 Prefilters should be located within aquatic organism rooms for ease of replacement.

2 Ventilation rates should ensure an acceptable atmosphere quality for aquatic organism welfare. If air cooling is required, this should be achieved through cooling coils mounted external to the occupied rooms.

3. Note that ,if HEPA exhaust filtration is omitted, the aquatic organism PC3 facility will not be suitable for other types of work at PC3 level.

(g) Where required in (f) above, the HEPA filter shall be installed, housed and maintained as specified in Clause 10.9.2.

(h) For each ventilation system a differential pressure gauge shall be visible and readable from immediately outside the facility.

(i) Where HEPA filtration is required by (f) above, any tubing that forms part of the facility pressure sensing and control equipment shall be fitted with a 0.2 µm hydrophobic membrane filter (such as a miniature disk filter), located as close as possible to the PC3 containment barrier. Filters and tubing shall be protected against mechanical damage.

(j) An emergency stop button shall be provided for each ventilation system. This shall be located outside the facility, adjacent to the exit.

The emergency stop button shall operate independently of the main ventilation control and main facility pressure control system such that emergency isolation of the ventilation can be implemented in event of central control system malfunction.

(k) An audible emergency alarm shall be provided within the facility to indicate a loss of negative pressure and a visible alarm shall be provided outside the facility to indicate the same.

NOTE: The selection of alarm type and the provision of mute switches should be considered to address aquatic organism welfare concerns associated with sudden or prolonged noises.

(l) Annual testing by competent persons shall include:

(A) Testing of the pressure differentials in accordance with AS 1807.10 to ensure compliance with the requirements in Item (a).

(B) Integrity testing of any installed HEPA filters in accordance with AS 1807.6 or AS 1807.7, as applicable.

(C) Checking that the control system is operating correctly and verifying alarms are set to operate in accordance with Item (j) on a loss of room pressure.

(D) Calibration of pressure control and indicating devices.

(E) A report of the testing in Items (A) to (D) and of any maintenance conducted shall be provided to the appropriate person for the facility.

(m) Exhaust air from Class III biological safety cabinets shall be discharged through the building exhaust system through direct ducting or a capture hood as described in AS/NZS 2647. It shall not be recirculated through the facility.

NOTES:

1 Care should be exercised with direct ducting with respect to pressure fluctuations. Capture hoods may be inappropriate for gases and vapours.

2 The exhaust air from Class I or Class II biological safety cabinets may be discharged into the facility or through the building exhaust system in accordance with AS/NZS 2647.

6.6.4 Access to services

Access to voids surrounding the immediate perimeter of the facility and to the ventilation equipment that serves the facility shall be restricted to authorized persons. Items of equipment, ducts and access panels to contained sections of the ventilation system shall be marked with biohazard labels to minimize the risk of accidental exposures to air or to contaminated surfaces. The installation of services shall ensure proper access to equipment such as HEPA filters for maintenance and testing personnel and their equipment.

6.6.5 Containment equipment

In addition to equipment specified for PC2 (Clause 6.5.4), a Class III Biological safety cabinet shall be provided where appropriate (see Clause 10.7.2).

6.6.6 Work practices

In addition to requirements for aquatic organism PC1 and aquatic organism PC2 facilities (see Clauses 6.4 and 6.5), the following work practices shall apply:

(a) All containment features of the completed facility shall be tested, commissioned and the results documented, before use.

(b) The facility shall be inspected at least annually by the BC or operator to ensure that its containment requirements comply with Clauses 6.6.2(a) and 6.6.3 by reviewing records including any HEPA filter integrity test reports and room pressure readings.

(c) The facility management shall establish policies and written procedures whereby only persons who have been advised of the biohazard, and who meet any medical requirements, shall enter the facility. See Clause 2.6.

(d) An effective emergency evacuation plan shall be devised and information on the plan shall be available to all facility staff and local emergency services.

(e) All facility staff shall have specific training in handling pathogenic aquatic organisms and in the use of safety equipment and controls. The staff shall be supervised by senior scientists who are experienced in working with pathogenic microorganisms.

(f) Only trained people authorized by the BC or operator shall enter the aquatic organism facility, and then only after they have been advised of the hazard and met all specific requirements, such as immunization.

(g) The facility door shall be locked when the room is unoccupied by personnel. See also Clause 6.6.2(a).

(h) Outer clothing and personal effects shall not be taken into the facility.

