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# CONTAINMENT STANDARDS FOR VETERINARY FACILITIES



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**CONTAINMENT STANDARDS  
FOR  
VETERINARY FACILITIES**

Food Production & Inspection Branch  
Agriculture & Agri-Food Canada

**First Edition  
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# CONTAINMENT STANDARDS FOR VETERINARY FACILITIES

## 1. INTRODUCTION

The Medical Research Council of Canada and Health Canada (MRC/HC) *Laboratory Biosafety Guidelines* published in 1990 and revised by HC in 1996, has been the recognized biological safety standard for the construction and operation of containment facilities. The MRC/HC guidelines provides guidance for those who build, design, operate or work in laboratories in which human pathogens are handled. The guidelines do not address work with strictly animal pathogens nor work with large animals in containment.

Containment requirements for veterinary facilities handling livestock or poultry diseases are unique. With the exception of zoonotic agents, these organisms are not known to cause disease in humans and are therefore classified on a lower level with respect to risk of infection to laboratory personnel. However, for non-indigenous animal pathogens, higher levels of containment are of vital importance to prevent their release into the environment with potential serious negative economic impact.

Work with large animals in containment poses a variety of special requirements. Animal and post-mortem rooms must be constructed to contain large numbers of microorganisms which may be present. The rooms must withstand a variety of stresses including physical impact, noise, temperature and cleaning. Operational protocols for personnel and animal handlers entering and exiting an infected animal area may have more stringent requirements than protocols for entering and exiting a containment laboratory.

The Health of Animals Act, 1990, and its Regulations gives Agriculture and Agri-Food Canada (AAFC) the legislative authority to control the use of pathogens which may cause disease in animals. AAFC will also establish for animal pathogens, the conditions under which they will be maintained and work will be carried out. It is on this basis that the Animal and Plant Health Directorate (APHD) of AAFC has taken a lead role in the development of standards for veterinary containment facilities.

The intended scope of this standard is to outline minimum design and operational requirements for APHD laboratories and animal containment facilities. In addition to having a direct impact on the APHD, this document provides guidance on the design and operation of veterinary containment facilities in general.



## Introduction

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The APHD *Containment Standards for Veterinary Facilities* were developed by an Agriculture and Agri-Food Canada containment team consisting of the following individuals:

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## 2. ANIMAL PATHOGEN CONTAINMENT LEVELS

Laboratories and animal facilities handling pathogens of veterinary significance must be constructed and operated to appropriate containment levels and standards. The level required depends not only on the risk to human health but on a variety of other factors including the prevention of cross-contamination and the prevention of escape of animal pathogens into the environment where they might infect the indigenous animal population.

The containment levels required for work with strictly animal pathogens are listed in a database maintained by the APHD. This is a dynamic listing which is continuously amended to include emerging pathogens. For each animal pathogen under the control of AAFC, APHD must be consulted for the level of containment needed (contact: Animal Health Division, 59 Camelot Dr., Nepean, ON K1A 0Y9, (613) 952-8000). This includes material of animal origin which may contain potential pathogens. The operational practices and physical design requirements for the animal pathogen (AP) containment levels are outlined in this document.

All APHD laboratories and animal facilities must comply with the minimum design and operational requirements listed in this standard. It should be noted that for each AP containment level described herein, the physical requirements meet or exceed the intent of the corresponding containment level listed in the *HC Laboratory Biosafety Guidelines*. However, some operational practices may differ where the animal pathogen does not represent a risk to human health (e.g. requirement for working in biological safety cabinet, requirement for use of positive-pressure ventilated suits).

Generally, work with endemic animal pathogens causing mild disease and of limited veterinary importance can be safely carried out in AP containment level 2 facilities. Facilities meeting level 2 criteria with specific level 3 enhancements (e.g. liquid effluent treatment) may be appropriate for resistant stages of certain animal parasites requiring an intermediate host. Pathogens causing serious livestock or poultry disease and spreading readily by the aerosol route require a higher level of containment (i.e. AP containment level 3 or 4, depending on the severity of disease).

Where the level of containment required is not specified in the APHD database, an assessment by the Chief, Laboratory Safety and principal investigator will establish specific containment requirements and operational protocols that must be followed.

Factors used to determine the required containment level include:

- infectious dose required to cause an infection
- route of infection (via aerosol transmission, injection, ingestion, absorption, invasion of mucous membranes or abraded skin)
- pathogenicity and virulence of the microorganism
- host range
- morbidity and mortality rates for the individual disease
- vector necessary for transmission and disease
- quantity and concentration of the agent (i.e. in vitro, in vivo)
- microorganism excreted in feces, urine, and/or exhaled
- inherent biological decay rate (specifically how long will the agent survive in the environment outside of a susceptible host or culture medium)
- endemicity of the microorganism
- availability of effective vaccines, prophylactics and therapeutic treatment

### 3. PHYSICAL REQUIREMENTS

The physical requirements for animal pathogen (AP) containment levels 2, 3 and 4 are described below. The laboratory facilities described meet or exceed the physical requirements set out in the HC guidelines and are appropriate for work with zoonotic agents in addition to strictly animal pathogens. The sections on animal facilities present design requirements unique to the handling of small and large animals in containment.

#### 3.1 LABORATORIES

A "laboratory zone" is defined as an laboratory area of equal containment level which may have multiple rooms and functions. The containment perimeter/barrier of the laboratory zone is continuous and non-intersecting (i.e. the zone is serviced by a single entry/exit).

LOCATION & ACCESS	
AP CONTAINMENT LEVEL	REQUIREMENT
2,3,4	dedicated and controlled access to be limited to authorized personnel into the laboratory zone
3,4	access to be controlled within the laboratory zone and into each level 3 and level 4 laboratory
2,3,4	laboratory room doors to have appropriate signage (i.e. hazard identification, name and phone number of contact person, entry requirements)
3	entry to laboratory zone to be provided via ventilated airlock (i.e. ventilation to be provided through leaky doors and/or HVAC systems) with, preferably, interlocking doors, or with a warning light or audible alarm to prevent both doors from opening at the same time (i.e. to prevent migration of air from higher to lower containment zones)
4	entry to laboratory zone to be provided via ventilated airlock with interlocking tightly sealed doors (e.g. inflatable or compression seal to provide room integrity in accordance with Section 7) to prevent migration of air from higher to lower containment zones (i.e. to ensure flow of air from low hazardous areas to high hazardous areas)



LOCATION & ACCESS - CONTINUED	
AP CONTAINMENT LEVEL	REQUIREMENT
3,4	entry to laboratory zone to be provided with clothing change area designed to separate personal clothing from laboratory clothing dedicated to that zone (i.e. "clean" change area separated from "dirty" change area)
3	entry to laboratory zone to be provided with a shower on the containment barrier (i.e. between "dirty" and "clean" change areas)
4	entry to laboratory zone to be provided with a suit change area, a chemical shower on the containment barrier (i.e. between the laboratory and suit change area) and water shower on exit from the zone (i.e. between "dirty" and "clean" change areas)
3,4	controlled entrance/exit doors to have emergency manual overrides
3,4	the containment barrier support systems (e.g. HEPA filters, effluent sterilization system) to be located as close as possible to the laboratory zone
2	office areas (i.e. a separated room providing a support lab function normally requiring no containment principles) to be located outside of laboratory work areas but can be located within the level 2 laboratory zone
3,4	office areas (i.e. a separated room providing a support lab function normally requiring no containment principles) to be located outside of laboratory zone
2,3,4	dedicated clerical work stations permitted within the laboratory work areas away from hazardous materials

**Note:** For new level 3 and level 4 construction it is recommended that containment laboratories be located away from exterior envelope walls. Most buildings are subjected to winds and the resulting positive or negative pressures on the building may be greater than the typical operating pressures of the containment laboratories. An interior design creates an environmental buffer to wind effects on exterior envelope walls. Section 4.1.4, *Live Loads Due to Wind*, of the 1995 National Building Code of Canada outlines the methodology for calculating external pressure or suction due to wind on a surface of a building.

Physical Requirements - Laboratories

<b>SURFACE (i.e. FLOORS, WALLS, CEILINGS, SEALANTS) FINISHES</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3,4	interior coatings to be gas and chemical resistant in accordance with laboratory function (e.g. will withstand chemical disinfectants, chemical spills, fumigation)
3,4	interior surfaces to be continuous (note: for level 3 floors, flooring with welded seams, is acceptable)
3,4	interior surfaces to minimize movement of gases and liquids through perimeter membrane (i.e. to provide room integrity in accordance with pressure decay testing specified in Section 7)
3,4	interior surfaces to provide impact resistance in accordance with laboratory function
3,4	interior surfaces to be compatible with adjacent and overlapping materials (i.e. to maintain adhesion and a continuous perimeter)
3,4	continuity of seal to be maintained between the floor and wall (a cove floor finish 15cm up the wall is recommended)
2,3,4	floors to be slip-resistant
3,4	doors and frames to be non-absorptive and have solid finishes (i.e. wood is not acceptable); hollow doors must be sealed

**Note:** For new level 2 construction, the requirements listed above for laboratory surface finishes should be taken into consideration.

<b>CONTAINMENT PERIMETER</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3,4	all mechanical, electrical and service piping penetrations to be sealed at containment perimeter
2,3,4	containment perimeter (e.g. doors and windows) to be kept closed in order to provide required containment of air systems
2,3,4	window design to be integrated with the heating/ventilation/air-conditioning (HVAC) system to avoid condensation, wetting and/or frost build-up
2,3,4	windows to provide required level of security

Physical Requirements - Laboratories

<b>CONTAINMENT PERIMETER - CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2,3,4	door openings to allow passage of required equipment (i.e. may be greater than standard width and height in accordance with equipment size)
2	autoclave to be located within laboratory zone
3,4	dedicated double-door barrier autoclave to be located and sealed on containment barrier of laboratory zone; body of autoclave to be located for ease of maintenance, preferably outside containment zone
3	barrier autoclave to be equipped with, preferably, interlocking doors, or with a warning light or audible alarms to prevent both doors from opening at the same time
4	barrier autoclave to be equipped with interlocking doors
2,3,4	autoclave to be equipped with a cycle log recorder (i.e. to record time, temperature, and pressure)
3,4	for materials that cannot be autoclaved (e.g. heat sensitive equipment, samples, film) other proven technologies for sterilization (e.g. incineration, chemical or gas sterilization, rendering, irradiation) to be provided at containment barrier
2,3,4	laboratory zone to be proofed against entry or exit of vermin or insects



Physical Requirements - Laboratories

<b>AIR HANDLING SYSTEM</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2,3,4	Inward directional airflow to be provided; non-recirculated air should be supplied to level 2 laboratories (note: this does not apply to the re-circulation of air through equipment such as biological safety cabinets)
2,3,4	Exhaust from the laboratories to provide a minimum of 10 air changes per hour under normal operations
2,3,4	HVAC air distribution design to minimize dead air spaces within the laboratory; supply and exhaust diffusers to be located to provide convection patterns that ensure airflow away from lab entrance; diffuser selection to provide minimal throw velocities, i.e. < 15 m/m @ 1 m.; supply and exhaust diffusers to be located with biological safety cabinets and fume hood locations taken into consideration
3,4	minimum of 25 Pa difference is recommended across a containment barrier
3,4	pressure monitoring devices are required at the laboratory zone entrance to monitor negative pressure between containment zones
3,4	room static pressure monitoring lines to be provided with filters of at least equal efficiency to HEPA filter
3,4	audible alarms to be provided in the laboratory work area and outside laboratory zone to detect positive pressurization and air handling systems failure
3,4	air supply HVAC system to be independent from adjacent laboratory zones (level 3 supply can be combined with areas of lower containment when provided with a bubble tight damper or HEPA filter after the connection, i.e. downstream from the connection)
3,4	air exhaust HVAC system to be independent from adjacent laboratory zones (level 3 exhaust can be combined with areas of lower containment when provided with a HEPA filter before the connection, i.e. upstream from the connection)
3	air supply to prevent backdraft of contaminated air through air supply duct (e.g. to be HEPA filtered or provided with a bubble tight damper)

Physical Requirements - Laboratories

**AIR HANDLING SYSTEM - CONTINUED**

<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
4	air supply to be HEPA filtered
3	air exhaust to be HEPA filtered
4	air exhaust to be passed through two stages of HEPA filtration
3,4	air supply and exhaust to be equipped with bubble tight dampers to permit gaseous decontamination (can be same bubble tight damper as required for backdraft protection and for isolation of the HEPA filters)
3,4	air supply HVAC system to be controlled (i.e. flow, pressure, electrical) with exhaust HVAC system, and vice versa, to prevent lab positive pressurization
3,4	airflow control devices and duct sensors to be located downstream of the exhaust HEPA filter and upstream of the supply bubble tight damper or HEPA filter
3,4	air supply and exhaust ductwork to be sealed airtight (in accordance with requirements specified in Section 7) between the room perimeter and bubble tight damper
3,4	all air supply and exhaust ductwork that is located outside the containment laboratory requires accessibility
3,4	bubble tight dampers and HEPA filters to be located as close as possible to the containment perimeter

**Note:** Flexibility should be designed into HVAC systems to accommodate for change (i.e. fan/motor capacity should be slightly oversized).

