1. PURPOSE
1.1. To provide vivaria/housing location managers and researchers with guidance on the IACUC’s expectations for a sanitation program to maintain sufficiently clean and dry bedding, adequate air quality, and clean cage surfaces and accessories.
1.2. To outline expectations for evaluating sanitation effectiveness for reusable specialized caging and research equipment used on a repeated basis for live animal activities when the items are not disinfected by CMP/ARU/PAR/PRF or autoclaved prior to use.

2. SCOPE
2.1. Applies to indoor-housed biomedical species; excludes agricultural animals covered by the Ag Guide and biomedical animals housed outdoors.
2.2. This applies to devices, equipment or items such as restraint devices or holders, behavioral or physiology testing equipment, induction chambers, imaging equipment, toys or enrichment items, other items, and non-standard caging food/water containers with repeated direct animal contact.
2.3. Specific guidelines as to the sanitation and/or sterilization of surgical instruments may be found in TAMU-G-013, TAMU-G-018, TAMU-G-022, TAMU-G-035, and TAMU-G-049.

3. RESPONSIBILITY
3.1. CMP/ARU/PAR/PRF will maintain records of efficacy testing and make logs available to the IACUC at semi-annual facility inspections, or to AWO staff during the performance of post-approval monitoring, upon request.
3.2. The PI (or designee) will prepare, and make available to the IACUC at semi-annual facility inspections, an SOP for each sanitization method not approved and routinely evaluated by CMP/ARU/PAR/PRF.
3.2.1. The PI will record efficacy testing and make the log available to the IACUC at semi-annual facility inspections or to AWO staff during the performance of post-approval monitoring, upon request.

4. DEFINITIONS AND/OR ACRONYMS
4.1. Centrally administered support service for animal research and teaching programs at Texas A&M University:
4.1.1. ARU: Animal Resource Unit supports the College of Dentistry vivarium
4.1.2. CMP: Comparative Medicine Program supports the Texas A&M College Station campus
4.1.3. PAR: Program for Animal Resources supports the Institute of Biosciences and Technology vivarium
4.1.4. PRF: Pharmaceutical Research Facility supports the Kingsville Pharmaceutical Science Facility vivarium
4.1.5. Sea Life: The Sea Life Facility support the Galveston campus
4.2. Cleaning: Removes excessive amounts of excrement, dirt, and debris.
4.3. Disinfection: Reduces or eliminates unacceptable concentrations of microorganisms.
4.4. Disinfectants: Are products used to reduce or eliminate microorganisms on non-living surfaces. Sporicidal disinfectants, such as Spor-Klenz, Wexcide, 10% Bleach (prepared daily), Opticide and Roccal-D, should be used, according to manufacturer’s instructions for all surfaces and equipment with direct animal contact.
4.4.1. Please note: Alcohol is neither a sterilant nor a high-level (sporicidal) disinfectant for surgical instruments. Please see surgical guidance referenced for additional information on sterilization of surgical instruments.
4.5. Direct Animal Contact: Refers to all surfaces the animal directly contacts when outside of the primary enclosure including, but not limited to behavioral testing equipment, restraint devices, and imaging equipment.
4.6. Life Support System: Refers to the physical structure used to contain the water and the animals as well as the ancillary equipment used to move and/or treat the water.
4.7. **PI**: Principal Investigator. The individual who has ultimate administrative and programmatic responsibility for the design, execution, and management of a project utilizing vertebrate animals.

4.8. **Sanitation**: Is the maintenance of environmental conditions conducive to health and well-being.


5. **GUIDELINES OR PROCEDURE**

5.1. **Sanitation**

5.1.1. Methods and frequencies of sanitation will vary with many factors, including the normal physiologic and behavioral characteristics of the animals; the type, physical characteristics, and size of the enclosure; the type, number, size, age, and reproductive status of the animals; the use and type of bedding materials; temperature and relative humidity; the nature of the materials that create the need for sanitation; and the rate of soiling of the surfaces of the enclosure.