(i) No one shall enter the facility for cleaning, servicing of equipment, repairs or other activities before the relevant, potentially contaminated surfaces have been decontaminated and authorization has been obtained from the facility supervisor or the safety officer. Dedicated cleaning equipment shall be stored within the facility.

(j) If the facility does not form the primary containment measure, and there is the potential for an aerosol hazard, all aquatic organism handling procedures with potentially infectious materials shall be done either in a Class I or II biological safety cabinet (or the equivalent).
NOTE: The provision of an uninterruptible power supply should be considered for BSCs.

(k) All equipment used in the facility shall be decontaminated prior to maintenance, service or removal.

(l) If a double-ended pressure steam sterilizer is installed across the barrier, it shall be decontaminated after each exposure to the facility environment.

(m) Microbiological wastes, aquatic organism excrement, liquid effluents, organism bedding, organism cages and aquatic organism carcasses shall be decontaminated in a pressure steam sterilizer. Waste material shall then be disposed of in accordance with Clause 12.2.

(n) Live aquatic organisms or viable biological material shall only be taken to an equivalent or higher level of containment.

(o) Viable biological materials to be removed from the facility shall be transferred to a non-breakable, sealed primary container, the external surface of which is decontaminated before enclosure in a non-breakable, sealed secondary container.

(p) Protective clothing shall be removed in a predetermined appropriate order before leaving the aquatic organism facility.

(q) Protective clothing shall not be worn outside the facility and shall be decontaminated by pressure steam sterilization prior to laundering or disposal.
NOTE: In most circumstances, the appropriate removal procedure is removing the gloves then decontaminating hands followed by removal of eye protection, gown and respiratory protection, taking care not to touch potentially contaminated parts of PPE when doing so, then decontaminating hands again.

(r) Where the aquatic organism facility forms the primary containment measure, and there is a risk of infectious material adhering to personnel, a full body shower shall be taken upon exiting the facility.

(s) Measures shall be taken to ensure no microbiological contamination is removed from the facility on footwear.

NOTE: Suitable measures include the use of dedicated facility footwear, the use of overshoes or a combination of these measures.

(t) In the event of a power failure, entry to the facility shall be restricted until services have been restored.

6.6.7 Health monitoring

See Clause 2.6.

6.7 REQUIREMENTS FOR AQUATIC ORGANISM PC4 FACILITIES

6.7.1 General

An aquatic organism PC4 facility is suitable for work with infectious microorganisms in Risk Group 4 and incorporates all equipment and practices for aquatic organism PC1 (Clause 6.4), aquatic organism PC2 (Clause 6.5) and aquatic organism PC3 (Clause 6.6); however, additional requirements on conditions of access and egress, safety equipment and staff training apply.

An aquatic organism PC4 facility may be one of two types—

(a) a facility where small aquatic organisms are kept in individually ventilated isolators or Class III biological safety cabinets in the aquatic organism PC4 facility; or

(b) a facility set up for the use of fully-encapsulated, positive pressure personnel suits ventilated by a life-support system.

When pathogenic microorganisms of Risk Group 3 or 4 are being used in association with small aquatic organisms, primary containment measures such as BSCs or individually ventilated isolators fitted with HEPA exhaust filters shall be used where possible. Where primary aquatic organism containment measures cannot be used, the facility forms the primary containment measure.

NOTE: The design of an aquatic organism PC4 facility is complex and those planning its construction should seek specialized advice. See also Appendix G for examples of recommended layouts for PC4 facilities showing the design principles involved and Appendix H for airtightness considerations.

6.7.2 Alternative locations for aquatic organism work

Aquatic organisms may be kept in PC4 facilities in the following ways:

(a) *PC4 laboratories* Small aquatic organisms in appropriate containers may be kept in Class I or Class II biological safety cabinets in a PC4 laboratory where staff wear one-piece positive pressure personnel suits ventilated by a life-support system.

NOTE: The exhaust air from Class I or Class II biological safety cabinets may be discharged into the facility or through the building exhaust system in accordance with AS/NZS 2647.

In addition to the work practices used in PC4 laboratories (see Clause 5.5.5), the following work practices apply:

(i) All staff shall have specific training in handling Risk Group 4 aquatic organisms in the relevant aquatic organism species and in the use of safety equipment and operation of the facility.

(ii) Microbiological wastes, aquatic organism excrement, liquid effluents, shower effluents, aquatic organism detritus, aquatic organism containers and aquatic organism carcasses shall be decontaminated, preferably by pressure steam sterilization, before disposal in accordance with Clause 12.2.