**LABORATORY SERVICES (i.e. WATER, COMPRESSED GASES, ELECTRICITY)**

<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2,3,4	exposed laboratory services piping with stand-offs to allow access for maintenance and cleaning
3,4	water supply control to be located outside laboratory zone

<b>LABORATORY SERVICES (i.e. WATER, COMPRESSED GASES, ELECTRICITY) CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3,4	supply water services to be provided with backflow prevention at the perimeter of laboratory zone (i.e. in addition to premises isolation); selection of backflow prevention device to be in accordance with the Canadian Standards Association (CSA) CAN/CSA-B64.10-94 <i>Manual for the Selection, Installation, Maintenance, and Field Testing of Backflow Prevention Devices (1994)</i>
3,4	compressed gas cylinders (with the exception of fire extinguishers) to be located outside of the laboratory zone
3,4	supply gas (e.g. carbon dioxide, compressed air, natural gas) services to be provided with backflow prevention at the perimeter of laboratory zone
3,4	vacuum services to be provided from within laboratory zone; internal contamination of vacuum pump to be minimized (e.g. HEPA filtration of vacuum line, use of disinfectant traps)
4	for activities involving zoonotic agents, compressed breathing air to be provided to positive-pressure personal protective equipment (i.e. for connection to the air hose of suits), equipped with breathing air compressors and back-up cylinders; air hose connections to be provided in all areas where suits are worn, including chemical shower and suit change room
3,4	handwashing sinks to be provided with "hands-free" capability
2,3	emergency eyewash facilities to be provided in the laboratory area in accordance with laboratory activities and applicable regulations (i.e. American National Standards Institute (ANSI) Z358.1 <i>Emergency Eyewash and Shower Equipment (1990)</i> )
2	emergency shower equipment to be provided in the laboratory area in accordance with laboratory activities and applicable regulations (i.e. ANSI Z358.1 <i>Emergency Eyewash and Shower Equipment</i> )



Physical Requirements - Laboratories

**LABORATORY SERVICES (i.e. WATER, COMPRESSED GASES, ELECTRICITY)  
CONTINUED**

<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3	where it is not possible to limit the quantities of hazardous materials within the laboratory, emergency shower equipment to be provided in the laboratory area in accordance with laboratory activities and applicable regulations (i.e. ANSI Z358.1 <i>Emergency Eyewash and Shower Equipment</i> )
3,4	drainage traps to be provided to required depth in accordance with air pressure differentials (15 cm P-traps are recommended)
3,4	drains and associated piping (including autoclave chamber condensate) to be separated from other laboratory zones (i.e. to go directly to main collector for sanitary sewer or liquid effluent treatment system as appropriate)
3	for non-indigenous agents, drains (including autoclave chamber condensate) and associated piping to be connected to an effluent sterilization system
3	for indigenous agents, drains (including autoclave chamber condensate) and associated piping to be connected to an effluent sterilization system consistent with laboratory activity and local regulations
4	drains (including autoclave chamber condensate) and associated piping to be connected to an effluent sterilization system
3,4	drains connected to effluent sterilization should be sloped towards sterilization system to ensure gravity flow; consideration should be given to the installation of valves to isolate sections for decontamination; piping to be heat and chemical resistant consistent with application; joints should be by thermo/chemical fusible means or welding to ensure integrity of entire system (i.e. in accordance with pressure decay testing specified in Section 7)

Physical Requirements - Laboratories

<b>LABORATORY SERVICES (i.e. WATER, COMPRESSED GASES, ELECTRICITY) CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3	for non-indigenous agents, plumbing vent lines (including effluent sterilization system) to be provided with filter of efficiency equivalent to HEPA
3	for indigenous agents, plumbing vent lines (including effluent sterilization system) to be provided with filter of efficiency equivalent to HEPA consistent with requirement for effluent sterilization system
4	plumbing vent lines (including effluent sterilization system) to be provided with filter of efficiency equivalent to HEPA; two series of filtration are required
3,4	plumbing vent lines to be heat-resistant consistent with application
3	plumbing vent lines can be combined with areas of lower containment when provided with a filter of efficiency equivalent to HEPA before the connection, i.e. upstream from the connection
3,4	supply conduit and wiring to be sealed at the containment barrier (i.e. to provide room integrity in accordance with pressure decay testing specified in Section 7)
2,3,4	light ballasts should be on a separate distribution layout from normal or emergency power to minimize harmonic current problems for sensitive lab equipment; High Intensity Discharge (HID) lamps with lengthy re-strike times should be avoided where there is no alternate quick strike light source
4	light ballasts and starters to be located outside containment perimeter
3,4	power system circuit breakers to be located outside containment perimeter
2,3,4	circuit-breakers and controls to be appropriately labelled
2,3,4	life-safety systems, lighting, biological safety cabinets and other essential equipment to be supported by normal emergency power

Physical Requirements - Laboratories

**LABORATORY SERVICES (i.e. WATER, COMPRESSED GASES, ELECTRICITY)  
CONTINUED**

<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3,4	HVAC systems to be supported by normal emergency power
3,4	communication system to be provided between laboratory area and outside laboratory zone
3,4	system (e.g. fax, computer) to electronically transfer information and data from laboratory area to outside laboratory zone to be provided (note: removing paperwork from the containment laboratory may be carried out after appropriate decontamination, i.e. autoclaving, irradiation, microwaving; such practices are generally not recommended for use on a routine basis)
4	work area to be visually monitored (e.g. closed circuit TV) from outside laboratory zone (e.g. security/biosafety office); observation windows are recommended

### 3.2 SMALL ANIMAL FACILITIES

Animal rooms for small animals (SA) should be designed for ease of cleaning and disinfection and have a minimum of built-in equipment. A small preparation area, storage area and handwashing sink are usually all that is required. The design should also facilitate the use of containment caging systems (e.g. laminar flow cabinets). Support facilities for cage washing, waste disposal, food and bedding storage should also be taken into consideration. Further, the design of the animal facility should permit adjustment of environmental controls to meet the need of the species as specified by the Canadian Council for Animal Care (CCAC) *Guide to the Care and Use of Experimental Animals*, 1993.

An "SA facility zone" is defined as an area of equal containment level. An "animal room" is defined as the room in which the SA is housed. The containment perimeter/barrier of the SA facility zone is continuous and non-intersecting (i.e. the zone is serviced by a single entry/exit).

LOCATION	
AP CONTAINMENT LEVEL	REQUIREMENT
2,3,4	SA facilities to be separated from other laboratory activities
2,3,4	office areas (i.e. a separated room providing a support function normally requiring no containment principles) to be located outside of SA facility zone
2,3,4	clerical work areas for animal handlers permitted within SA facility zone but outside of animal rooms and corridors
2,3,4	feed and bedding storage areas to be provided within the SA facility
2,3,4	clean and dirty cage washing area to be provided within the SA facility (cage washer may be located on containment barrier)
2,3,4	experimental areas (i.e. for necropsy, surgical procedures, etc.) separate from animal rooms should be provided
3,4	experimental areas to be provided with a biological safety cabinet for manipulation of animals infected with zoonotic agents



**Physical Requirements - SA Facilities**

**LOCATION - CONTINUED**

<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
<b>3,4</b>	the containment barrier support systems (e.g. HEPA housings, effluent sterilization system) to be located as close as possible to the SA Facility zone

**Note:** For new level 3 and level 4 construction it is recommended that animal rooms be located away from exterior envelope walls.

**PERSONNEL ACCESS**

<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
<b>2,3,4</b>	dedicated and controlled access to be limited to authorized personnel into the SA facility zone
<b>3,4</b>	access to be controlled within the SA facility zone and into each animal room
<b>2,3,4</b>	entry doors to the SA facility zone to have appropriate signage (i.e. hazard identification, name and phone number of contact person, entry requirements)
<b>2,3,4</b>	where the animal rooms within the SA facility zone have unique hazards, entry doors to each cubicle to have appropriate signage (i.e. hazard identification, personal protective equipment requirements)
<b>2</b>	entry to SA facility zone to be provided via ventilated airlock (i.e. ventilation to be provided through leaky doors and/or HVAC systems)
<b>3</b>	entry to SA facility zone to be provided via ventilated airlock (i.e. ventilation to be provided through leaky doors and/or HVAC systems) with, preferably, interlocking doors, or with a warning light or audible alarm to prevent both doors from opening at the same time
<b>4</b>	entry to SA facility zone to be provided via ventilated airlock with tightly sealed interlocking doors (e.g. inflatable or compression seal to provide room integrity in accordance with Section 7)

Physical Requirements - SA Facilities

<b>PERSONNEL ACCESS - CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2,3,4	entry to SA facility zone to be provided with clothing change area designed to separate personal clothing from SA facility clothing dedicated to that zone (i.e. "clean" change area separated from "dirty" change area)
3	SA facility zone to be provided with a shower on the containment barrier (i.e. between "dirty" and "clean" change areas)
4	SA facility to be provided with a chemical shower on the containment barrier, a suit change area, and water shower on exit from the SA facility zone
3,4	controlled entrance/exit doors to have emergency manual overrides

<b>ANIMAL ACCESS</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2	animal entry to SA facility zone to be provided via ventilated airlock (i.e. ventilation to be provided through leaky doors and/or HVAC systems)
3	animal entry to SA facility zone to be provided via ventilated airlock (i.e. ventilation to be provided through leaky doors and/or HVAC systems) with interlocking doors
4	animal entry to SA facility zone to be provided via ventilated airlock with tightly sealed interlocking doors (e.g. inflatable or compression seal)

<b>SURFACE (i.e. FLOORS, WALLS, CEILINGS, SEALANTS) FINISHES</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2,3,4	interior coatings to be gas and chemical resistant in accordance with SA facility and animal room function (e.g. will withstand cleaning, chemical disinfection, fumigation)
2,3,4	interior surfaces to be continuous

**Physical Requirements - SA Facilities**

<b>SURFACE (i.e. FLOORS, WALLS, CEILINGS, SEALANTS) FINISHES - CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2,3,4	interior surfaces to minimize movement of gases and liquids through perimeter membrane (i.e. animal rooms to provide room integrity in accordance with pressure decay testing specified in Section 7)
2,3,4	interior surfaces to provide impact resistance in accordance with SA facility and animal room function
2,3,4	interior surfaces to be compatible with adjacent and overlapping materials (i.e. to maintain adhesion and a continuous perimeter)
2,3,4	continuity of seal to be maintained between the floor and wall (a cove floor finish 15 cm up the animal room and dirty corridor wall is recommended)
2,3,4	SA facility floors to be slip-resistant and cleanable
2,3,4	animal room and corridor floors to slope towards floor drain (recommended pitch of slope is 2.1cm/m)
2,3,4	doors and frames to be non-absorptive and have solid finishes (i.e. wood is not acceptable); hollow doors must be sealed

