STANDARD OPERATING PROCEDURES

DIVISION OF COMPARATIVE MEDICINE
UNIVERSITY OF SOUTH FLORIDA

4. Racks of bottles filled with acidified drinking water must be validated as sterile in accordance with SOP 1002 with an Integrator strip present with each autoclaved load, and the racks of bottles **covered** prior to transport to isolation**.**

5. Ensure that all caging and water bottles have been properly autoclaved by checking Integrator strips present with each autoclaved load of cages or water bottles. Refer to Monitoring Autoclave Sterilization (SOP 1002), Autoclave Sterilization (SOP 1006), and Monitoring Steam Sterilization of Liquids (SOP 1013) for proper sterilization techniques. If steam exposure indicator has not changed, do not use cages/supplies and notify your supervisor immediately.

6. Whenever covered autoclaved transport racks of microisolators or covered racks of sterile drinking water bottle racks or empty transport racks are delivered to the isolation housing room door, a **two person transfer procedure** should be utilized with:
    a. The interior individual donning PPE in advance in accordance with entry procedures described below,
    b. Keeping the door **closed** while the exterior individual uncovers, then **saturate sprays** the exterior of equipment/supplies with **Oxivir**,
    c. The door is opened, and the sprayed equipment/supplies either transferred to the interior cart, or the cart is rolled over the **antimicrobial adhesive mat** just inside the door,
    d. The door is closed, and the cart, rack, and equipment/supplies **sprayed again** by the interior individual, and allowed to **sit for 5 minutes** inside isolation.
    e. Racks of water bottles are covered with a Tyvek cover when kept within the room.

III.B. Preparation of the Secondary Enclosure

1. Isolation housing and use rooms shou2(k)-1o0 Td [bJ /TT1 1 Tf [(an)11p(f)-5.3(epar)-4(f)-1(.

decontamination.

5. Decontaminate the newly prepared isolation room and its contents, and validate efficacy of the decontaminating process in advance of occupancy, using **vaporized hydrogen peroxide** in accordance with SOP 1016 *Hydrogen Peroxide Vapor Decontamination* and SOP 1162 *Bioquell Z-2 Hydrogen Peroxide Vapor Generator System*, and at the discretion of the veterinarian, using environmental sampling for **detection of opportunistic bacteria** in accordance with SOP 410 *Sentinel Rodent Health Surveillance*.

6. Supplies and equipment must be dedicated to isolation housing and use.

7. If additional consumable supplies, equipment, reagents or biologics are needed during study, they must be placed in **nonporous secondary containers** (e.g., Ziploc bags, conical tubes, screw-top containers, autoclave-able bags) and either sterilized by autoclaving or vaporized hydrogen peroxide, or disinfected by saturate spraying with Oxivir TB. Containers are then delivered using a **two person transfer procedure** as described above.

III.C. Personnel Entry

1. Access to isolation rodent housing and use is limited to essential personnel, and only those with **documented didactic tutorial and in-person training** presented to the IACUC.

2. Work in isolation housing and use must precede all other work in any other animal facility area.

3. After donning disposable shoe covers and gown at the facility entrance, proceed directly to isolation housing and use and complete personnel entry procedures below.

4. Prior to entry, ensure that all portable **equipment and supplies** required for work have been **dedicated to isolation** housing and use.

5. The isolation housing room door must remain locked.

6. Access to isolation housing is limited only to essential personnel with documented didactic tutorial and in-person training presented to the IACUC.

7. Signage on the isolation housing room door must indicate that immunodeficient rodents are housed in isolation and must describe the PPE and procedures required for entrance.

8. Husbandry, care and use of rodents housed in isolation must be **completed first**, before accessing other areas.

9. At the isolation housing room door, all staff must already be wearing a disposable **gown** and **shoe covers** donned at the facility entrance, and then outside the isolation door **don in sequence** (a) **bouffant**, (b) surgical **mask** that covers the

10. If using portable **equipment** within the biosafety cabinet, ensure these are saturate sprayed with Oxivir, including all surfaces that may come into contact with mice.

11. If inhalational **anesthesia** is planned, saturate spray the induction chambers and rodent nosecone of the quad anesthesia unit and let it sit for 5 minutes.

12. If **imaging** in isolation, disconnect the induction chamber from the anesthesia machine, saturate spray both the interior and exterior of the induction chamber, and enter it into the biosafety cabinet and let it sit for 5 minutes8.9(ham)-5.9(ber6(t)-.end)10.5(l)1eu

keeping the door **closed** while the interior individual **identifies and saturate sprays the microisolators** with Oxivir on an interior cart, (c) the door is **opened**, and the sprayed microisolators transferred to the exterior individual and an **exterior cart**, (d) the door is closed, and the exterior cart is used to deliver the microisolators to the common procedure area.