(b) *Primary containment devices in Aquatic organism PC4 facilities* Aquatic organisms may be kept in individually ventilated isolators or Class III biological safety cabinets in an Aquatic organism PC4 facility if the exhaust air from the Class III biological safety cabinets is discharged through the building exhaust system through a capture hood as described in AS/NZS 2647. The exhaust air shall not be recirculated. In such aquatic organism PC4 facilities staff do not need to wear positive pressure personnel suits.

In addition to the requirements in Clauses 6.7.3 to 6.7.7 (other than those in Clauses 6.7.3.2, 6.7.4.2 and 6.7.6.2), procedures for handling aquatic organisms in individually ventilated isolators or Class III biological safety cabinets fitted with an exhaust HEPA filter shall be documented following a risk assessment of the hazards involved with the infectious agent and aquatic organism species.

(c) *Aquatic organism PC4 facilities where the room is the primary containment measure* In such aquatic organism PC4 facilities staff wear one-piece positive pressure personnel suits ventilated by a life-support system and additional requirements specified in Clauses 6.7.3 to 6.7.7 apply, including those in Clauses 6.7.3.2, 6.7.4.2 and 6.7.6.2.

6.7.3 Construction

6.7.3.1 *General*

In addition to the design features and facilities specified for aquatic organism PC1, aquatic organism PC2 and aquatic organism PC3, the following shall be provided:

(a) The facility shall be housed in a separate building or shall form an isolated part of a building. Full access to all exterior surfaces of the contained structure and service penetrations shall be provided to facilitate periodic integrity testing. The perimeter of the facility shall be protected against adverse pressure fluctuations due to wind or other external factors. The facility negative pressure shall be maintained across the floor, wall and ceiling boundaries at all times.

NOTE: Recommendations on acceptable room airtightness are given in Appendix H.

(b) Any transparent sections shall be constructed of impact-resistant materials.

(c) An outer and inner change room, separated by a shower airlock with self-closing doors, shall be provided for personnel entering and leaving the facility. The outer door of the facility shall be lockable.

NOTE: A security card access procedure, with additional numerical pad or biometric access control, is preferred as a means of entry.

The outer shower door shall form the facility containment boundary for decontamination purposes.

The four doors of each entry/exit path shall raise an alarm if left open.

An entry and egress 'traffic light' alarm system or door interlock control system shall be provided to prevent the simultaneous opening of the doors on each side of the shower. An audible alarm shall operate if both doors are simultaneously open at any time.

NOTES:

1 The use of pneumatically sealed doors should be considered on both sides of the shower.

2 A timer should be provided to permit personnel to shower for a defined period as part of the exit procedure.

3 Privacy for changing and showering may require door access features and interlocks or alarms additional to the above biocontainment requirements.

4 The use of interlocks requires the provision of manual overrides in case of emergencies.

5 The inner change room may provide the functions of the anteroom as set out in Clause 6.5.2(p).

(d) Walls, floors and ceilings of the facility shall be constructed in such a manner as to form a sealed internal shell which facilitates gaseous decontamination. The internal surfaces of the shell shall be resistant to liquids and chemicals used in the facility and shall facilitate easy cleaning and decontamination. All apertures in the structures and surfaces shall be sealed to prevent vermin or insects from entering the area. Glazing in windows shall be of laminated security glass selected to withstand the maximum pressure differential imposed during all operating conditions, including all possible failure modes, and during testing.

(e) A double-ended pressure steam sterilizer shall be provided to decontaminate materials from the facility and from the inner clothing change room. The outer sterilizer door shall open to the area external to the facility, and shall be sealed to the containment perimeter of the facility. The inner door shall automatically interlock with the outer door in such a manner that the outer door can be opened only after the sterilization cycle has been completed. The sterilizer shall comply with the requirements of Clause 10.6.

(f) A pass-through dunk tank, decontamination chamber or equivalent decontamination equipment shall be provided, so that materials and equipment that cannot be decontaminated in the pressure steam sterilizer can be rendered safe for removal from the facility.

(g) A suitable decontamination system shall be provided for handling all effluents, including those from any showering facility, in accordance with Section 12.

(h) An automatic changeover emergency power source, emergency lighting and communication systems shall be provided. The emergency power source shall be adequate to operate the ventilation systems, BSCs, room access and shower controls. An uninterruptible power supply shall be provided to ensure uninterrupted operation of the shower controls and ventilation control system.