<b>CONTAINMENT PERIMETER</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3,4	all mechanical, electrical and service piping penetrations to be sealed at containment perimeter
2,3,4	containment perimeter (e.g. doors) to be kept closed in order to provide required containment of air systems
2,3,4	windows to provide required level of security
2,3,4	windows with direct access between outside the SA facility and animal rooms not to be provided
2	proven technologies for sterilization (e.g. incineration, chemical or gas sterilization, rendering, irradiation, autoclaving) to be provided within SA facility zone

Physical Requirements - SA Facilities

<b>CONTAINMENT PERIMETER - CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3,4	proven technologies for sterilization (e.g. incineration, chemical or gas sterilization, rendering, irradiation, autoclaving) to be provided at containment barrier
2,3,4	SA facility zone to be proofed against entry or exit of vermin or insects

<b>AIR HANDLING SYSTEM</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2,3,4	inward directional airflow to be provided; non-recirculated air should be supplied to level 2 SA Facilities and must be supplied to level 3 and level 4 SA Facilities (note: this does not apply to the re-circulation of air through equipment such as containment caging systems)
2,3,4	exhaust from animal rooms to provide specified number of air changes as required by the CCAC
2,3,4	HVAC air distribution design to minimize dead air spaces within the animal room; supply and exhaust diffusers to be located to provide convection patterns that ensure airflow away from room entrance; diffuser selection to provide minimal throw velocities, i.e. < 15 m/m @ 1 m.
3,4	minimum of 25 Pa difference is recommended across a containment barrier
3,4	pressure monitoring devices to be provided at the SA facility zone entrance to monitor negative pressure between containment barriers; monitors are also recommended at animal room entrance
3,4	room static pressure monitoring lines to be provided with filters of at least equal efficiency to HEPA filter
3,4	audible alarms to be provided in the SA facility work area and outside SA facility zone to detect positive pressurization and air handling systems failure



Physical Requirements - SA Facilities

<b>AIR HANDLING SYSTEM - CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
<b>3,4</b>	air supply and exhaust HVAC system to be independent from adjacent zones (level 3 supply can be combined with areas of lower containment when provided with a bubble tight damper or HEPA filter after the connection, i.e. downstream of the connection; level 3 exhaust can be combined with areas of lower containment when provided with a HEPA filter before the connection, i.e. upstream from the connection)
<b>3</b>	air supply to prevent backdraft of contaminated air through the air supply duct (i.e. to be HEPA filtered or provided with a bubble tight damper)
<b>4</b>	air supply to be HEPA filtered
<b>3</b>	air exhaust to be HEPA filtered
<b>4</b>	air exhaust to be pass through two stage of HEPA filtration
<b>3,4</b>	roughing pre-filters (e.g. 30% and 85% - in accordance with American Society of Heating, Refrigerating and Air-conditioning Engineers (ASHRAE) Standard 52.1-1992 <i>Gravimetric and Dust-spot Procedures for Testing Air-cleaning Devices Used in General Ventilation for Removing Particulate Matter</i> ) to be provided to protect the HEPA filter
<b>3,4</b>	air supply and exhaust to be equipped with bubble tight dampers to permit gaseous decontamination (can be same bubble tight damper as required for backdraft protection and for isolation of the HEPA filters)
<b>3,4</b>	air supply HVAC system to be controlled (i.e. flow, pressure, electrical) with exhaust HVAC system, and vice versa, to prevent animal room positive pressurization
<b>3,4</b>	airflow control devices and duct sensors to be located downstream of the exhaust HEPA filter and upstream of the supply bubble tight damper or HEPA filter

Physical Requirements - SA Facilities

<b>AIR HANDLING SYSTEM - CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3,4	air supply and exhaust ductwork to be sealed airtight (in accordance with requirements specified in Section 7) between the animal room perimeter and bubble tight damper
3,4	all air supply and exhaust ductwork that is located outside the rooms to be accessible
3,4	bubble tight dampers and HEPA filters to be located as close as possible to the containment perimeter

**Note:** Flexibility should be designed into HVAC systems to accommodate for change (i.e. fan/motor capacity should be slightly oversized).

<b>SA FACILITY SERVICES (i.e. WATER, ELECTRICITY)</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2,3,4	exposed SA facility services piping with stand-offs to allow access for maintenance and cleaning
3,4	water supply control to be located outside SA facility zone
3,4	supply water services to be provided with backflow prevention at the perimeter of SA facility zone (i.e. in addition to premises isolation); selection of backflow prevention device to be in accordance with <i>CAN/CSA-B64.10-94 Manual for the Selection, Installation, Maintenance, and Field Testing of Backflow Prevention Devices</i>
4	for activities involving zoonotic agents, compressed breathing air to be provided to positive-pressure personal protective equipment (i.e. for connection to the air hose of suits), equipped with breathing air compressors and back-up cylinders; air hose connections to be provided in all areas where suits are worn, including chemical shower and suit change room
2,3,4	handwashing sinks to be provided with "hands-free" capability
2,3,4	cage washer to be provided with temperature of final rinse water to be at least 82°C

Physical Requirements - SA Facilities

**SA FACILITY SERVICES (i.e. WATER, ELECTRICITY) - CONTINUED**

<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2,3,4	drainage traps to be provided at depth in accordance with pressure differentials (i.e. to maintain water seal); 15 cm P-traps are recommended
3,4	drains and associated piping to be separated from other zones (i.e. to go directly to liquid effluent treatment system)
3,4	drains and associated piping to be connected to an effluent sterilization system and should be sloped towards sterilization system to ensure gravity flow; consideration should be given to the installation of valves to isolate sections for decontamination; piping to be heat and chemical resistant consistent with application; joints should be by thermo/chemical fusible means or welding to ensure integrity of entire system (i.e. in accordance with pressure decay testing specified in Section 7)
3,4	plumbing vent lines (including effluent sterilization system) to be provided with filter of efficiency equivalent to HEPA; level 4 requires two stages of filtration
3	plumbing vent lines can be combined with areas of lower containment when provided with a filter of efficiency equivalent to HEPA before the connection, i.e. upstream from the connection
3,4	plumbing vent lines to be heat-resistant consistent with application
3,4	supply conduit and wiring to be sealed at the containment barrier (i.e. to provide room integrity in accordance with pressure decay testing specified in Section 7)
2,3,4	light ballasts should be on a separate distribution layout from normal or emergency power to minimize harmonic current problems for sensitive lab equipment; High Intensity Discharge (HID) lamps with lengthy re-strike times should be avoided where there is no alternate quick strike light source
4	light ballasts and starters to be located outside containment perimeter



Physical Requirements - SA Facilities

<b>SA FACILITY SERVICES (i.e. WATER, ELECTRICITY) - CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3,4	power system circuit breakers to be located outside containment perimeter
2,3,4	circuit-breakers and controls to be appropriately labelled
2,3,4	life-safety systems, lighting, and essential equipment to be supported by normal emergency power
3,4	HVAC systems to be supported by normal emergency power
3,4	communication system to be provided between work area and outside SA Facility zone
3,4	system (e.g. fax, computer) to electronically transfer information and data from the SA Facility to outside the containment zone to be provided (note: removing paperwork from the containment zone may be carried out after appropriate decontamination, i.e. autoclaving, irradiation, microwaving; such practices are generally not recommended for use on a routine basis)
4	animal rooms to be visually monitored (e.g. closed circuit TV) from outside containment zone (e.g. security/biosafety office); observation windows are recommended

### 3.3 LARGE ANIMAL FACILITIES

Large animal (LA) facilities are suitable for work with livestock, deer and other game ranching animals not normally housed in cages. Small animals and birds may be housed in cages or isolators in an LA facility providing the requirements listed above for SA facilities are met (e.g. cage washing capability). The manipulation of animal pathogens and other laboratory work associated with LA facilities is to be carried out in laboratories as described in Section 3.1.

Animal cubicles must be constructed to contain large numbers of microorganisms which may be present. Unlike a laboratory room where the biological safety cabinet provides primary containment, the animal cubicle serves as both the primary and secondary barrier. A "clean and dirty" (i.e. "entry and exit") corridor concept is operationally preferable to a "single" corridor design. The clean and dirty corridor facilitates the traffic flow of animal handlers, scientific staff, animals, feed, equipment and samples. This design also minimizes the risks of cross-contamination between animal rooms.

The post-mortem (PM) room should be located within the animal facility. It is likely the area of greatest contamination and as such, should be the area of greatest negative pressure. Air and traffic should flow from the animal cubicle toward the PM room. The design requirements listed below for LA facilities (e.g. access, surface finishes, containment perimeter, HVAC, and services) apply equally to "stand-alone" (i.e. not located within an animal facility) PM rooms.

An "LA facility zone" is defined as an area of equal containment level. A "cubicle" is defined as the room in which the LA is housed. The containment perimeter/barrier of the LA facility zone is continuous and non-intersecting (i.e. the zone is serviced by a single entry/exit)

LOCATION	
AP CONTAINMENT LEVEL	REQUIREMENT
2,3,4	LA facility zones to be separated from other laboratory activities

**Note:** For new level 3 and level 4 construction it is recommended that animal cubicles be located away from exterior envelope walls.

Physical Requirements - LA Facilities

LOCATION - CONTINUED	
AP CONTAINMENT LEVEL	REQUIREMENT
2,3,4	office areas (i.e. a separated room providing a support function normally requiring no containment principles) to be located outside of LA facility zone
2,3,4	clerical work areas for animal handlers permitted within LA facility zone but outside of cubicles and corridors
2,3,4	storage areas for short-term storage of small amounts of feed (i.e. not for bulk storage) to be provided within the LA facility
2,3,4	a PM room should be provided within the LA facility zone
2,3	the PM room should be provided with an integral cold room for storage of LA carcasses awaiting necropsy or disposal and a laboratory support area

**Note:** The level 4 PM room may be the same room as the animal cubicle (i.e. necropsy is conducted within the cubicle).

PERSONNEL ACCESS	
AP CONTAINMENT LEVEL	REQUIREMENT
2,3,4	dedicated and controlled access to be limited to authorized personnel into the LA facility zone
3,4	access to be controlled within the LA facility zone, into each cubicle, and into post-mortem room
2,3,4	entry doors to the LA facility zone to have appropriate signage (i.e. hazard identification, name and phone number of contact person, entry requirements)
2	entry to LA facility zone to be provided via ventilated airlock (i.e. ventilation to be provided through leaky doors and/or HVAC systems)
3	entry to LA facility zone to be provided via ventilated airlock (i.e. ventilation to be provided through leaky doors and/or HVAC systems) with interlocking doors

Physical Requirements - LA Facilities

<b>PERSONNEL ACCESS - CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
<b>4</b>	entry to LA facility zone to be provided via ventilated airlock with tightly sealed interlocking doors to provide room integrity in accordance with Section 7
<b>2,3,4</b>	entry to LA facility zone to be provided with clothing change area designed to separate personal clothing from LA facility clothing dedicated to that zone (i.e. "clean" change area separated from "dirty" change area)
<b>3,4</b>	entry to LA facility zone to be provided with a shower on the containment barrier (i.e. between "dirty" and "clean" change areas)
<b>2,3,4</b>	where the cubicles within the LA facility zone have unique hazards, entry doors to each cubicle to have appropriate signage (i.e. hazard identification, personal protective equipment requirements)
<b>3</b>	where the LA facility uses a single corridor design, entry to cubicles to be provided via ventilated airlock (i.e. ventilation to be provided through leaky doors and/or HVAC systems) with interlocking doors to prevent migration of air from the cubicle to corridor
<b>3</b>	where the LA facility uses a single corridor design, entry to cubicles to be provided with area designed to separate LA facility clothing from dedicated cubicle clothing
<b>3</b>	where the LA facility uses a single corridor design, entry to cubicle to be provided with a shower on the containment barrier (i.e. between cubicle and corridor)
<b>4</b>	entry to cubicles to be provided via ventilated airlock with tightly sealed interlocking doors (e.g. inflatable or compression seal) to prevent migration of air from the cubicle to corridor
<b>4</b>	entry to cubicle to be provided with area designed to separate LA facility clothing from dedicated cubicle clothing and with a suit change areas
<b>4</b>	entry to cubicle to be provided with a chemical shower on the containment barrier (i.e. cubicle and corridor)



