PURPOSE

- To provide the highest quality of care of Xenopus frogs
- To prevent the spread of disease that could compromise research studies.

IDENTIFICATION

A. Holding and individual/experimental tanks will be labeled:
   1. PI's name
   2. IACUC protocol number
   3. Species
   4. Number of animals
   5. Date of arrival

ANIMAL HEALTH MONITORING

A. The Principle Investigators, Graduate Students, Undergraduate Students, and/or Animal Laboratory Technicians will observe all animals daily.

B. All animals are observed for signs of illness or changes in behavior and the “Daily Observation Record” is completed. Signs of illness or distress in xenopus include, buoyancy problems, weight loss, dropsy (coelomic distention), bloat, cottony tufts attached to skin, petechial, ecchymoses on the belly, legs, nose, or feet; excessive slime coat mucous production, bite wounds on legs or body, cloudy corneas, hyperemic, distended cloaca, swimming in circles, tilted or upside down, unusual amounts of shed skin in the tank water.

C. The temperature and relative humidity of the room are recorded daily in the room temperature log. All problems are reported to the ACS Facilities Manager.

D. The room is checked for operational deficiencies. All deficiencies are reported to the ACS Facilities Manager.

E. Temperature ranges for Xenopus macroenvironment shall remain between 65 and 75°F

F. Photoperiod is set at 12:12. Lights are controlled by an automatic wall-mounted timer and lights come on a 2 am and go off at 2 pm. (note: time on/off or photoperiod split may be altered to accommodate specific experiments; signs will be posted with light schedule if it is altered)
G. Frogs are fed ~2 teaspoons of frog food every Monday, Wednesday and Friday morning.
   1. This amount is an estimate; feeding typically breaks down to 3-6 pellets of food brittle per frog.
   2. If frogs eat the entire amount within 20 minutes, they will be fed another teaspoon per cage.

H. