IACUC Standard Operating Procedure SOP-05-2017: Isolation of Hazardous Chemicals/Agents in Use in Rodents

This document describes the procedures used when handling rodents exposed to hazardous chemicals/agents, as well as their cages, contact bedding, cage contents and waste. This document should be used in conjunction with Protocol specific Appendix Ds which must be available in the room where the chemicals are in use.

Animals treated with following hazardous chemicals/agents, BrdU, EdU, STZ, Tamoxifen, Doxorubicin, Carbon tetrachloride, DEN, and Sodium Arsenite are currently designated to require housing in the Chemical Isolation Room or other IACUC approved satellite chemical isolation space while the hazard is present. Furthermore, new chemical agents will be reviewed on a case-by-case basis to determine if they require isolation. Risk assessment will include hazard type, routes of exposure, routes of administration, routes of excretion and dose. Approved satellite chemical isolation spaces must be clearly posted with an approved “Caution Satellite Chemical Isolation Room” sign provided by EHS which will stay posted as long as the hazard is in use in that space.

Access to chemical isolation spaces will only be granted to trained personnel; training on this SOP must be completed prior to access being granted. Training will be provided by designated Animal Care Service (ACS) staff.

In all circumstances, animal manipulations and handling of cages will be conducted solely by trained personnel (ACS or research) utilizing appropriate engineering controls and personal protective equipment (PPE). Specifically, during the period that the hazard is present (72 hours after the final administration unless otherwise described in an IACUC approved Appendix D), as denoted by the presence of the orange card, handling of animals and cages will be limited to research staff when necessary for the research procedures, or ACS staff in the event of an emergency. When the hazard is present (as denoted by the orange card), all cage changes and husbandry should be performed by research staff; trained ACS staff may provide emergency care as needed. When the hazard is no longer present (after the clearance period and as denoted by the movement of the orange card to the back of the series of cards) either ACS or research staff may handle animals and cages and provide routine husbandry. In addition to those duties described above, ACS staff will also be expected to conduct health checks and restock supplies, as needed.

Once the hazard is no longer present at the cage level, animals may be able to be moved back to regular housing in the general population. This movement will depend on two factors: (1) health status as determined by ACS, and (2) risk assessment as determined by EHS. In some cases animals will be free to move out of isolation after a 72 hour clearance period has elapsed since the final administration of the chemical/agent, in other cases permanent isolation (i.e. until termination) of the animals will be required. This clearance period will be based upon the EHS risk assessment and will be documented in the approved Appendix D and associated Appendix K. Once the clearance period has elapsed, animals must be transferred...
to a clean cage by research staff and the orange cage card must be moved to the back of the series before the animal can be moved out of isolation.

I. Chemical Isolation Room Entry Procedure

All personnel entering the Chemical Isolation Room must don the following Personal Protective Equipment in the anteroom:

Disposable gown, bonnet, extra pair of shoe covers, nitrile gloves, and safety glasses. Personnel handling animals and/or performing husbandry chores are to don 6 ml purple nitrile or double standard nitrile gloves.

II. Procedures for Hazards That Require a 72 Hr Hold Period (e.g. tamoxifen, BrdU, EDU)

For those chemicals/agents that require a 72 hour hold (as described in Appendix D and the associated Appendix K) the following procedures are indicated:

A. Transfer of animals
   1. Animals from the housing room will be moved to the Chemical Isolation Room in clean caging compatible with the housing rack in the isolation room. Original housing cage card(s) will accompany the animal(s) coming to the room.

B. Dosing the animals
   1. Administration of hazardous chemical agents will be performed in a Chemical Fume Hood (in the Chemical Isolation Room available in anteroom 1K) or other approved engineering controls (such as Animal Transfer Station (ATS) or Biosafety Cabinet (BSC) in the satellite areas – refer to Appendix D for each protocol.
   2. All needles, syringes, and vials that come in contact with these agents will be disposed of in an approved Sharps container. A Sharps container will be located in the Chemical Fume Hood in the Chemical Isolation room. Place a yellow “Do Not Autoclave” sticker on the sharps container.
      a. For satellite locations a sharps container must be placed in the approved engineering control during procedures. Place a yellow “Do Not Autoclave” sticker on the sharps container.
   3. If dosing is performed in the water, the water bottle must be labeled with a “Dispose as Hazardous Waste” sticker; unless otherwise indicated in Appendix D, only the contents should be disposed of as hazardous waste.
   4. After administration of the hazardous chemical agent, decontaminate all surfaces with appropriate disinfectant as identified in Appendix D of the research protocol.
   5. After administration of hazardous chemical agents, place the rodents in a clean cage (either back
to the transfer cage, or a new cage in the case of multiple administrations over time). Provide water bottle and food; wipe the outside of the cage with appropriate disinfectant and return the cage to the cage rack.