Physical Requirements - LA Facilities

<b>PERSONNEL ACCESS - CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3	entry to PM room (other than from dirty corridor) to be provided via ventilated airlock (i.e. ventilation to be provided through leaky doors and/or HVAC systems) with interlocking doors
4	entry to PM room (other than from dirty corridor) to be provided via ventilated airlock with tightly sealed interlocking doors (e.g. inflatable or compression seal)
2,3,4	entry to PM room (other than from dirty corridor) to be provided with clothing change area designed to separate personal clothing from LA facility clothing dedicated to that zone (i.e. "clean" change area separated from "dirty" change area)
3,4	entry to PM room (other than from dirty corridor) to be provided with a shower on the containment barrier (i.e. between "dirty" and "clean" change areas)
4	entry to PM room (other than from dirty corridor) to be provided with a chemical shower on the containment barrier
3,4	controlled entrance/exit doors to have emergency manual overrides

<b>ANIMAL ACCESS</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2	animal entry to LA facility zone to be provided via ventilated airlock (i.e. ventilation to be provided through leaky doors and/or HVAC systems)
3	animal entry to LA facility zone to be provided via ventilated airlock (i.e. ventilation to be provided through leaky doors and/or HVAC systems) with interlocking doors
4	animal entry to LA facility zone to be provided via ventilated airlock with tightly sealed interlocking doors (e.g. inflatable or compression seal) to prevent migration of air from higher to lower containment zones

Physical Requirements - LA Facilities

<b>ANIMAL ACCESS - CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3,4	animal entry/exit to cubicle to be provided via sealed door (i.e. level 3 - four sided door jam; level 4 - inflatable or compression seal) between cubicle and corridor
3,4	animal entry to PM room (other than from dirty corridor) to be provided via sealed door (i.e. level 3 - four sided door jam; level 4 - inflatable or compression seal)

<b>SURFACE (i.e. FLOORS, WALLS, CEILINGS, SEALANTS) FINISHES</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2,3,4	interior coatings to be gas and chemical resistant in accordance with LA facility and cubicle function (e.g. will withstand cleaning, chemical disinfection, fumigation)
2,3,4	interior surfaces to be continuous
2,3,4	interior surfaces to minimize movement of gases and liquids through perimeter membrane (i.e. cubicles to provide room integrity in accordance with pressure decay testing specified in Section 7)
2,3,4	interior surfaces to provide impact resistance in accordance with LA facility and cubicle function
2,3,4	interior surfaces to maintain adherence and integrity under high pressure washing stresses (e.g. 90°C @ 1500 psi)
2,3,4	interior surfaces to be compatible with adjacent and overlapping materials (i.e. to maintain adhesion and a continuous perimeter)
2,3,4	continuity of seal to be maintained between the floor and wall (a cove floor finish 1m up the cubicle, dirty corridor and PM room wall is recommended)
2,3,4	LA facility floors to be slip-resistant and cleanable
2,3,4	animal cubicle, corridor and PM room floors to slope towards floor drain (recommended pitch of slope is 2.1cm/m)

Physical Requirements - LA Facilities

<b>SURFACE (i.e. FLOORS, WALLS, CEILINGS, SEALANTS) FINISHES - CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2,3,4	animal cubicle, corridor and PM room floors to withstand required loading and cleaning in accordance with function
2,3,4	where applicable, cubicle floor matting to be chemical gas and liquid resistant in accordance with cubicle function
2,3,4	doors and frames to be non-absorptive and have solid finishes (i.e. wood is not acceptable); hollow doors must be sealed

<b>CONTAINMENT PERIMETER</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2,3,4	protruding obstructions to be minimized in animal cubicles and corridors; to protect animals, unguarded projections to be at a height of at least 213cm (may be higher for certain species, e.g. deer)
3,4	all mechanical, electrical and service piping penetrations to be sealed at containment perimeter
2,3,4	containment perimeter (e.g. doors) to be kept closed in order to provide required containment of air systems
2,3,4	windows to provide required level of security
2,3,4	windows with direct access between outside the LA facility and cubicles/PM room not to be provided; viewing windows into cubicles acceptable
2	proven technologies for sterilization (e.g. incineration, chemical or gas sterilization, rendering, irradiation, autoclaving) to be provided within LA facility zone
3,4	proven technologies for sterilization (e.g. incineration, chemical or gas sterilization, rendering, irradiation, autoclaving) to be provided at containment barrier
2,3,4	LA facility zone to be proofed against entry or exit of vermin or insects



Physical Requirements - LA Facilities

<b>AIR HANDLING SYSTEM</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
<b>2,3,4</b>	inward directional airflow to be provided; non-recirculated air should be supplied to level 2 LA Facilities and must be supplied to level 3 and level 4 LA Facilities
<b>2,3,4</b>	HVAC from animal cubicles to provide specified number of air changes as required by the CCAC and to minimize dead air spaces within the cubicle; supply and exhaust diffusers to be located to provide convection patterns that ensure airflow away from room entrance; diffuser selection to provide minimal throw velocities, i.e. < 15 m/m @ 1 m.
<b>3,4</b>	minimum of 25 Pa difference is recommended across a containment barrier
<b>3,4</b>	pressure monitoring devices to be provided at the LA facility zone entrance to monitor negative pressure between containment barriers; monitors are also recommended at cubicle and PM room entrance
<b>3,4</b>	room static pressure monitoring lines to be provided with filters of at least equal efficiency to HEPA filter
<b>3,4</b>	audible alarms to be provided in the LA facility work area and outside LA facility zone to detect positive pressurization and air handling systems failure
<b>3,4</b>	air supply and exhaust HVAC system to be independent from adjacent zones (level 3 supply can be combined with areas of lower containment when provided with a bubble tight damper or HEPA filter after the connection, i.e. downstream of the connection; level 3 exhaust can be combined with areas of lower containment when provided with a HEPA filter before the connection, i.e. upstream from the connection)
<b>3</b>	air supply to prevent backdraft of contaminated air through air supply duct (i.e. to be HEPA filtered or provided with a bubble tight damper)
<b>4</b>	air supply to be HEPA filtered
<b>3</b>	air exhaust to be HEPA filtered
<b>4</b>	air exhaust to be pass through two stage of HEPA filtration

Physical Requirements - LA Facilities

<b>AIR HANDLING SYSTEM - CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3,4	roughing pre-filters (e.g. 30% and 85% - in accordance ASHRAE Standard 52.1-1992 <i>Gravimetric and Dust-spot Procedures for Testing Air-cleaning Devices Used in General Ventilation for Removing Particulate Matter</i> ) to be provided to protect the HEPA filter
3,4	air supply and exhaust to be equipped with bubble tight dampers to permit gaseous decontamination (can be same bubble tight damper as required for backdraft protection and for isolation of the HEPA filters)
3,4	air supply HVAC system to be controlled (i.e. flow, pressure, electrical) with exhaust HVAC system, and vice versa, to prevent cubicle/PM room positive pressurization
3,4	airflow control devices and duct sensors to be located downstream of the exhaust HEPA filter or upstream of the supply bubble tight damper or HEPA filter
3,4	air supply and exhaust ductwork to be sealed airtight (in accordance with requirements specified in Section 7) between the cubicle/PM room perimeter and bubble tight damper
3,4	all air supply and exhaust ductwork that is located outside the room to be accessible
3,4	bubble tight dampers and HEPA filters to be located as close as possible to the containment perimeter

**Note:** Flexibility should be designed into HVAC systems to accommodate for change (e.g. fan/motor should be slightly oversized).

<b>LA FACILITY SERVICES (i.e. WATER, ELECTRICITY)</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
2,3,4	exposed LA facility services piping with stand-offs to allow access for maintenance
3,4	water supply control to be located outside LA facility zone

Physical Requirements - LA Facilities

**LA FACILITY SERVICES (i.e. WATER, ELECTRICITY) - CONTINUED**

<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3,4	supply water services to be provided with backflow prevention at the perimeter of LA facility zone (i.e. in addition to premises isolation); selection of backflow prevention device to be in accordance with <i>CAN/CSA-B64.10-94 Manual for the Selection, Installation, Maintenance, and Field Testing of Backflow Prevention Devices</i>
4	for activities involving zoonotic agents, compressed breathing air to be provided to positive-pressure personal protective equipment (i.e. for connection to the air hose of suits), equipped with breathing air compressors and back-up cylinders; air hose connections to be provided in all areas where suits are worn, including chemical shower and suit change room
2,3,4	handwashing sinks to be provided with "hands-free" capability
2,3,4	drainage traps to be provided at depth in accordance with pressure differentials (i.e. to maintain water seal); 15 cm P-traps are recommended
3,4	drains and associated piping to be separated from other zones (i.e. to go directly to liquid effluent treatment system)
3,4	drains and associated piping to be connected to an effluent sterilization system and should be sloped towards sterilization system to ensure gravity flow; consideration should be given to the installation of valves to isolate sections for decontamination; piping to be heat and chemical resistant consistent with application; joints should be by thermo/chemical fusible means or welding to ensure integrity of entire system (i.e. in accordance with pressure decay testing specified in Section 7)
3,4	plumbing vent lines (including effluent sterilization system) to be provided with filter of efficiency equivalent to HEPA; level 4 requires two stages of filtration
3,4	plumbing vent lines to be heat-resistant consistent with application



Physical Requirements - LA Facilities

<b>LA FACILITY SERVICES (i.e. WATER, ELECTRICITY) - CONTINUED</b>	
<b>AP CONTAINMENT LEVEL</b>	<b>REQUIREMENT</b>
3	plumbing vent lines can be combined with vent lines from areas of lower containment when provided with a filter of efficiency equivalent to HEPA before the connection, i.e. upstream from the connection
3,4	supply conduit and wiring to be sealed at the containment barrier (i.e. to provide room integrity in accordance with pressure decay testing specified in Section 7)
2,3,4	light ballasts should be on a separate distribution layout from normal or emergency power to minimize harmonic current problems for sensitive lab equipment; High Intensity Discharge (HID) lamps with lengthy re-strike times should be avoided where there is no alternate quick strike light source
4	light ballasts and starters to be located outside containment perimeter
3,4	power system circuit breakers to be located outside containment perimeter
2,3,4	circuit-breakers and controls to be appropriately labelled
2,3,4	life-safety systems, lighting, and essential equipment to be supported by normal emergency power
3,4	HVAC systems to be supported by normal emergency power
3,4	communication system to be provided between work area and outside LA Facility zone
3,4	system (e.g. fax, computer) to electronically transfer information and data from the LA Facility to outside the containment zone to be provided (note: removing paperwork from the containment zone may be carried out after appropriate decontamination, i.e. autoclaving, irradiation, microwaving; such practices are generally not recommended for use on a routine basis)
4	cubicles to be visually monitored (e.g. closed circuit TV) from outside the LA Facility zone (e.g. security/biosafety office); observation windows are recommended

### 3.4 HIGH EFFICIENCY PARTICULATE AIR (HEPA) FILTERS

HEPA filters are fabricated of pleated sheet of borosilicate fibres divided by corrugated aluminum separators. This fragile medium can damage easily if stored improperly, dropped or otherwise handled carelessly. Filter integrity must be verified after installation or relocation and regularly thereafter. The frequency of reverification depends on a variety of factors including hours of operation and can be determined by the monitoring the filter integrity over time. Filters should be initially tested on an annual basis.

The filter medium is glued into a wood or metal frame. Wood frames, although easy to dispose of, have the disadvantage of absorbing vapours (e.g. vaporized hydrogen peroxide) during routine sterilization of the filter. Vapours are subsequently slowly released from the frame which lengthens the time needed to reduce to the concentration to the acceptable values (i.e. total cycle time for gaseous decontamination is extended). Metal frames do not absorb such vapours however present difficulties during disposal.