6.7.3.2 *Positive pressure suit area*

For certain requirements, a specially designed suit area may be provided within the facility. Personnel who enter this area shall wear a one-piece positive pressure suit that is ventilated by a life support system. If provided, positive pressure suit areas shall comply with the following additional requirements:

(a) Entry shall be via an outer change room that leads to an airlock fitted with a personal body shower then into an anteroom leading to a second airlock fitted with a chemical disinfectant shower provided to decontaminate the surface of the suit before the worker leaves the area.

(b) A breathing quality air supply for connection to the positive pressure suit shall be provided in the anteroom, chemical shower and work area. Quick-connect fittings shall be provided on the suit and the air supply lines to ensure prompt disconnect and reconnect when moving between anteroom, chemical shower and work areas.

(c) All penetrations into the internal shell of the suit area shall be sealed. (d) An alarm and emergency back-up breathing air system shall be provided.

6.7.4 Ventilation

6.7.4.1 *General*

The facility ventilation shall comply with the following:

(a) A separate supply and exhaust, non-recirculating air ventilation system shall be provided. The system shall maintain such pressure differentials and directional airflow to ensure airflows toward areas of highest potential risk within the facility. There shall be a differential pressure of at least 25 Pa between each area (see Figure G5). The system shall be provided with an alarm to detect malfunction. The supply and exhaust airflow shall be interlocked to assure inward (or zero) airflow at all times. Differential air pressures between facility zones shall be monitored by use of a differential pressure gauge as specified in Clause 6.6.3(h).

(b) Both supply and exhaust air shall be filtered through HEPA filters as specified in Clause 10.9.1. The HEPA filters shall be installed and housed as specified in Clause 10.9.2. Prefilters to both the supply and exhaust HEPA filters shall be provided as specified in Clause 6.6.3(c) and (e).

The supply air HEPA filter shall prevent the outflow of contaminated air if air pressures become imbalanced within the facility.

(c) The ventilation control system shall raise an audible alarm within the facility and at an attended location when room differential air pressures depart from set points by more than 15 Pa for a period of greater than 2 min.

(d) Annual testing by a competent person shall include:

(i) Testing of the pressure differentials in accordance with AS 1807.10 to ensure compliance with (a).

(ii) Integrity testing of all installed HEPA filters in accordance with AS 1807.6 or AS 1807.7, as applicable.

(iii) Checking that the control system is operating correctly and verifying alarms are set to operate in accordance with Item (c).

(iv) A report of the testing in Items (i) to (iii) and of any maintenance conducted shall be provided to the appropriate person for the facility.

6.7.4.2 *Positive pressure suit area*

In addition to the requirements in Clause 6.7.4.1, the following ventilation system features shall be provided for a positive pressure suit area:

(a) The exhaust air shall be filtered through two HEPA filters installed in series.

(b) Duplicate ventilation equipment shall be provided to automatically re-establish facility ventilation and pressure conditions in event of equipment failure. Controls and equipment operation shall prevent a positive pressure occurring within the facility at all times, including the failure of an exhaust fan.

(c) The air pressure within the experimental area shall be lower than that of the chemical shower airlock which, in turn, shall be lower than the adjacent entry/exit and non-suit areas.
NOTE: A 25 Pa differential for each airlock is recommended.

6.7.5 Containment equipment

For work with agents of Risk Group 4, either of the following shall be provided:

(a) A Class I or Class II biological safety cabinet (see Clause 6.7.6.1(h)) where the facility forms the primary containment measure.

(b) A Class III biological safety cabinet (see Clause 10.7).

6.7.6 Work practices

6.7.6.1 *General*

In addition to the work practices specified for aquatic organism Physical Containment Levels 1 (Clause 6.4.3), 2 (Clause 6.5.4) and 3 (Clause 6.6.5), the following practices shall be observed:

(a) All staff shall be trained in the specific working aspects of the facility, including the safety equipment, the operation of the facility and containment and clean-up of infectious spills. Staff shall use the safety equipment provided. Staff shall receive specific training, with written instructions and information in the handling of the relevant microorganisms in the relevant aquatic organism species.

(b) Staff shall be supervised by senior scientists who are trained and experienced in working with the relevant microorganisms and aquatic organism species. A facility operations manual shall be prepared.

(c) Access shall be controlled to allow entry by authorized staff only. A list of contact names and phone numbers shall be provided outside the entry point

(d) Personnel shall enter and leave the facility through the clothing change and shower rooms, except in cases of emergency, where alternative exits may be used.

(e) All requisite facility clothing, including footwear, shall be provided for use by personnel in the facility

(f) On entering the facility, personnel shall don complete facility clothing, including shoes. All street clothing, including underwear, shall be removed and retained in the outer clothing change room.