6. Spills of hazardous chemical agents should be cleaned with absorbent paper towels and appropriate disinfectant as identified in Appendix D of research protocol. Large spills are unlikely as large amounts of chemical agents are not permitted in this facility.
7. Dispose of all waste generated from spills through UConn EHS.

C. Orange Cage Cards
   Fill out the Orange Cage Card (used for chemical hazards). As prompted by the card, the researcher must:
   1. Provide the Agent and Hazard Identification (refer to Appendix D)
   2. Complete the date and time of administration
   3. Complete the “Special Handling Required Until” date and time
   4. Provide the initials of the person administering the Chemical Agent
   5. Provide contact information (cell phone) of person administering the Chemical Agent
   6. For repeated/multiple injections of the Chemical Agent each administration should be documented on the card; additional cards will be needed when more than 5 injections or doses are administered.
   7. Orange cards must remain with the cage until the end of the study; placed either at the front when the hazard is present, or moved to the back of the series of cards when the hazard is no longer present.

D. Handling Cages, Bedding, Water Bottles and Animals after Administration of Hazardous Chemical Agents
   1. Animals will be housed in the Chemical Isolation Room for a minimum of 72 hours (unless otherwise described in an IACUC approved Appendix D) after the final administration of hazardous chemicals/agents; during that period in which the hazard is present (as denoted by the orange card), handling of animals and cages will be limited to research staff when necessary for the research procedures, or ACS staff in the event of an emergency. When the hazard is present (as denoted by the orange card), all cage changes and husbandry should be performed by research staff; trained ACS staff may provide emergency care as needed. When the hazard is no longer present (after the clearance period and as denoted by the movement of the orange card to the back of the series of cards) either ACS or research staff may handle animals and cages and provide routine husbandry. Please note, in the case of repeated administration of hazardous chemicals/agents, husbandry and cage changes will be performed by research staff, as the hazard will be present throughout that period.
   2. The cages and animals will be handled in the Animal Transfer Station (ATS) or other appropriate engineering control such as a biosafety cabinet (BSC). Verify blower motor is on before manipulations.
3. Research staff will conduct husbandry as needed when the hazard is present. Cage changing technique will be demonstrated by trained ACS staff as a part of training on this SOP.

4. After each use of the ATS, decontaminate all surfaces with an appropriate disinfectant.

5. After the 72 hour clearance period, animals should be transferred to a clean cage, the orange card should be moved to the back of the series, and animals maybe moved back into regular housing areas. **NOTE:** the movement of animals back into the general population is dependent upon: (1) risk assessment as determined by EHS, and (2) health status of the individual animals as determined by ACS.

6. Trained staff will dispose of soiled cages and their contents as follows:
   a. Disposal will take place in the ATS or other appropriate engineering control such as a BSC. Supplies, yellow bags, tape and yellow “do not autoclave” stickers will be provided in the room.
   b. Entire cage, including bedding, clean water and agent-laced chow, will be placed in a yellow plastic bag. Maximum of two (2) cages per bag.

   **(NOTE - Agent laced water will be emptied into waste containers labeled with the agent. These bottles will have been labeled with a “Dispose as Hazardous Waste” sticker which will alert staff to the waste requirement. Once emptied, stickers will be removed and the empty bottles will be placed in a yellow bag. Wastewater will be arranged for collection and disposed of as hazardous waste through EHS by completing the online Chemical Waste Pickup Form.)**
   c. Twist the top of the bag and seal with tape.
   d. Attach a yellow “Do Not Autoclave” sticker onto the bag and write the administered chemical name on the bag with marker provided in room.
   e. Decontaminate all surfaces of ATS with appropriate disinfectant.
   f. Bags will be left in the isolation room to be removed only by trained ACS staff. ACS staff will deliver yellow bags to the dirty side of the cage wash and place them on the dedicated cart.