HEPA filters are typically installed in filter housings by means of a gasket (e.g. neoprene) or fluid (e.g. gel) seal. Common problems with gaskets is that they may lose "memory" due to compression, may tear and may be incompatible with gaseous decontaminants. For example, some neoprenes (e.g. open-celled black neoprene gaskets) are degraded by hydrogen peroxide. Dense gasket material lasts for more sterilization cycles than an open-celled gasket made of similar material. Gel seals establish an airtight seal between the filter and housing by means of a channel around the filter perimeter filled with gel. The housing knife edge flange seals into this channel. Gel seals are not prone to the compression and compatibility problems associated with gasket seals.

Requirement for all HEPA filters (i.e. in biological safety cabinets or air handling systems) are as follows:

- HEPA filters to have minimum efficiency of 99.97% at 0.3 $\mu$ m in accordance with the Institute of Environmental Sciences IES-RP-CC-001-86 *Recommended Practice for HEPA Filters* (1986); filters to be factory tested by particle challenge testing using the scanning method
- the static pressure (i.e. cleanliness) of HEPA filters to be monitored by pressure monitoring devices, e.g. magnehelic gauges



## HEPA Filters

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- the in-situ integrity of HEPA filters to be verified by in-situ particle challenge testing using the scanning method (particle penetration not to exceed 0.01%)

Note: the integrity of other in-line filters (e.g. plumbing vent lines, gas supply lines, autoclave exhaust ducts) to be verified by particle challenge tests; filter efficiency to be equal to that of HEPA filter

Requirements for in-line HEPA filter housings installed into air handling systems at any containment level are as follows:

- HEPA filter housings to be provided with bubble-tight dampers on the air inlets and outlets for shut-off and isolation of the filter
- HEPA filter housings to be provided with upstream and downstream fumigation ports to allow for in situ gaseous decontamination
- HEPA filter housings to be provided with upstream injection and downstream access ports to allow for in situ particle challenge tests by the scanning method
- HEPA filter housings to be leaktight in accordance with pressure decay testing specified in Section 7

### 3.5 FURNISHINGS

Generally, laboratory casework should be designed in accordance with function, the hazardous materials likely to be encountered, and ease of decontamination.

Modularity, a desirable feature accommodating for change, is less valuable in containment facilities. The more modular the unit, the more seams, crevices, and joints there are.

The following outlines desirable features of furnishings which can be applied in a generic fashion. Some requirements will differ with the facility type and containment level and these are so indicated below.

- surfaces to be scratch, stain, moisture, chemical and heat resistant in accordance with function
- solid-core materials to be used in AP containment level 3; wood is not recommended
- stainless steel construction to be used in AP containment level 4, SA and LA facility animal rooms, and PM rooms
- floor contact surface to be rust resistant except where stainless steel is specified
- bench tops to be continuous (i.e. with no open seams) and to contain spillage of materials (e.g. with marine edges and drip stops)
- benches, doors, drawers, door handles, etc. to have rounded rims and corners in AP containment level 4 where positive-pressure suits are used
- backsplashes to be continuous with work surfaces (i.e. with no open seams)
- backsplashes to be installed tight to wall and sealed at wall-bench junction
- service raceway channels and upper cabinets to be sealed at junction to bench
- benching in AP containment level 4 to be continuous but not fixed (i.e. entire system to be removable but with no modular elements)
- modularity in AP containment level 3 to be limited to the movability and removability of banks of drawers

## Furnishings

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- it is recommended that doors beneath counters swing out, not slide; it is recommended that doors above counters slide, not swing out
- it is recommended that doors not be self-closing
- drawers to be equipped with catches, i.e. to prevent the drawer from being pulled out of the cabinet
- drawer castors to be nylon rollers, not sliders
- drawers in AP containment level 4 to be of one piece construction
- reagent shelving to be equipped with lip edges
- sinks to be integral with bench top where same material is used
- handwashing sinks to be of hands-free operation

## 4. FUME HOODS

Fume hoods should be routinely used to contain hazardous gases, vapours, mists, aerosols and particulates generated during the manipulation of chemical substances. To be effective, a fume hood must confine contaminants within the hood, remove them through the ductwork, and filter or disperse them so that they do not return to the building through air intake systems. All laboratory fume hoods can be described as one of three types:

- Conventional - basic enclosure with a movable front sash; as sash is opened the air volume entering the hood remains constant, but the air velocity decreases due to reduced flow constriction
- By-pass - operates at a constant exhaust air volume, regardless of sash position; accomplished with openings above the hood sash through which air passes when the sash is closed
- Auxilliary-air - equipped with a supply air system separate from the room supply system; discouraged by the ASHRAE Handbook, *Heating, Ventilating, and Air-conditioning Applications* (1995) due to difficulties and installation criteria associated with the supply of auxilliary air

Note: recirculating ductless fume hoods have limited use in the laboratory because of the wide variety of chemicals used. Inappropriate filter selection, inappropriate chemicals and/or chemical concentrations, and inadequate monitoring systems for filter replacement can lead to contaminated air being recirculated into the work environment.

Materials exhausted from fume hoods are often disposed of by dilution into the atmosphere. The inclusion of exhaust air treatment devices (e.g. activated carbon filters) must be consistent with applicable local regulations. Where required, filters must be selected according to the type of contaminant to be removed, the efficiency required to meet occupational and/or environmental exposure limits and the required residence time. Filters must be located upstream of the exhaust fan and must permit replacement without contaminating the surrounding environment. Filters present an initial high cost and are associated with high maintenance associated. Carbon filters also present present a pollution source during disposal.

Fume hoods located inside containment level 3 and 4 laboratories must comply with the requirements for HEPA filtration (see Section 4). The installation of a charcoal filter prior to the HEPA filter may be considered as a measure to protect the HEPA



filter from deleterious effects from chemical vapours and to protect the individual performing maintenance and certification testing of the HEPA filter.

Fume hoods and associated exhaust systems must comply with design and installation requirements outlined in the CSA standard Z316.5-94, *Fume Hoods and Associated Exhaust Systems* (1994). Fume hoods should be located as follows:

- away from normal traffic patterns and interfering room air currents
- the front sash should be 1.5 m away from room air exhaust diffuser and 1.5 m away from air supply diffusers (this distance can be reduced when using velocity controlled diffusers, i.e. maximum throw velocity of 15 m/m @ 1 m from diffuser or 1/2 face velocity of fume hood, and when proven that fume hood performance is not affected)
- the front sash should be 1.5 m away from doorways and,, 2.0 m away from opposing walls or other obstructions,
- the side of the fume hood should be 0.3 m away from walls or other obstructions projecting beyond the plane of the sash and 1 m away from doorways
- the hood should not be located directly opposite seated work stations, other fume hoods or biological safety cabinets

The CSA Z316.5-94 standard for fume hoods requires the performance of tests conducted on all fume hoods upon initial installation and periodically thereafter to maintain them in good operating condition. All requirements for test conditions, equipment accuracy and calibration, and outlined in the CSA standard must be followed. This includes the performance of tests in accordance with ANSI/ASHRAE 110, *Method of Testing Performance of Laboratory Hoods*, (1995).

## 5. BIOLOGICAL SAFETY CABINETS

When properly maintained and used in conjunction with good laboratory techniques, biological safety cabinets (BSCs) provide effective primary containment for work with zoonotic pathogens. In AP containment level 2 facilities, BSCs are used for procedures with potential for producing infectious aerosols and with high concentrations or large volumes of infectious material. In AP containment level 3, all open vessel activities with zoonotic materials are conducted in a biological safety cabinet. In AP containment level 4, all activities with microorganisms are conducted in a BSC in order to minimize contamination of the lab. Every employee working in a BSC must be trained in its correct use and have a good understanding of the different types of cabinets and how they work.

Detailed information on the selection, function and use of BSCs can be found in the Centers for Disease Control/National Institutes of Health *Primary Containment for Biohazards: Selection, Installation and Use of Biological Safety Cabinets* (1995).

### 5.1 CLASSES OF BIOLOGICAL SAFETY CABINETS

There are three classes of biological safety cabinets: Class I, Class II and Class III. Class I cabinets have unrecirculated air flow away from the operator that is discharged through a HEPA filter. Class I cabinets provide good operator protection but do not protect the material within the cabinet (the product) from contamination. Class II cabinets have inward air flow for personnel protection, downward HEPA-filtered air for product protection and HEPA-filtered exhaust air for environmental protection. They are divided into two types (A and B) based on construction, air flow velocities and patterns, and exhaust systems. Class III cabinets are totally enclosed and gas-tight with HEPA filtered supply and exhaust air. Work is performed with attached long-sleeved gloves. Class III cabinets protect the worker and the product.

**Note:** Horizontal clean benches which direct air towards the operator are not biological safety cabinets and must not be used for handling infectious, toxic or sensitizing materials.

Selection of the proper class of BSC requires careful evaluation of the work involved. Class II (i.e. Type A and Type B) cabinets are designed for work involving microorganisms in AP containment level 2, 3 and 4 facilities. Cabinet air from Type A cabinets may be recirculated back into the laboratory in level 2 and 3 facilities. Ducting a Type A cabinet out of the building is possible, providing the method of ducting uses a "thimble" connection (i.e. a small opening around the cabinet exhaust filter housing) and the balance of the cabinet exhaust system is not disturbed. The thimble must be removeable or be designed to allow for proper certification of the cabinet (i.e. bubble tight damper to seal off the cabinet for decontamination, access port to allow scan testing of the HEPA filter).

Class II, Type B cabinets can be used when manipulating small quantities of chemicals as an adjunct to work with microorganisms in AP containment level 2, 3 and 4 laboratories. Type B1 cabinets recirculate 30% of the air within the cabinet and are suitable for work with minute amounts of chemicals and radionuclides. These cabinets must be "hard-ducted" (i.e. direct connection), preferably to their own dedicated exhaust system. The exhaust canopy must allow for proper BSC certification. Type B2 cabinets are total-exhaust cabinets with no air recirculation within them and are suitable for work with small amounts of volatile chemicals and radionuclides. These cabinets are also hard-ducted. The Type B3 cabinet is similar to a ducted Type A cabinet (also referred to as Type A/B3) with negative air pressure plenums. Type B3 cabinets are not recommended for work with volatile chemicals as recirculation of 70% of air can cause a build-up of chemical vapours in the cabinet.

Class III cabinets are designed for work with HC level 4 pathogens and have traditionally been installed in maximum containment laboratories. Cabinet lines consisting of several class III cabinets (e.g. for centrifuges, incubators, refrigerators) and transfer devices joined together must be custom built. Caution should be exercised for their use in level 4 laboratories as they require specialized laboratory support systems (e.g. transfer devices, HVAC, emergency power, controlled access). Class III cabinets may also have a role in field applications where full supporting structures are not necessarily needed.

**Selection of Class II Biological Safety Cabinets**

AP Containment Level	Application	BSC Class	Exhaust Pattern
2, 3	- microorganisms	Type A	Recirculated
2, 3	- microorganisms	Type B3	Thimble-ducted
2, 3, 4	- microorganisms - volatile chemicals and radionuclides	Type B2	Hard-ducted
2, 3	- microorganisms - volatile chemicals and radionuclides (minute amounts)	Type B1	Hard-ducted

**Note:** Only cabinets which meet the National Sanitation Foundation (NSF) Standard No. 49 *Class II Biohazard Cabinetry* (1992) (design, materials and construction specifications for BSCs) and bear an NSF 49 seal are to be purchased.

**5.2 INSTALLATION AND CERTIFICATION**

The air curtain at the front of the cabinet is fragile and can easily be disrupted by people walking parallel to it, by open windows, air supply registers, or laboratory equipment that creates air movement (e.g. vacuum pumps, centrifuges). The following outline recommendations for BSC placement:

- BSCs to be located away from high traffic areas, doors and air supply/exhaust ducts that may interrupt air flow patterns (generally, the same principles outlined for fume hood locations should apply)
- minimum clearance of 30cm to be provided between exhaust outlet on top of cabinet and any overhead obstructions
- whenever possible, a 30cm clearance to be provided on each side of the cabinet to allow for access



- for ducted cabinets, blowers on the exhaust system should be located at the terminal end of the ductwork; failure of exhaust flow should signal an alarm to the user; to prevent pressurization of the cabinet, an interlock system should be installed to prevent the cabinet blower from operating whenever the exhaust flow is insufficient (e.g. flow/electrical control); an anti-backflow device to prevent reverse airflow through the HEPA filter may be required

Continuous operation of BSCs helps to control dust levels and other airborne particulates in the laboratory. Operating BSCs only when needed to conserve energy must consider the balancing of laboratory room air. In some cases, room exhaust is balanced to include the air exhausted through ducted BSCs and these cabinets must not be turned off.