5.1.2. Some housing systems or experimental protocols may require specific husbandry techniques, such as aseptic handling or modification in the frequency of bedding change.

5.2. **Sanitation of the Microenvironment**

5.2.1. *In general*, enclosures and accessories, such as tops, should be sanitized at least once every 2 weeks.

5.2.2. Primary enclosures can be disinfected with chemicals, hot water, or a combination of both.

5.2.3. Effective disinfection can be achieved with wash and rinse water at 143-180°F or more. The traditional 82.2°C (180°F) temperature requirement for rinse water refers to the water in the tank or in the sprayer manifold.

5.2.4. Detergents and chemical disinfectants enhance the effectiveness of hot water but should be thoroughly rinsed from surfaces before reuse of the equipment.

5.2.4.1. May be contraindicated for some aquatic species, as residue may be highly deleterious.

5.2.5. Mechanical washers (e.g., cage and rack, tunnel, and bottle washers) are recommended for cleaning quantities of caging and movable equipment.

5.2.6. **Handwashing**

5.2.6.1. Can be effective but requires considerable attention to detail.

5.2.6.2. Must ensure that surfaces are rinsed free of residual chemicals.

5.2.6.3. Must ensure that personnel have appropriate equipment to protect themselves from exposure to hot water or chemical agents used in the process.

5.2.7. **Smaller Pieces of Equipment**

5.2.7.1. Should be washed with detergents and/or hot water and, where appropriate, chemical agents to destroy microorganisms.

5.2.7.2. Cleaning with ultrasound may be a useful method for small pieces of equipment.

5.2.7.3. Examples: water bottles, sipper tubes, stoppers, feeders.

5.2.8. **Automated Watering Systems**

5.2.8.1. To ensure that microorganisms and debris do not build up in the watering devices:

5.2.8.1.1. Flush periodically with large volumes of water or appropriate chemical agents followed by a thorough rinsing, and/or

5.2.8.1.2. Use constant recirculation loops that use properly maintained filters, ultraviolet lights, or other devices to disinfect recirculated water.

5.2.8.1.3. Include the routine sanitation of automatic water delivery valves (i.e., lixits).

5.2.9. **Sterilization**

5.2.9.1. It may be necessary to sterilize caging (and associated equipment)

5.2.9.1.1. To ensure that pathogenic or opportunistic microorganisms are not introduced into specific-pathogen-free or immunocompromised animals, or

5.2.9.1.2. To ensure the destruction of experimental biologic hazards before cleaning.

5.2.9.2. Sterilizers should be regularly evaluated and monitored to ensure their safety and effectiveness.

5.2.9.3. Autoclaves can be monitored through the use of sterilization indictors that validate materials have been properly sterilized.
5.2.10. Pens/Runs
   5.2.10.1. Solid surfaces can be sufficiently maintained with frequent flushing with water and periodic use of detergents or disinfectants, as appropriate.
   5.2.10.2. Animal waste removed by flushing, will need to be done at least once a day at a time that considers the normal behavioral and physiologic processes of the animals (e.g. gastrocolic reflux in meal-fed animals).
   5.2.10.3. Animals should be kept dry during flushing.