7. Trained ACS staff will dump the yellow bags of soiled cages from the chemical isolation room (identified by a yellow “Do Not Autoclave” sticker) in the HEPA filtered dumping station wearing N-95 masks and safety glasses; clean water will be dumped in the garbel. The waste will be placed in red waste bags and transferred to a Regulated Medical Waste box-bag unit for collection by UConn EHS.

8. **Euthanasia/Carcass Disposal:** Trained staff will perform euthanasia in the anteroom. ACS will make arrangements to have a carbon dioxide tank and chamber present. Alternatively, the animals may be taken directly to the research lab for immediate euthanasia. Transfer the animals to a clean cage for transport to the lab. If the 72 hour clearance period has elapsed, the animals can be transported to the lab without further special handling required. If the animal must be transported for euthanasia within the 72 hour clearance period, research staff must take a yellow bag and “Do Not Autoclave” sticker to ensure the waste disposal and labeling procedures described in Step 6 above are followed for bagging soiled cages. Procedures in “D” above must be followed for handling soiled bedding and cages. The one change in procedure will be that
research staff must deliver the yellow bags to the dirty side of the cage wash and place them on the dedicated cart. If there will be carcasses of injected animals, the carcasses will be handled by research staff and they will be placed in a small red carcass bag and delivered to the necropsy room and placed in the carcass freezer until final collection by UConn EHS.

E. Handling spilled cages of animals that have been dosed within the restricted period.
   1. Research staff must exit the Chemical Isolation Room and place a Do Not Enter sign on the door (provided in the anteroom).
   2. Notify ACS supervisor (contact numbers are listed on door).
   3. Remove PPE and exit the suite.

III. Procedures for Hazards That Require Permanent Isolation

Some chemicals/agents will require permanent isolation (as described in Appendix D and the associated Appendix K). **NOTE:** In this category of hazards there can be two waste streams: (1) Regulated Medical Waste (RMW) is the most common and uses the yellow bags with yellow “Do Not Autoclave” stickers in the isolation rooms, (2) Hazardous Waste is less common and could include liquid wastes from agent laced water and/or bedding and animals. If the EHS risk assessment of the chemical/agent in use requires disposal through the Hazardous Waste Stream then the water bottle and/or cage card will be labeled with the “Dispose as Hazardous Waste” sticker; this indicates that the entire contents of the bottle and/or cage should be disposed of as Hazardous Waste.

For chemicals/agents requiring permanent isolation, the following procedures are indicated:

A. Transfer of animals
   Animals from the housing room will be moved to the Chemical Isolation Room in clean caging compatible with the housing rack in the isolation room. Original housing cage card(s) will accompany the animal(s) coming to the room.

B. Dosing the animals
   1. Administration of hazardous chemical agents will be performed in a Chemical Fume Hood (in the Chemical Isolation Room available in anteroom 1K) or other approved engineering controls such as Animal Transfer Station (ATS) or Biosafety Cabinet (BSC) (in the satellite areas – refer to Appendix D for each protocol).
   2. All needles, syringes, and vials that come in contact with these agents will be disposed of in an approved Sharps container. A Sharps container will be located in the Chemical Fume Hood in the Chemical Isolation room. **Place a yellow “Do Not Autoclave” sticker on the sharps container.**
      a. For satellite locations a sharps container will need to be placed in the approved engineering control during procedures. **Place a yellow “Do Not Autoclave” sticker on the sharps**
container.
3. If dosing is performed in the water, the water bottle must be labeled with a “Dispose as Hazardous Waste” sticker; unless otherwise indicated in Appendix D only the contents should be disposed of as hazardous waste.
4. After administration of the hazardous chemical agent, decontaminate all surfaces with appropriate disinfectant as identified in Appendix D of the research protocol.
5. After administration of hazardous chemical agents, place the rodents in a clean cage. Provide water bottle and food, wipe the outside of the cage with appropriate disinfectant and return the cage to the cage rack.
6. Spills of hazardous chemical agents should be cleaned with absorbent paper towels and appropriate disinfectant as identified in Appendix D of research protocol. Large spills are unlikely as large amounts of chemical agents are not permitted in this facility.
7. Dispose of all waste generated from spills through UConn EHS.