The provision of propane gas to BSCs is not generally recommended. Open flames in the BSC create turbulence, disrupt air flow patterns and can damage the HEPA filter. When suitable alternatives (e.g. disposable sterile loops, micro-incinerators) are not possible, touch-plate microburners with a pilot light to provide a flame on demand may be used.

The correct operation of BSCs must be verified before they are used, after any repairs or relocation. Moving a cabinet can cause damage to the HEPA filter and its seals. Testing must be performed by qualified individuals and it is recommended that only NSF accredited field certifiers be used. The following outline requirements for BSC certification:

- BSCs to be certified by NSF accredited field certifiers in accordance with the CSA Standard Z316.3-95 *Biological Containment Cabinets: Installation and Field Testing* (1995) at least annually and whenever the cabinet has been moved or serviced
- a copy of the certification report must be provided to the user and kept on file (ideally, a pocket containing the report should be affixed to the cabinet exterior)
- a label indicating the date of certification, the date of the next certification, to what standard the tests were performed and the name of the certifier must be affixed to the exterior of the cabinet

## **6. OPERATIONAL PRACTICES**

### **6.1 GENERAL REQUIREMENTS**

The following general practices are required when working in any containment laboratory or animal facility:

- entry must be restricted to laboratory staff, animal handlers, maintenance staff and other persons on official business
- only persons meeting specific entry requirements (e.g. immunization, serum screening) may enter containment laboratories unless the facility has been appropriately decontaminated
- a health and medical surveillance program must be provided as recommended by Health Canada
- personnel must receive training on the potential hazards associated with the work involved and the necessary precautions to prevent exposures to zoonotic agents and release of non-indigenous agents; personnel must show evidence that they understood the training provided; training must be documented and signed by both the employee and supervisor
- a documented procedural manual must be written and followed
- all persons (including visitors, maintenance staff, etc.) entering the containment area must be trained and know and follow the operational protocols for the project in process; trainees must be accompanied by a trained staff member
- persons entering a containment facility must be well prepared and bring all materials they will need with them; if something has been forgotten, traffic patterns must still be adhered to (ie. do not go back to get it; either phone for someone to bring it or exit via proper protocols)
- employees working in the containment area must have general knowledge of the physical operation and design of the facility (e.g. air pressure gradients between zones, directional air flow patterns, alarm signals for air pressure failure, containment perimeter)

## Operational Practices - General

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- traffic flow patterns from clean to dirty areas must be established and adhered to (i.e. move from least to most contaminated areas)
- smoke testing (i.e. with a smoke pencil) should be done periodically by lab staff to verify correct airflow
- entry/exit protocols for persons, animals, equipment, samples, waste, etc. must be written, posted and followed; general protocols must be supplemented with protocols specific for each project in progress
- emergency procedures for entry/exit, spill clean-up, air handling/biosafety cabinet failure, fire, animal escape and other emergencies must be written, posted and followed
- in the event of life-threatening emergencies, personal health and safety are a priority; exit protocols must be established whereby routine procedures are bypassed; a reporting area must be identified where further steps must be taken (e.g. disinfecting footwear, changing, showering) prior to leaving
- all spills, accidents, overt or potential exposures to infectious materials, and losses of containment (e.g. lab positive pressurization) must be reported immediately to the laboratory supervisor; written records of such incidents must be maintained
- an effective rodent and insect control program must be maintained

## 6.2 LABORATORIES

The following describes the minimum operational practices required at **AP containment level 2**:

- laboratory personnel must be trained in and follow the safe use of laboratory equipment, biological safety cabinets, procedures to minimize the production of aerosols, decontamination and emergency response
- open wounds, cuts, scratches and grazes should be covered with waterproof dressings
- eating, chewing gum, drinking, smoking, storing food, and applying cosmetics are prohibited
- personal items such as purses and outdoor clothing should be kept separate from work areas
- the work area containing hazardous materials should be kept free from materials not pertinent to the work and that cannot be easily decontaminated (e.g. journals, books, correspondence); paperwork and report writing should be kept separate from such work areas
- laboratory reference material should be kept in the laboratory zone
- hands should be washed frequently (after handling infectious materials, after removing gloves, and before leaving the laboratory)
- open-toed and high-heeled shoes must not be worn in the laboratory
- long hair should be tied back so that it cannot come into contact with hands, specimens, containers, or equipment
- gloves (e.g. intact vinyl or latex) must be worn when handling infectious materials; metal mesh gloves can be worn underneath the latex or vinyl glove to provide protection from sharps and needles
- laboratory coats, gowns or coveralls must be worn when working in the laboratory; this clothing must not be worn in non-laboratory areas (e.g. offices, staff rooms, canteens, libraries)



## Operational Practices - Laboratories

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- protective lab clothing should not be stored in the same locker as street clothing
- contaminated clothing must be decontaminated prior to laundering (unless laundering facilities are within the laboratory zone and have been proven to be effective in decontamination)
- eye and face protection must be worn when it is necessary to guard against splashing hazardous materials, flying particles, and harmful light or other rays
- laboratory doors must be kept closed as required by the facility design
- biological safety cabinets must be used for procedures with potential for producing infectious aerosols (e.g. with zoonotic agents) and with high concentrations or large volumes of zoonotic materials
- contaminated work surfaces must be decontaminated
- all contaminated materials must be decontaminated before disposal or cleaning for reuse
- contaminated equipment leaving the laboratory for servicing or disposal must be appropriately decontaminated
- efficacy monitoring of autoclaves using biological indicators must be done at least weekly, depending on the frequency of use of the autoclave, and records of the results kept on file; cycle log records (i.e. time, temperature and pressure) must also be kept on file

In addition to the general operational practices listed for level 2, the following describes the minimum operational practices required at **AP containment level 3**:

- a protocol specific to the operation of the lab must be developed and read by personnel; employees must certify in writing that they have understood the material in the protocol
- the laboratory zone must be kept locked
- infectious agents should be stored inside the laboratory zone; agents stored outside the zone must be kept locked, in leakproof containers

## Operational Practices - Laboratories

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- personnel must have demonstrated proficiency in microbiological practices and techniques (e.g. experience in handling infectious organisms or cell cultures)
- personal items such as purses and outdoor clothing must not be brought into the laboratory zone
- a containment check must be performed prior to entering the laboratory zone (ie. verify negative lab pressurization as designed)
- water seals must be maintained in drainage traps (i.e. through regular sink/shower usage and/or by filling traps in areas that are not being used)
- laboratory samples and supplies may be carried into the laboratory zone or passed through a ventilated pass-box; where the barrier autoclave is used to pass materials into the laboratory, the autoclave must have been cycled prior to opening the outer "clean side" door
- personnel entering the laboratory zone must remove street clothing and jewellery, and change into dedicated laboratory clothing and shoes
- where full body protective clothing is not worn a shower is required on exit from the laboratory; where a known or suspected aerosol exposure has occurred (e.g. dropping infectious materials) a shower is required on exit from the laboratory zone
- a shower (including washing hair, beards) is required on exit from a laboratory zone handling non-indigenous animal pathogens; eye glasses must be disinfected at the containment barrier
- a second layer of protective clothing (ie. solid-front gowns with tight-fitting wrists, gloves) should be worn over laboratory clothing when directly handling infectious materials (e.g. dedicated for use at the biological safety cabinet)
- contaminated clothing must be decontaminated prior to laundering (unless laundering facilities are within the laboratory zone and have been proven to be effective in decontamination of the microorganisms likely to be encountered)

## Operational Practices - Laboratories

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- all activities with infectious materials are conducted in a biological safety cabinet; where this is not possible, other physical containment devices in combination with personal protective clothing and equipment must be used; no work with open vessels containing infectious materials is conducted on the open bench
- centrifugation of infectious materials must be carried out in sealed safety cups or rotors that are loaded and unloaded in a biological safety cabinet
- all contaminated waste materials leaving the laboratory zone must be decontaminated through a double-door autoclave at the barrier before disposal; both doors of the autoclave must not be opened simultaneously
- heat sensitive materials that cannot be autoclaved out of the laboratory zone must be decontaminated at the containment barrier (e.g. fumigated with formaldehyde or vaporized hydrogen peroxide, disinfected using liquid chemicals, or other technology proven to be effective)

In addition to the general operational practices listed for level 2 and 3, the following describes the minimum operational practices required at **AP containment level 4**:

- all persons entering the laboratory zone must have completed a training course in procedures specific to the level 4 lab and must show evidence of having understood the training; training must be documented and signed by the employee and supervisor
- protocols must be established for emergencies including personnel suit damage, loss of breathing air, and loss of chemical shower
- where HC level 4 agents are used, employees must carry an illness surveillance card (e.g. with employee name, supervisor's and alternate's name and phone number, facility phone number); employees must immediately notify their supervisor of any febrile illness; supervisors must contact any employee with unexplained work absences
- a log record is maintained of activities in the laboratory
- infectious agents must be stored inside the laboratory zone
- a daily check of lab systems must be carried out

## Operational Practices - Laboratories

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- personnel entering the laboratory must remove street clothing and jewellery, and change into dedicated laboratory clothing and shoes
- when HC containment level 4 zoonotic agents are manipulated, positive-pressure suits must be worn; suit integrity must be routinely checked for leaks
- a shower is required on exit from the laboratory zone
- a chemical shower of appropriate duration is required when suits are worn; the disinfectant used must be effective against the agents of concern, be diluted as specified and prepared fresh as required



### 6.3 ANIMAL FACILITIES

Work with animals poses a variety of special hazards including exposure to zoonotic agents (naturally occurring or experimentally infected), animal bites and scratches, kicks and crushing injuries, physical hazards (e.g. noise, temperature) and chemical hazards (e.g. cleaning agents, disinfectants). Allergic conditions can result from contact with animal fur or hair, bedding, and animal wastes. At least one-fifth of people who work with laboratory rodents, guinea pigs and rabbits develop allergies. Protection from allergens must be provided through engineering controls, ventilation, use of isolators and cages with filter tops and appropriate use of respiratory protection.

Animal handlers must have knowledge of the species' general characteristics such as behaviour, instincts and physical attributes. Consideration should also be given to their natural ecto- and endoparasites and the zoonotic diseases to which they are susceptible including their route of excretion and dissemination.

