

UNIVERSITY OF WESTERN ONTARIO
ANIMAL CARE AND VETERINARY SERVICES

**PROTECTION PROCEDURES
FIFTH FLOOR EXCLUSION FACILITY - Health Sciences**

Standard Operating Procedure #168-03

1 INTRODUCTION

The exclusion facility is designed to contain nucleus breeding colonies and immunodeficient, transgenic or knockout mice separate from conventional housing in the Health Sciences animal facility. The purpose is to reduce the risk of the mice contracting murine pathogens.

The exclusion facility on the fifth floor includes rooms 5000A to 5000I and the associated corridors, TA503 and TA504. This area has controlled access with special procedures for entry, exit, material handling, and animal handling (see Appendix A).

The exclusion facility airflow is positive to the surrounding area, with the animal rooms more positive than the hallway TA504. Air is directed from the animal rooms to TA504. The animal rooms are “cleaner” than the hallways.

- 1.1 **Personnel must enter the Exclusion unit prior to entering the Health Sciences and/or Conventional or Inclusion rodent facilities.**
- 1.2 **Personnel must not have entered any non-Health Sciences, ACVS animal facility within 48 hours of entering the Exclusion Barrier.**
- 1.3 Access to this facility is only between 9:00 a.m. and 5:00 p.m. Monday to Friday, unless special arrangements are made with the Facility Manager.
- 1.4 Animals cannot return to the Exclusion Barrier facility once removed.
- 1.5 Animal Care Staff attend to Exclusion Barrier housed animals first thing in the morning.
- 1.6 Items brought into the exclusion barrier **must** be wiped off with Clidox prior to crossing the yellow line (please refer to 2.9)

2 ENTRY AND EXIT

Animals leaving the barrier unit may never return.

- 2.1 Research personnel entering the Exclusion Barrier within 48 hours of entering "on campus" facilities (Health Sciences Conventional or Robarts Research Institute) must shower at home with a complete change of clothes prior to entry.
- 2.2 ACVS personnel may enter the Exclusion Unit following a shower and a complete change of clothes after work within the conventional area(s) of Health Sciences.
- 2.3 ACVS personnel will wear autoclaved blues within the Exclusion.
- 2.4 ACVS personnel will wear dedicated Exclusion Barrier shoes where possible, otherwise double booties as per 2.7.3.
- 2.5 Only those research personnel with security access will be permitted entry. The number of people entering the barrier facility should be kept to a minimum.
- 2.6 To reduce traffic into the Exclusion Barrier, researchers are encouraged to hire ACVS to perform routine procedures and monitoring.
- 2.7 Entry to the barrier facility is only permitted via the door by the Dental Sciences Building elevator