C. Orange Cage Cards
Fill out the Orange Cage Card (used for chemical hazards). As prompted by the card, the researcher must:
1. Provide the Agent and Hazard Identification (refer to Appendix D)
2. Complete the date and time of administration
3. Under “Special Handling Required Until” indicate the following: “until project termination”.
4. **NOTE:** Cage cards for projects requiring disposal of bedding and animals through the Hazardous Waste stream will also be labeled with a “Dispose as Hazardous Waste” sticker
5. Provide the initials of the person administering the Chemical Agent
6. Provide contact information (cell phone) of person administering the Chemical Agent
7. For repeated/multiple injections of the Chemical Agent each administration should be documented on the card; additional cards will be needed when more than 5 injections or doses are administered.
8. Orange cards must remain in the front of the series of cards with the cage until the end of the study.

D. Handling Cages, Bedding, Water Bottles and Animals after Administration of Hazardous Chemical Agents
1. Animals under this category will be housed in the Chemical Isolation Room until project termination. Routine husbandry will be performed by trained research staff while the hazard is present.
2. The cages and animals will be handled in the Animal Transfer Station (ATS) or other appropriate engineering control such as a BSC. Verify blower motor is on before manipulations.
3. Research staff will conduct routine husbandry as needed when the hazard is present. Cage
changing technique will be demonstrated by trained ACS staff as a part of training on this SOP.

4. After each use of the ATS, decontaminate all surfaces with an appropriate disinfectant.

5. Trained staff will dispose of soiled cages and their contents as follows:
   a. Disposal will take place in the ATS. Supplies, yellow bags, tape and yellow “do not autoclave” stickers will be provided in the room.
   b. Entire cage, including bedding, clean water and agent-laced chow, will be placed in a yellow plastic bag. Maximum of two (2) cages per bag.
      i. NOTE - Agent laced water will be emptied into waste containers labeled with the agent. These bottles will have been labeled with a “Dispose as Hazardous Waste” sticker which will alert staff to the waste requirement. Once emptied, stickers will be removed and the empty bottles will be placed in a yellow bag. Wastewater will be arranged for collection and disposed of as hazardous waste through EHS by completing the online Chemical Waste Pickup Form.
   c. Twist the top of the bag and seal with tape.
   d. Attach a yellow “Do Not Autoclave” sticker onto the bag and write the administered chemical name on the bag with marker provided in room.
   e. Decontaminate all surfaces of ATS with appropriated disinfectant.
   f. Bags will be left in the isolation room to be removed only by trained ACS staff. ACS staff will deliver yellow bags to the dirty side of the cage wash and place them on the dedicated cart.

   NOTE: Projects requiring Hazardous Waste disposal will have special instructions as to handling cages, bedding waste, water bottles and/or the animal carcasses. Refer to the project specific Appendix D.

6. Trained ACS staff will dump the yellow bags of soiled cages from the chemical isolation room (identified by a yellow “Do Not Autoclave” sticker) in the HEPA filtered dumping station wearing N-95 masks and safety glasses. The waste will be placed in red waste bags and transferred to a Regulated Medical Waste box-bag unit for collection by UConn EHS.

7. Euthanasia/Carcass Disposal: Trained staff will perform euthanasia in the anteroom. ACS will make arrangements to have a carbon dioxide tank and chamber present. Alternatively, the animals may be taken directly to the research lab for immediate euthanasia. Transfer the animals to a clean cage for transport to the lab. If the animal must be transported to the lab for euthanasia, research staff must take a yellow bag and “Do Not Autoclave” sticker to ensure the waste disposal and labeling procedures described in Step 5 above are followed for bagging soiled cages. Procedures in “D” above must be followed for handling soiled bedding and cages. The one change in procedure will be that research staff must deliver the yellow bags to the dirty side of the cage wash and place them on the dedicated cart. If there will be carcasses of injected animals, the carcasses will be handled by research staff and they will be placed in a small red carcass bag and delivered to the necropsy room and placed in the carcass freezer until final collection by UConn EHS unless otherwise specified in the protocol specific Appendix D.
E. **Handling spilled cages of animals that have been dosed within the restricted period.**
   1. Research staff must exit the Chemical Isolation Room and place a Do Not Enter sign on the door (provided in the ante room).
   2. Notify ACS supervisor (contact numbers are listed on door).
   3. Remove PPE and exit the suite.
The Undersigned acknowledge having read this SOP-05-2017, received demonstrations on procedures, demonstrated the procedures, and had the opportunity to ask questions regarding tasks associated with working in the Chemical Isolation Room.

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