The following describes the minimum operational practices required in **small animal facilities (SA facilities)**:

- coveralls and footwear must be worn when working in AP containment level 2 SA facilities
- personnel entering AP containment level 3 and 4 SA facilities must change into dedicated facility clothing and shoes
- gloves must be worn when handling infected animals
- hands must be washed after handling animals, after removing gloves and before leaving the facility
- HEPA-filtered respirators are required for handling animals in AP containment level 2 and 3 SA facilities where infectious aerosols of zoonotic agents may be generated and cannot be contained within a primary containment device
- positive pressure ventilated suits are required for handling animals infected with HC containment level 4 zoonotic agents
- a shower (including washing hair, beards) is required on exit from AP containment level 3 SA facilities handling non-indigenous agents and when handling other agents where aerosols cannot be contained in primary

containment devices; eye glasses must be disinfected at the containment barrier

- a shower is required on exit from AP containment level 4 SA facilities; a chemical shower is required when suits are worn
- where a clothing change is not performed on exit from each animal room, disinfectant footbaths are required (the disinfectant must be effective against the microorganisms of concern and changed regularly in accordance with the active life of the disinfectant)
- contaminated clothing must be decontaminated prior to laundering (unless laundering facilities are within the laboratory zone and have been proven to be effective against the microorganisms likely to be encountered)
- each animal room must be labelled with unique hazards and entry requirements (e.g. respiratory protection)
- animal room doors must be kept closed as required by facility design
- water seals must be maintained in drainage traps (i.e. through regular sink/shower/floor drain usage and/or by filling traps in areas that are not being used)
- cages housing infected animals must be appropriately labelled
- containment caging systems should be used in AP containment level 3 SA facilities to contain aerosols (e.g. laminar flow cabinets, solid wall and bottom cages covered with filter bonnets)
- containment caging systems must be used in AP containment level 4 SA facilities
- careful handling procedures must be employed so as to minimize the creation of aerosols and dissemination of dust from cages, refuse and animals
- proper methods of restraint must be used to minimize scratches, bites and accidental self-inoculations

## Operational Practices - Animal Facilities

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- necropsy of small animals infected with a zoonotic agent and intranasal inoculation of animals in AP containment level 3 and 4 SA facilities should be conducted in a biological safety cabinet; animals must be securely transported to the biological safety cabinet
- supplies and materials are brought into the AP containment level 3 and 4 SA facilities by carrying them in or by way of a ventilated pass-through; barrier autoclaves, fumigation chambers or airlocks may also be used providing that they have been decontaminated before opening the outer "clean side" door
- animal bedding must be removed in a manner that minimizes the generation of aerosols and dust; for AP containment level 3 and 4 SA facilities handling zoonotic agents, cages must be decontaminated prior to removing bedding
- all contaminated materials must be decontaminated before disposal or cleaning for reuse
- autoclaving is the preferred method of decontaminating cages prior to washing; temperature of the final rinse water in mechanical cage washers should be at least 82°C
- animal carcasses and tissues must be incinerated or processed through new technology proven to be effective (e.g. tissue autoclave); carcasses must be transported from the animal room for disposal in leakproof containers that are appropriately labelled

The following describes the minimum operational practices required in **large animal facilities (LA facilities)**:

- coveralls and footwear must be worn when working in AP containment level 2 LA facilities
- personnel entering AP containment level 3 and 4 LA facilities must remove street clothing and jewellery, and change into dedicated facility clothing and shoes
- personnel entering AP containment level 3 animal cubicles where non-indigenous agents are used, level 3 animal cubicles with a single corridor design and level 4 animal cubicles must change into dedicated cubicle clothing and boots at the cubicle entrance

- where a clothing change is not performed on entry/exit to the animal cubicle, a second layer of protective clothing must be donned when entering the animal cubicle (e.g. rubber boots, gloves, rubber suit); this layer must be decontaminated on exit from the animal cubicle (e.g. disinfectant foot baths)
- gloves must be worn when handling infected animals
- hands must be washed after handling animals, after removing gloves and before leaving the facility
- HEPA-filtered respirators are required for handling animals where infectious aerosols of zoonotic agents may be generated
- positive-pressure ventilated suits are required for handling animals infected with HC containment level 4 zoonotic agents
- a shower (including washing hair, beards) is required on exit from AP containment level 3 LA and level 4 LA facilities; eye glasses must be disinfected at the containment barrier
- a shower is required on exit from AP containment level 3 animal cubicles where non-indigenous agents are used, level 3 animal cubicles with a single corridor design, and level 4 animal cubicles; eye glasses must be disinfected at the containment barrier
- contaminated clothing must be decontaminated prior to laundering (unless laundering facilities are within the laboratory zone and have been proven to be effective against the microorganisms likely to be encountered) (note: autoclaving heavily soiled laundry (e.g. blood, feces) may cause stains to "lock in"; in such cases it may be necessary to launder the clothing first, providing the laundry machines are located within containment)
- each animal cubicle must be labelled with unique hazards and entry requirements (e.g. respiratory protection)
- animal cubicle doors must be kept closed as required by facility design
- water seals must be maintained in drainage traps (i.e. through regular sink/shower/floor drain usage and/or by filling traps in areas that are not being used)



## Operational Practices - Animal Facilities

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- proper methods of restraint must be used to minimize kicks, crushing injuries and accidental self-inoculations
- supplies and materials are brought into the AP containment level 3 and 4 LA facilities by carrying them in or by way of a ventilated pass-through; barrier autoclaves, fumigation chambers or airlocks may also be used providing that they have been decontaminated before opening the outer "clean side" door
- animals are brought into the AP containment level 2,3, and 4 LA facility by means of an airlock
- entering more than one animal cubicle from a clean corridor is generally not acceptable; entering more than one animal cubicle from the dirty corridor may be acceptable depending on the project (e.g. moving between contaminated areas of equal status; working with negative control animals first before proceeding to infected animals)
- the exterior surfaces of containers of biological samples to be removed from contaminated animal rooms must be decontaminated; heat sensitive samples can be chemically disinfected (e.g. immersion in disinfectant on the barrier)
- at the end of the experiment all supplies remaining in the animal cubicles (e.g. supplies, feed) must be removed and decontaminated
- animal carcasses and tissues must be incinerated or processed through new technology proven to be effective (e.g. tissue autoclave); carcasses must be transported from the animal cubicle for disposal via the dirty corridor (alternatively, leakproof containers may be used for transport)
- animal cubicles and the dirty corridor must be cleaned and decontaminated at the end of an experiment using an appropriate procedure; the disinfectant must be effective against the microorganisms of concern; preliminary washing using a general purpose disinfectant/detergent should be done using low-pressure hoses; decontamination can then be achieved by spraying or fumigating with a disinfectant as appropriate

## 6.4 POST-MORTEM ROOMS

Hazards in the post-mortem (PM) room are not limited to splashes and aerosols of infectious materials. Accidents can be caused by cutting instruments, sharp ends of cracked bones, slippery floors, electrical equipment, chemical fixatives and disinfectants.

General precautions:

- only authorized staff are allowed to use the necropsy facilities
- staff must be trained in the use of all equipment and tools (e.g. electric hoist/monorail, tools, PM table, incinerator)
- staff must be trained in proper disinfection and cleaning procedures
- the area must be kept neat and tidy; equipment, paper, reports, etc. should be stored securely and not be accumulated in the PM room to facilitate cleaning and decontamination; floors should be clear of obstructions
- specific protocols for each project must be developed and followed; these include entry/exit protocols (for people, animals, equipment and samples), protective clothing and equipment, disinfection and cleaning protocols, use of the incinerator and autoclaves, and emergency procedures

Preparation for necropsy:

- protective clothing appropriate to the AP containment level and potential hazards must be worn in the PM room; this should include the removal of street clothing and donning of protective clothing and footwear; HEPA-filtered respirators are required when the potential for infectious aerosols exist; waterproof aprons, gloves and eye/face protection (face shield, goggles) should also be worn; a safety helmet is required when operating an electrical hoist/monorail
- specific protocols must be developed for the movement of animals and carcasses into the PM room (e.g. hoist for large animals, cart for small livestock, secure containers for poultry and laboratory animals)

Necropsy Procedures:

- necropsy safety procedures specific to the species involved must be followed (i.e. use of cutting instruments to avoid injury)
- the animals (especially birds and small lab animals) should be wetted with water and/or disinfectant prior to necropsy
- skilful technique is required to prevent excessive spread of contamination and the formation of aerosols originating from fluids and tissues (this is particularly important for work with zoonotic agents); every effort should be made to confine the spread of contamination; this is especially true when there is a likelihood of material being dropped from an elevated position

Cleaning and disposal procedures:

- upon completion of the post mortem, all necropsy tools and instruments must be decontaminated by autoclaving or disinfection (the disinfectant must be effective against the microorganisms of concern); as some disinfectants are inactivated in the presence of organic materials, gross contamination should be removed prior to disinfection
- disposable sharps, needles, blades, glass slides, etc. must be discarded into an appropriate sharps container for decontamination
- the necropsy table, floor and other contaminated work areas must be cleaned and disinfected at the end of an experiment using an appropriate procedure; preliminary washing using a general purpose disinfectant/detergent should be done; special care must be exercised when using a hose to wash the area (i.e. prevent the spread of contamination and formation of aerosols); decontamination of the PM room can then be achieved by spraying or fumigating with a disinfectant effective against the microorganisms of concern
- specimens (fresh, frozen or fixed) for further study should be placed in leakproof containers, appropriately labelled; the outside of the container must be cleaned and disinfected at end of necropsy or upon exit from the PM room; samples may only be opened in a laboratory zone of the same AP containment level

- all animal waste must be incinerated or processed through new technology proven to be effective (e.g. tissue autoclave); the incinerator or tissue autoclave should be located adjacent to the PM room
- where large specimens must be divided into smaller pieces and transported to the incinerator, pieces should be placed carefully into leakproof containers to avoid splashes and aerosols; the outsides of containers must be cleaned down and disinfected prior to transport out of the PM room; the containers must be labelled with the contents and the name and phone number of a contact person

### Exit procedures:

- the requirement for showering out of the PM room is dependent on the microorganism of concern; a full shower out of the facility (including washing hair, beards and glasses) is mandatory when working with zoonotic level 3 and 4 agents, and non-indigenous agents
- contaminated protective clothing must be decontaminated prior to disposal or re-use; contaminated laundry is autoclaved prior to processing (unless using a pass-through laundry machines proven to be effective against the microorganism of concern)



## 6.5 DECONTAMINATION PROCEDURES

Decontamination is defined as the removal of contamination and includes both sterilization (the complete destruction of all microorganisms) and disinfection (the destruction of specific types of microorganisms). Decontamination procedures for waste disposal, for removing materials, equipment, samples from containment zones, for laundry, for contaminated surfaces and rooms, etc. represent a critical containment barrier. Failure in the procedure can result in the unintentional release of agents from the containment facility. It is the responsibility of each facility to see that proper procedures are followed and that containment is not breached. The choice of method is determined by the nature of the material to be treated.

- all decontamination and waste management procedures must be in accordance with applicable federal, provincial, and municipal regulations
- all employees must be trained in decontamination procedures specific to their activities and know the factors affecting the effectiveness of the treatment procedure
- written procedures must be available for each specific decontamination method being used
- all records of efficacy testing and logs of decontamination cycles must be kept on file and be available for inspection as necessary

Several treatment options are briefly discussed below:

### **Autoclaves:**

The effectiveness of decontamination by steam autoclaving is dependent upon the temperature to which the material is subjected as well as the length of time it is exposed. Particular attention must be given to packaging including the size of containers and their distribution in the autoclave. Containers must have good steam permeability and must be arranged in the autoclave in a manner that permits free circulation of steam. Tight fitting containers do not permit steam penetration. Piling containers above one another and overloading can result in decontamination failure.

Effective operating parameters for autoclaves should be established by developing standard loads and their processing times through the use of thermocouples and biological indicators placed at the centre of the load. Biological indicators are also used on a regular basis (e.g. weekly, based on the frequency of use) to monitor the effectiveness of the autoclaving cycle. Records must also be kept of the time, temperature and pressure for each load.

Note: Chemical indicators for steam, time and temperature are useful for day-to-day monitoring that the load has been processed, however, must not be used as an indicator of sterility.

### **Chemical disinfection and fumigation:**

Chemical disinfectants are used for the decontamination of surfaces and equipment which cannot be autoclaved; of specimen containers removed from containment; of spills of infectious materials; of protective equipment and clothing; of laboratories, animal cubicles and other rooms; and a variety of other decontamination procedures where heat treatment is not feasible. The choice of chemical disinfectant is dependent on the resistance of the microorganisms of concern (e.g. resistant mycobacteria vs susceptible bacterial species), the application (e.g. liquid vs gaseous fumigation) and the nature of the material to be disinfected (hard surfaces vs porous materials).

The effectiveness of the disinfection procedure can be influenced by the presence of organic material, temperature, relative humidity, concentration of the disinfectant, and contact time. Each of these parameters must be carefully evaluated and defined in accordance with the properties of the disinfectant product and specific disinfection procedure.