23. If working in **bidirectional** isolation, whenever occupied microisolators are returned to the bidirectional isolation housing room door, a **two person transfer procedure** must be utilized with (a) the interior individual donning PPE in advance in accordance with entry procedures described above, including a **Tyvek suit**, (b) keeping the door **closed** while the exterior individual saturate sprays the microisolators with Oxivir, (c) the door is **or, , ,n2.3(()-6(P. Tw (a.6(h O)-6.6(hp er)-6(i)2.6(or)-6**

using the **two person transfer** procedure described above.

10. Individually ventilated caging (IVC) in isolation is **changed weekly**.

11. Any cage excessively soiled is changed as often as necessary to maintain an acceptable level of sanitation.

12. Ensure multiple **padded forceps** are soaking within a beaker or pan of Oxivir solution in the chamber of the biosafety cabinet.

13. Cage changing is noted on the *Room Status Sheet*.

14. IVC microisolators in isolation are **changed as an entire unit, including the bottle** with acidified water, the wire rack top with autoclaved feed, lid and bottom. Feeding and water bottle changing is noted on the *Room Status Sheet*.

15. **Saturate spray gloves and sleeves** with Oxivir

16. Prepare the biosafety cabinet for husbandry. Ensure the biosafety cabinet is on. **Saturate spray every surface of the biosafety cabinet interior** with Oxivir and allow it to sit for 5 minutes.

17. **After uncovering/opening** equipment/supplies needed for change outs (e.g., empty water bottle rack, IVC microisolators with autoclaved feed and bedding, racks of autoclaved water bottles filled with acidified drinking water), **saturate spray with Oxivir** and allow it to sit for 5 minutes.

18. Uncover/open only the supplies/equipment needed for change-outs.

19.

25. Nothing should touch the interior surfaces of the microisolator, including the biosafety cabinet surface. Cage

2. Whenever mice in **bidirectional** isolation must exit for a **common procedural area**, PPE must be donned in advance in accordance with isolation **entry procedures** described above, the microisolator to be transported is identified and saturate sprayed with Oxivir, and the sprayed microisolator transferred to the common procedural area by **exiting in your PPE**.

3. If **several** microisolators are to be transferred to a common procedural area, a **two person transfer procedure** should be utilized to exit with (a) the interior individual donning PPE in advance in accordance with entry procedures described above, (b) keeping the door **closed** while the interior individual **identifies and saturate sprays the microisolators** with Oxivir on an interior cart, (c) the door is **opened**, and the sprayed microisolators transferred to the exterior individual and an **exterior cart**, (d) the door is closed, and the exterior cart is used to deliver the microisolators to the common procedure area.

4. Work in common procedural areas involving mice from bidirectional isolation should **precede work involving other mice** from other housing areas. If multiple cohorts from separate housing rooms are present in the common procedural area, work with cohorts must occur in sequence, with work involving mice from isolation, including those administered uncharacterized human specimens, preceding the other cohort, rather than concurrently. **Only one cage should be opened at a time**.

5. If you have exited bidirectional isolation in your Tyvek suit, dispose of your Tyvek suit after exiting.

6. In the common procedural area, all staff must already be wearing a disposable **gown** and **shoe covers** donned at the facility entrance, and must also don in sequence outside the common procedural room (a) bouffant, (b) surgical mask that covers the nose.

7. Staff working with mice from bidirectional isolation must also then (a) push your thumb through the seam of the sleeve of the gown, (b) put on a pair of **gloves** ensuring cuffs **overlap the gown**, (c) **Tyvek sleeves**, and (d) put on a **second pair of gloves** ensuring gloves overlap sleeves and no skin is exposed.

8. **Saturate spray your sleeves and gloves** with Oxivir and wait 5 minutes before handling anything.

9. **Decontaminate all surfaces that may come in econ Tc 0a/P <</MCr and wTcbce-17.5(/MC**

11. If **imaging**, ensure imaging equipment surfaces that may come into contact with mice are decontaminated with Oxivir in accordance with SOP 1015 *Decontamination of Common Procedural Areas*.

12. After ensuring that all supplies and equipment are available and decontaminated, **saturate spray the cage** and water bottle with Oxivir and allow it to sit for 5 minutes.

13. **Only one cage is to be opened at a time**.

14. **Saturate spray gloves and sleeves** with Oxivir. A pan of Oxivir solution may be used to decontaminate gloved hands by immersion before opening each microisolator.

15. Remove the water bottle and place it on the work surface. Carefully unclamp and remove the microisolator lid and invert it onto the work surface and place the wire lid on top of the inner surface of the lid. The wire lid can be used to assist in handling rodents.

16. Nothing should touch the interior surfaces of the microisolator, even when retrieving a mouse with 0.6(a t)-5.Tw 57i(l)21.007 Tw 0.25 06hou2esttTc 0.002 -5.eemove thdf440 -1.152

Approved: **Date:**