5.2.11. Aquatic Systems
   5.2.11.1. A properly designed and functioning life support system will maintain nitrogenous wastes within an acceptable range.
   5.2.11.2. Because waste is dissolved in the water and/or removed as solids by siphoning (hydrocleaning) or filtration, regular changing of tanks is not integral to maintaining adequate hygiene in typical aquatic systems.
   5.2.11.3. Filters need routine cleaning or replacement or proper maintenance, depending on the type.
   5.2.11.4. Nitrites are generally removed through water changes or by a specialized denitrification unit.
   5.2.11.5. Dissolved proteins may be removed by protein skimmers (saltwater systems)
   5.2.11.6. Disinfection is usually accomplished through filtration and application of UV light or ozone and/or water changes.
   5.2.11.6.1. Note: Chlorine and most chemical disinfectants are inappropriate for aquatic systems containing animals as they are toxic.
   5.2.11.7. The type of monitoring and frequency varies depending on the disinfection method, the system, and the animals. The frequency of cleaning and disinfection should be determined by water quality, which should permit adequate viewing of the animals, and animal health monitoring.
   5.2.11.7.1. Algal growth is common in aquatic systems and algal species seen with recirculating systems are generally nontoxic, although species capable of producing toxins exist. Algae are typically removed using mechanical methods (i.e., scrubbing or scraping). Limiting algal growth is important to allow viewing of the animals in the enclosure.
   5.2.11.8. System components such as lids on fish tanks, which may accumulate feed, may require sanitation as often as weekly depending on the frequency and type of feed and the system’s design.

5.3. Sanitation of the Macroenvironment
   5.3.1. All components of the animal facility, including animal rooms and support spaces should be regularly cleaned and disinfected as appropriate to the circumstances and at a frequency based on the use of the area and the nature of likely contamination.
   5.3.2. Vaporized hydrogen peroxide or chlorine dioxide are effective compounds for room decontamination.
   5.3.3. Cleaning implements
       5.3.3.1. Should be made of materials that resist corrosion and withstand regular sanitation.
       5.3.3.2. Should be assigned to specific areas and should not be transported between areas with different risks of contamination without prior disinfection.
       5.3.3.3. Worn items should be replaced regularly.
       5.3.3.4. Should be stored in a neat, organized fashion that facilitates drying and minimizes contamination or harborage of vermin.

5.4. Sanitation by CMP/ARU/PAR/PRF
   5.4.1. Equipment and items should be sanitized by CMP/ARU/PAR/PRF whenever possible. CMP/ARU/PAR/PRF routinely performs sanitization efficacy testing, thus removing the need for investigators to independently perform this testing.
   5.4.2. An SOP(s) with multiple methods for the sanitization of specialized caging and research equipment that comes into direct contact with live animals has been developed by CMP/ARU/PAR/PRF for use in the research lab.
5.4.3. Use of these standard sanitization methods are strongly recommended when not utilizing CMP/ARU/PAR/PRF’s direct services.

5.4.4. CMP/ARU/PAR/PRF regularly tests and documents the sanitation effectiveness of these standardized methods. Investigators are not required to perform independent efficacy testing when sanitizing items as described in the CMP/ARU/PAR/PRF SOP.

5.5. **Sanitation by Principal Investigator or Facility Manager**

5.5.1. If an investigator/manager utilizes a sanitization method not approved and routinely evaluated for efficacy by CMP/ARU/PAR/PRF, the investigator/manager must develop an SOP and independently perform and document efficacy testing.

5.5.2. The SOP(s) should minimally include: the item(s) being disinfected, the frequency of disinfection, the disinfectant(s) used, contact time, dilution and appropriate personal protective equipment needed.  
5.5.2.1. The SOP should also contain cleaning/disinfection instructions as specified by the manufacturer, where applicable.

5.5.3. Solid/Non-Porous/Non-Permeable (i.e. stainless steel, hard plastic) should be sanitized using sporicidal disinfectants at a frequency to maintain sanitary conditions.

5.5.4. Porous materials cannot be sanitized and therefore must be cleaned and replaced regularly to help ensure a clean environment for the animals.

5.5.5. Performance measures such as visual inspection of equipment and assessment of animal health must be evaluated on a continual basis.

5.6. **Assessing the Effectiveness of Sanitation**

5.6.1. Monitoring of sanitation practices should occur regularly and fit the process and materials being cleaned.

5.6.1.1. Methods relying on mechanical means of sanitation require the regular evaluation of the components, such as spray arms, moving headers, and spray nozzles.