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- (due to airborne pathogens in the conventional facility the Exclusion Barrier facility must not be accessed from within the Health Sciences Animal Care Facility). Please refer to Attachment A.
- 2.8 To access, exit the elevator on the 5th floor. Once off the elevator, proceed north (turn right) along corridor TA501 to Barrier entrance.
 - 2.9 No personal items are to be brought into corridor TA503 (including carts from your lab and lab coats).
 - 2.7.1 Dedicated Exclusion carts are provided exclusively for use within the barrier area (see Section 2.8 for guidelines on the use of carts).
 - 2.7.2 Minimal hooks are provided by exclusion entrance exterior for lab coats, so please come without one if possible.
 - 2.7.3 A pair of booties must be put on over street shoes prior to entering the Barrier facility if dedicated Exclusion barrier shoes are not worn (as per 2.4).
 - 2.8 If you have brought a cart from your lab and are transporting items into the barrier facility, the cart must remain in the corridor TA501 (outside barrier). Items taken off the cart must be placed on the floor inside corridor TA503 (the entrance to the barrier facility) near to but not crossing over the yellow line.
 - 2.9 All materials including Robarts mouse transfer boxes must be wiped down with 1:5:1 Clidox prior to crossing the yellow line. Clidox is made up every 24 hours by Animal Care staff (check date and time posted by the Clidox), and located in a sealed container located on the shelf unit next to the yellow line.
 - 2.10 Once all items are within the barrier area, enter corridor TA503. In this corridor you will don protective outer wear before crossing the yellow line. First put on a mask (a beard cover if necessary), then cap and one pair of gloves.
 - 2.11 Don safety glasses and wipe transfer boxes and all other material which enters the exclusion area with cloth soaked in Clidox and place over yellow line. Remove safety glasses and return to shelf unit.
 - 2.12 If dedicated exclusion barrier shoes are not being worn, take a second pair of booties and place one on your right foot, ensuring it contacts the floor over the yellow line. Repeat with your left foot.
 - 2.12 If dedicated exclusion barrier shoes **are** worn, take a 1st pair of booties and place one on your right foot, ensuring it contacts the floor over the yellow line. Repeat with your left foot.
 - 2.13 **Always check protective outer wear for holes and tears. These should be thrown out and not worn.**
 - 2.1.3.1 If you require a cart you will find one in corridor TA504. This cart MUST NOT cross the yellow line in corridor TA503. Push the cart close to the yellow line and transfer the materials wiped with CLidox from the floor across the yellow line to the cart taking care not to cross over the line.
 - 2.15 From this corridor proceed directly to your room. Carts must always remain in the hall. Items can be transferred to your animal room from the hallway to the floor of the animal room beyond the yellow line.
 - 2.16 Once in your animal room, stay within the yellow square by the entrance door. Put your right foot in a tyvek suit making sure you cross over the yellow line before touching the ground with your foot. Repeat with your other foot. Ensure Tyvek is completely zipped up.
 - 2.16.1 Room 5000A has an anteroom prior to the animal room. The anteroom will be the equivalent of the yellow square within this SOP.
 - 2.17 If handling animals, don a second pair of gloves, taping the wrists to your tyvek suit. The taped-off square cannot be re-entered until you are ready to exit the room. Proceed with your work

3 ANIMAL PROCEDURES

- 3.1 Clidox is (MUST BE MADE FRESH DAILY AND USED WITHIN 24 HOURS OF BEING

PREPARED) to be used exclusively in the Biological Safety Cabinet. Exclusion Barrier Block rooms will have the Clidox premixed and supplied to the room every a.m. by ACVS staff.

3.2 BIOLOGICAL SAFETY CABINET SETUP

- 3.2.1 Wet towels and one j-cloth in the plastic container containing the activated CLIDOX solution. Lift and pull the glass sash of Biological Safety Cabinet. Wipe all surfaces inside the cabinet (except the vent in the ceiling) with the j-cloth. Be sure to wipe all inner walls, work surface and the interior of the glass sash. Put down sash. NOTE: ANY TIME THE SASH IS LIFTED AND RETURNED TO ITS ORIGINAL POSITION THE AIRFLOW MUST BE RE-ESTABLISHED. WAIT AT LEAST 1 MINUTE BEFORE CONTINUING WITH WORK INSIDE THE CABINET.
- 3.2.2 Cover **entire** worksurface with the Clidox soaked towels provided and place j-cloth inside cabinet. TOWELS AND J-CLOTH MUST REMAIN MOIST AT ALL TIMES.
- 3.2.3 Pour some of the Clidox solution into the squeeze bottle marked "Clidox 1:5:1". Wipe outer surface of squeeze bottle with damp j-cloth and place in cabinet. Use this squeeze bottle to ensure work surface towels remains moist at all times.
- 3.2.4 Everything must be wiped with Clidox as you introduce items not enclosed in a sterile container (forceps/scissors/scales/heat lamps) into the Biological Safety Cabinet. Wipe each with a damp j-cloth, followed by your gloved hands (only exception is 3.3.5).
- 3.2.5 After handling the outside of the sterile containers containing supplies (water bottle and food container) hands must be wiped with Clidox prior to handling the sterile contents.