(g) Before leaving the facility, a full body shower shall be taken. Personnel shall remove their facility clothing and store or discard it in the inner change room before showering.

(h) Personnel entering or leaving the facility shall indicate, either manually or electronically, the time of each exit and entry.

(i) All procedures within the facility involving agents assigned to Risk Group 4 shall be conducted in Class III biological safety cabinets in accordance with Clause 6.7.2(b) or alternatively in Class I or Class II biological safety cabinets used in conjunction with one-piece positive pressure personnel suits ventilated by a life-support system in accordance with Clause 6.7.2(c).

(j) Prior to disposal, all facility effluents, including those from the shower, shall be decontaminated by either heat or chemical treatment in accordance with Section 12.

(k) The double-ended pressure steam sterilizer and decontamination chamber opening across the barrier shall be decontaminated after each exposure to the facility environment.

(l) Unless working with one-piece positive pressure personnel suits ventilated by a life- support system, viable biological materials to be removed from Class III BSCs shall be transferred to a non-breakable, sealed primary container, the external surface of which is decontaminated before enclosure in a non-breakable, sealed secondary container. A primary container holding viable or intact biological materials shall be opened only in another Class III BSC in the PC4 facility or another PC4 facility (see Item (l)).

NOTE: Containers may be opened in non-PC4 facilities only if the biological material has been rendered non-infectious or non-toxic, and the space in the primary and secondary containers has been decontaminated.

(m) Viable biological materials to be removed from the containment facility shall be transferred to a non-breakable, sealed primary container, the external surface of which is decontaminated before enclosure in a non-breakable, sealed secondary container. The secondary container shall be removed from the facility through the disinfectant dunk tank or gaseous decontamination chamber or an airlock designed for this purpose.

(n) No other materials shall be removed from the maximum containment facility unless they have been decontaminated. Equipment or materials which might be damaged by high temperatures or steam shall be decontaminated in an airlock or specially designed chamber, by means of sterilizing gas or vapour.

A primary container holding viable or intact biological materials shall be opened only in a Class III BSC in another PC4 facility.

NOTE: Containers may be opened in non-PC4 facilities only if the biological material has been rendered non-infectious or non-toxic, and the space in the primary and secondary containers has been decontaminated.

(o) Risk Group 4 material shall be stored only within the facility.

(p) A risk assessment of the working environment shall be undertaken encompassing all matters influencing the personal safety of staff. Monitoring and emergency procedures shall be prepared and implemented at a level commensurate with the outcome of the risk assessment.

NOTE: The presence of a co-worker either inside the facility or observing the work from outside the facility should be considered.

(q) Checks shall be carried out to ensure monitoring and communication arrangements result in the emergency procedures being initiated in a timely manner and to ensure the procedures are adequate.

6.7.6.2 *Positive pressure suit area*

In addition, the following work practices apply for positive pressure suit areas:

(a) Upon entering the anteroom, the person shall don a positive pressure suit prior to entering the second (chemical shower) airlock.

(b) When exiting the suit area, a chemical disinfectant shower shall be taken to decontaminate the outer surface of the suit. The disinfectant shall be effective against the microorganisms used in the suit area at the concentration at which it is used in the shower taking into account the time and temperature defined for the shower. The suit and clothing shall be removed in the anteroom and a full body shower shall be taken in the shower airlock.

(c) Microbiological wastes, aquatic organism excrement, liquid effluents, shower effluents, aquatic organism bedding, small aquatic organism cages and aquatic organism carcasses shall be decontaminated, preferably by pressure steam sterilization, before disposal in accordance with Clause 12.2. Additionally, if floor drains are present, all effluents shall be decontaminated in accordance with Clause 12.2 before discharge.

6.7.7 Health monitoring

See Clause 2.6.

REFERENCE ADDITIONS:

1.34 Australian Code of Practice for the care and use of animals for scientific purposes, 7th edition 2004. Australian Government National Health and Medical Research Council (NHMRC)

1.35 Guidelines to promote the wellbeing of animals used for scientific purposes. Australian Government National Health and Medical Research Council (NHMRC)

1.36 Guide for the care and use of laboratory animals, 8th edition 2011. US National Research Council, the National Academic Press, pp 77-88 on aquatic species

1.37 United States Department of Agriculture, Agricultural Research Services, National Agricultural Library, Animal Welfare Center. Refer in particular to “Fish Welfare” and “Care and Use of Molluscs” information resources.

Add clause in app E for backflow prevention for aquatics