5.6.1.2. Temperature dependent processes should be evaluated with the use of temperature-sensing devices (e.g., thermometers, probes, or temperature-sensitive indicator strips) to ensure that the equipment being sanitized is exposed to the desired conditions.

5.6.1.3. Visual inspection and microbiologic and water temperature monitoring may be indicated.

5.6.1.4. **Note**: The intensity of animal odors, particularly that of ammonia, should not be used as the sole means of assessing the effectiveness of the sanitation program.

5.6.2. Whether the sanitation process is automated or manual, regular evaluation of sanitation effectiveness is recommended.

5.6.2.1. This can be performed by evaluating processed materials by microbiologic culture or the use of organic material detection systems (e.g., adenosine triphosphate [ATP] bioluminescence) and/or by confirming the removal of artificial soil applied to equipment surfaces before washing.

5.6.2.2. Surfaces that do not pass efficacy testing, must be re-sanitized until a passing level is achieved.

5.6.2.3. Each distinct method of sanitization should be assessed individually for efficacy. For example, hand-wash/rinse and chemical disinfection are two distinct methods of sanitization, requiring assessment of efficacy for each method.

5.6.2.4. Evaluation of sanitation effectiveness should be performed on a regular basis, at least once every 3 years, or whenever there is a change to the sanitization procedure.

5.6.2.4.1. In addition, a representative population of items from survival surgery areas, behavioral testing equipment, along with other animal use areas, as deemed appropriate, will be monitored quarterly by CMP/ARU/PAR/PRF for sanitation efficacy.

5.7. **Recordkeeping**

5.7.1. Each efficacy testing episode should be recorded in a log and should include the sanitization method used, the type of specialized cage or research item subjected to efficacy testing, the date the items were sanitized, and the date the efficacy testing was performed.
5.7.2. Both the log and SOP(s) must be available for review upon request.

6. EXCEPTIONS

6.1. The PI may request an exception to the above standards by describing the departure in the AUP

6.2. For programmatic exceptions, the facility director or manager may submit a request for the exception using TAMU-F-013

7. REFERENCES, MATERIALS, AND/OR ADDITIONAL INFORMATION

7.1. References/Resources


7.1.2. Biosafety in Microbiological and Biomedical Laboratories, 5th Edition


7.1.4. Hygiena SystemSURE Plus Luminometer, Ultrasnap ATP Swabs


7.2. IACUC/AWO Referenced Documents: (requires TAMU NetID authentication)

7.2.1. TAMU-F-013 Request for Programmatic Exception from Animal Welfare Standards

7.2.2. TAMU-G-013 Guidelines for Survival Surgical Procedures in Rodents

7.2.3. TAMU-G-018 Guidelines for Performing Surgical Procedures in Non-Rodent Mammals

7.2.4. TAMU-G-022 Guidelines on the Performance of Non-Survival Surgery

7.2.5. TAMU-G-035 Guidelines for Performing Surgery in Fish

7.2.6. TAMU-G-049 Guidelines for Performing Surgery in Amphibians and Reptiles

7.3. For more information on sanitation methods and available SOPs, please contact:

7.3.1. CMP at (979) 845-7433

7.3.2. ARU: at (214) 828-8149

7.3.3. PAR: at (713) 677-7471

7.3.4. PRF: at (361) 221-0770

7.3.5. Sea Life Center: at (409) 740-4574

7.4. Acknowledgements

7.4.1. This document was partially adapted using materials obtained from the University of Arizona and the Ohio State University.

8. HISTORY

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<td>College Station/Galveston: New Document; replaced unnumbered document titled “Maintenance and Storage of Sterilized Items”</td>
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<tr>
<td>12/16/2019</td>
<td>001</td>
<td>Houston/Kingsville: New Document</td>
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<td>Dallas: New Document</td>
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<td>003</td>
<td>College Station/Galveston: Renewal of expiring document; updated definitions. Reviewed and approved via email.</td>
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