3.3 CAGE SETUP

In order to allow bagged caging material to be placed into the cabinet, the sash must be lifted. Allow for the airflow to be re-established (after returning the sash to the original position) one minute before continuing with work inside the cabinet.

- 3.3.1. Extra autoclaved caging supplies are kept within individual rooms. Minimum supplies will be kept in 5000H, and once removed from this area can not be returned.
- 3.3.2. All caging supplies and other supplies within the Biological Safety Cabinet must be covered (water bottles and food containers) prior to opening the sash to introduce more supplies.
- 3.3.3 Locate green bag containing sterile shoebox caging and place inside Biological Safety Cabinet (DO NOT WIPE OUTER SURFACE OF GREEN BAGS)
- 3.3.4 Untie bag and wipe hands before touching the contents inside of bags (cages/filter tops, etc.)
- 3.3.5 Remove the number of cages that you require.
- 3.3.6 Tie up green bag in the hoods and return to shelf.
- 3.3.7 Repeat 3.3.3 to 3.3.8 procedure for the filter tops/wires/water bottles.
- 3.3.8 Locate the food container and place in Biological Safety Cabinet. Wipe all outer surfaces of container and rewipe hands with Clidox-soaked j-cloth.
- 3.3.9 WIPE HANDS EVERYTIME THEY ARE RE-INTRODUCED INTO THE BIOLOGICAL SAFETY CABINET.
- 3.3.10 Assemble cages, and ensure the water bottle stopper is secure (prevent flooding). Check to make sure water comes out of sipper tube by inverting bottle, placing finger on tip and watching for air bubbles run in the bottle.
- 3.3.11 Remove lid of food container, scoop out food and place in hopper. Return scoop and lid to food container.

3.4 MOUSE MANIPULATION/CAGE CHANGING

In order to allow the caging material to be placed into the cabinet the sash must be lifted. To allow for the airflow to be re-established (after returning sash to the original position) wait one minute before continuing with work inside the cabinet.

- 3.4.1 For changing cages, establish a "dirty" area and "clean" area in Biological Safety Cabinet. ONCE DIRTY CAGING IS PLACED INSIDE BIOLOGICAL SAFETY CABINET HANDS MUST BE WIPED WITH CLIDOX SOAKED J-CLOTH AFTER TOUCHING EXTERIOR SURFACES OF CAGING. (I.E. such as lifting of the cage top and before continuing with any mouse manipulations).
- 3.4.2 Remove forceps from table/shelf, wipe with J-cloth and place in Biological Safety Cabinet.
- 3.4.3 Retrieve soiled cage from rack. Remove cage card and place in area outside Biological Safety Cabinet. Set soiled cage down in designated "dirty" area in Biological Safety Cabinet. Repeat 3.2.3.
- 3.4.4 Rewipe hands with Clidox. Remove filter top from previously assembled clean cage and flip it upside down so its exterior surface rests on the Clidox soaked towels.
- 3.4.5 Rewipe hands with Clidox and slide the wire hopper lid toward the back of the cage creating an opening just big enough to transfer the animals through.
- 3.4.6 Remove the filter top from the dirty cage and flip upside down so the exterior surface rests on the Clidox soaked towels.
- 3.4.7 Rewipe hands with Clidox and lift up the wire hopper lid from the soiled cage. Re-wipe forceps with Clidox-soaked j-cloth.
- 3.4.8 Using the forceps, pick up the mice by the base of the tail, or scruff of the neck if young, and transfer into the clean cage. Re-wipe forceps with Clidox-soaked j-cloth.
- 3.4.9 Slide the clean wire hopper back into place on the clean cage. Put sufficient food in the hopper, place a fresh water bottle in the holder and check that stopper is secure.
- 3.4.10 Put the clean filter back on the cage. Remove the cage from the work area and return it to its original place on the rack and replace the cage card.
- 3.4.11 Rewipe hands with Clidox and replace filter top on dirty cage and remove from Biological Safety Cabinet. Place intact cage on floor.
 - 3.4.11.1 All cages must have bedding dumped out after cage changing is complete and prior to removing cages from exclusion facility. Cages removed from facility must go to Room 611 (cagewasher room).
- 3.4.12 For vented racks, please spray ports using spray bottle with Quatricide PV-15 prior to docking new cages (such as after weaning).
 - 3.4.12.1 Repeat all procedures from 3.4.4 to 3.4.12 until each cage has been changed.
 - 3.4.12.2 WIPE HANDS EVERY TIME THEY ARE RE-INTRODUCED INTO THE BIOLOGICAL SAFETY CABINET.
- 3.4.13 If a mouse escapes and remains within the BSC work station, transfer into the clean cage.
- 3.4.14 If a mouse escapes from the work station, capture it and put it in an appropriate new microisolator cage, identify with a cage card and notify the Facility Manager or Leadhand. It cannot be re-housed with its cagemates.