### **Liquid effluent treatment systems:**

Liquid effluent treatment systems are typically heat-based and are used for decontaminating liquid waste streams from building sources including sinks, showers, water closets, autoclaves, washing machines and floor drains. The decontamination parameters (i.e. time and temperature) must be defined and must be effective against the microorganisms of concern. The internal temperature and pressure of the effluent tanks and decontamination time must be logged throughout the cycle. Chemical-based decontamination systems may be practical on a small-scale where smaller volumes of liquid effluent require treatment.

Decontaminated liquids released from the treatment system must meet all applicable regulations (e.g. municipal bylaws for temperature, chemical/metal content, suspended solids, oil/grease and biochemical oxygen demand).

### **Carcass disposal systems:**

Animal anatomical wastes may be incinerated. Effective incineration depends on proper equipment design; provision for the time, temperature, turbulence, and air required for complete oxidation; and careful feeding of the unit. Other technologies proven to be effective in decontamination (e.g. tissue autoclaves) present an acceptable alternative to incineration. The tissue autoclave parameters (i.e. time, temperature and pressure) must be defined and logged throughout the cycle. Provincial and territorial regulatory requirements for incinerator operation/emissions and tissue autoclave discharges must be followed.

### **Radiation:**

Gamma irradiation (e.g.  $^{60}\text{Co}$ ) can be used for the decontamination of heat-sensitive materials and is an effective means of decontaminating chemicals and solvents removed from a containment facility. The efficacy of the treatment technology is dependent on the penetration of the treated items by gamma irradiation and therefore, it is dependent upon the density of the treated substance as well as the strength of the irradiation source.

Microwave radiation is not widely used for decontamination in containment facilities. As in steam autoclaving, heat is the critical factor for eliminating viable microorganisms and the autoclave is usually the technology of choice. The factors which affect microwave treatment include frequency and wavelength of the irradiation, the duration of exposure and the destruction and moisture content of the material to be decontaminated.

Ultraviolet radiation (UV) should not be relied upon as the sole method of decontamination for materials removed from containment facilities (UV does not penetrate, microorganisms vary in susceptibility to UV). It is effective in reducing airborne and surface contamination providing the lamps are properly cleaned, maintained and checked to ensure that the appropriate intensity is being emitted.



## **7. CERTIFICATION**

For the purposes of this document, "certification" is defined as the verification of the physical construction and performance of critical containment components. Certification of containment systems may be included as part of the overall commissioning processes normally undertaken to verify that the design meets applicable codes and standards and that it has been constructed in accordance with the design intent.

To ensure the physical requirements for the intended containment level and use of the facility have been met, each laboratory and animal facility must undergo a detailed certification regimen. This requires verification and documentation of critical containment components. A complete set of "as built" and "as modified" drawings, an understanding of the intended use and work to be performed, a list of equipment requirements, all test results and an understanding of the intent of the systems operation are all required before certification can proceed.

A checklist of critical containment components to be verified during initial certification is provided below. Re-certification should also be carried out on a regular basis to monitor the system's performance. The frequency of recertification depends on a variety of factors including the frequency of use and can only be determined by monitoring the facility over time. Initially, re-certification should be carried out on an annual basis. A comparison should be made to the baseline established during initial certification. Detailed records of the certification process and test results must be maintained.

Operational protocols must also be established before work with pathogens at the specified containment level can be carried out. Training of personnel is a critical aspect of this process and may involve initial work with pathogens normally requiring a lower containment level. Users must understand the containment systems and their operation in addition to scientific procedures.

### **7.1 ROOM INTEGRITY**

Smoke testing the integrity of a containment room can be used to detect any visual leaks in the room perimeter. All joints, corners and sealed penetrations should be surveyed for leaks. Pressure decay testing the integrity of the containment room provides an indication of the tightness of the room perimeter (i.e. the ability of gases and liquids to move through the perimeter membrane and service penetrations).



The basic procedure for room pressure decay testing under vacuum is as follows:

- isolate area by closing and securing all doors, valves and bubble tight dampers at the containment barrier (avoid temporary sealing measures in doors, windows and services that would cover permanent seals and not permit their testing for leakage); plug all pressure sensor lines, e.g. magnehelic gauges
- connect a vacuum source to the room and create a negative pressure above 600 Pa negative pressure; allow room to stabilize and shut off vacuum
- dynamically trend pressure loss starting at 600 Pa until the room reaches 200 Pa negative pressure; record at 10 second intervals for minimum of 30 minutes
- record room temperature and exterior barometric pressure at beginning and end of test
- acceptance criteria outlined below must be satisfied (note: sources of air leakage can be identified using a soap or commercial detector solution on joints, corners, sealed penetrations, etc.)

Testing requirements and acceptance criteria for certification of **laboratories**:

- integrity of AP containment level 3 laboratories to be tested by smoke testing
- integrity of AP containment level 4 laboratories to be tested by pressure decay testing; rate of air leakage not to exceed 12.5 Pa/min at 500 Pa over a twenty minute period

Testing requirements and acceptance criteria for certification of **small animal facilities**:

- integrity of AP containment level 3 small animal facilities to be tested by smoke testing
- pressure decay testing of AP containment level 3 animal rooms is recommended

- pressure decay testing of AP containment level 3 animal rooms is required where animals are not housed in containment cages; rate of air leakage not to exceed 12.5 Pa/min at 500 Pa over a twenty minute period
- integrity of AP containment level 4 animal rooms to be tested by pressure decay testing; rate of air leakage not to exceed 12.5 Pa/min at 500 Pa over a twenty minute period

**Testing requirements and acceptance criteria for certification of large animal facilities:**

- room integrity of AP containment level 3 animal cubicles and PM room to be tested by smoke testing
- pressure decay testing of AP containment level 3 animal cubicles and PM room is recommended
- pressure decay testing of AP containment level 3 animal cubicles and PM room is required where non-indigenous agents are present; rate of air leakage not to exceed 12.5 Pa/min at 500 Pa over a twenty minute period
- integrity of AP containment level 4 animal cubicles and PM room to be tested by pressure decay testing; rate of air leakage not to exceed 12.5 Pa/min at 500 Pa over a twenty minute period

## **7.2 AIR HANDLING SYSTEM**

Various components of a containment room's air handling system require certification. Manufacturer's requirements for airflows for BSCs must be met. Particle challenge testing of HEPA filters must be performed to ensure they do not contain leaks in the filter media, the bond between the media and frame, or around the frame gasket and support. Ductwork systems should be pressure decay tested to confirm that specified leakage rates are not exceeded. The American Society of Mechanical Engineers (ASME) Standard N510 *Testing of Nuclear Air Treatment Systems*, 1989, gives procedures for testing the leak-tightness of ducts and plenums. Room pressure control systems must operate as specified (e.g. ensure negative pressures are maintained).

**Biological safety cabinets:**

- all biological safety cabinets to be tested in-situ in accordance with CSA Z316.3-95, *Biological Containment Cabinets: Installation and Field Testing* (1995)
- interlocks (i.e. between BSC motor and exhaust fan) to be tested for specified response
- alarms to be tested for detection of BSC and/or exhaust fan failure by simulation of alarm conditions

**Fume hoods:**

- all fume hoods and associated exhaust systems to be tested in-situ in accordance with CSA Z316.5-94, *Fume Hoods and Associated Exhaust Systems* (1994)

**HEPA filters and filter housings:**

- integrity of HEPA filters installed into supply and exhaust ductwork to be tested in-situ by particle challenge testing using the scanning method; particle penetration not to exceed 0.01%
- integrity of HEPA filter housings with inlet and outlet bubble tight dampers installed into supply and exhaust ductwork to be tested in-situ by pressure decay testing in accordance with ASME N510 *Testing of Nuclear Air Treatment Systems* (1989); rate of air leakage not to exceed 0.2% of vol/min at 2500 Pa
- in-line filters (e.g. plumbing vent lines, gas supply lines, autoclave exhaust ducts) to be tested by particle challenge testing; filter efficiency to be equivalent to HEPA

**Supply and exhaust ductwork:**

The basic procedure for ductwork pressure decay testing as outlined by ASME N510, *Testing of Nuclear Air Treatment Systems*, is as follows:

- isolate and seal the ductwork to be tested
- attach a temperature and pressure sensor to the test system to indicate interior temperature and pressure
- pressurize ductwork with air to at least 500 Pa and allow to stabilize
- record temperature inside the ductwork at beginning and end of the test
- dynamically trend pressure loss; record at 10 second intervals for minimum of 30 minutes
- the acceptance criteria outlined below must be satisfied (note: sources of air leakage can be identified using a soap or commercial detector solution on joints, corners, sealed penetrations, etc.)

**Ductwork testing requirements and acceptance criteria:**

- for AP containment level 3 and 4 laboratories, SA rooms, and LA cubicles and PM rooms, supply and exhaust ductwork between containment room perimeter and bubble tight damper to be in accordance with Sheet Metal and Air Conditioning Contractors National Association (SMACNA) Seal Class A, *HVAC Air Duct Leakage Test Manual* (1985)
- for AP containment level 3 SA rooms where animals are not housed in containment caging, LA cubicles and PM rooms where non-indigenous agents are present, supply and exhaust ductwork between containment room perimeter and bubble tight damper to be tested in-situ by pressure decay testing; rate of air leakage not to exceed 0.2% of duct vol/min at 500 Pa
- for AP containment level 4 laboratories, SA rooms, and LA cubicles and PM rooms, supply and exhaust ductwork between containment room perimeter and bubble tight damper to be tested in-situ by pressure decay testing; rate of air leakage not to exceed 0.1% duct vol/min at 500 Pa



**Air balancing and pressurization relationships:**

- inward directional airflow to be visually demonstrated by smoke testing
- pressurization relationships across a containment barrier (e.g. at airlocks, laboratories, animal rooms and cubicles, corridors) to be verified; minimum of 25 Pa is recommended across a containment barrier

**HVAC control systems:**

- control systems to be tested for fail-safe operation by failure of system components (e.g. door is open, fan failure, electrical failure, BSC failure); room/cubicle positive pressurization to be prevented
- control system performance verification should include speed of response, accuracy, and repeatability
- audible alarms to be tested for detection of positive pressurization and air handling systems failure by simulation of alarm conditions

**7.3 LABORATORY SERVICES**

- water supply backflow prevention to be tested in accordance with CAN/CSA-B64.10-94, *Manual for the Selection, Installation, Maintenance, and Field Testing of Backflow Prevention Devices* (1994)
- backflow prevention for other services (e.g. gases) to be verified to ensure system will operate as specified
- the water flow rate and temperature of eyewash stations and emergency showers to be verified in accordance with ANSI Z3858.1, *Emergency Eyewash and Shower Equipment*
- compressed breathing air and systems to be verified in accordance with CAN3-Z180.1-M85, *Compressed Breathing Air and Systems* (1994)
- operation of positive-personal protective equipment (i.e. suit) to be tested in-situ to ensure suit will operate as specified

- water and chemical shower systems to be tested to ensure systems will operate as specified
- cage washers to be tested to ensure system will operate as specified; water temperature of final rinse to be at least 82°C
- emergency electrical generator to be tested under appropriate load conditions to ensure systems will operate as specified
- security systems (e.g. controlled access, closed circuit TV) to be verified to ensure system will operate as specified
- communication and electronic paper transfer systems (e.g. intercom, telephone, fax) to be verified to ensure system will operate as specified
- all sterilization systems (e.g. autoclaves, liquid effluent treatment systems) to be verified for operation as specified and to be microbiologically tested using representative loads; for technologies based on heat - using *Bacillus stearothermophilus* spores; for technologies based on chemicals - using *Bacillus subtilis* spores
- drains and associated piping leading to liquid effluent treatment systems (including associated vent lines) to be tested in accordance with Section 3.6 of the Canadian Plumbing Code, *Testing of Drainage and Venting Systems* (1990); pressure for air test on drainage system shall be at a factor of safety beyond standard code requirements of 35 kPa (e.g. 2 X code)
- all disinfection systems (e.g. dunk tanks, fumigation chambers) to be verified for operation as specified and microbiologically tested using representative loads; resistance of test organism to be representative of organisms likely to be encountered

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