3.5 CLOSING DOWN BIOLOGICAL SAFETY CABINET (BSC)

- 3.5.1 Remove all towels from hood and be sure to remove any bedding by shaking towels out into garbage can, then squeeze out excess fluid from towels into sink.
- 3.5.2 Rinse towels thoroughly with water and wring out excess in the sink. Fold towels and hang them over the edge of shoebox or handles of cart. Place all items back on

table/shelves.

- 3.5.3 The sash of the Biological Safety Cabinet to clean the inside surface.
- 3.5.4 Using the squeeze bottle with “Clidox 1:5:1 solution” squirt fresh Clidox solution onto work surface. Using your j-cloth wipe all surfaces inside the cabinet (including the inside surface of the sash).
- 3.5.5 Squirt alcohol onto the work surface of the clean bench. Saturate paper towel with alcohol and wipe all surfaces of the cabinet to distribute the alcohol (neutralizes the Clidox to prevent damage to the stainless steel).
- 3.5.6 Clean glass visor with Windex and paper towel.
- 3.5.7 Wait a few moments, then turn off light and fan of cabinet. Do not leave UV light on.

3.6 ANIMAL PROCEDURES WITHIN THE BARRIER FACILITY

- 3.6.1 All manipulations (injections and surgeries, blood or urine collection) must be carried out within the Biological Safety Cabinet. **Procedures must be recorded in the animal health binder within the room for the purpose of accurate health monitoring of animals.**
- 3.6.2 Notify a Veterinary Services technician or veterinarian of any animal concerns (sick, injured, deaths or post mortem requests) and record in Animal Health Binder within the room.
- 3.6.3 Euthanasia may only be performed inside the Biological Safety Cabinet using barbiturate overdose. Contact ACVS if any other euthanasia techniques are required.

3.7 LEAVING THE BARRIER FACILITY

- 3.7.1 To leave animal room, first untape wrists and remove outer gloves. Discard in provided garbage
- 3.7.2 Remove Tyvek.
- 3.7.3 Place booted foot into yellow square as you remove each leg.
 - 3.7.3.1 Room 5000A has an anteroom prior to the animal room. The anteroom will be the equivalent of the yellow square within this SOP.
- 3.7.4 Ensure booties are still on your shoes while stepping back into the yellow square. Discard outer gloves in the garbage provided. You should be left wearing your cap, mask, beard cover (if necessary), booties and first pair of gloves.
- 3.7.5 Leave the animal room. Proceed directly to corridor TA503, and approach but do not cross the yellow line.
- 3.7.6 Fax machine can be used to fax any paperwork out or into the facility - located in 5000H.
- 3.7.7 Remove one boot cover, making sure your foot crosses over the yellow line before touching the ground. Repeat with your other foot.
- 3.7.8 Next, remove cap, mask, beard cover (if necessary), and your gloves last. Discard in labeled containers provided.
- 3.7.9 Exit the barrier area the same way you entered by pressing the blue button on the wall.
- 3.7.10 Retrieve any personal effects, lab carts, etc. from the hallway.
- 3.7.11 Remove first booties and place in green garbage can outside the Exclusion barrier facility main entrance.

Attachment A
Map of Area.

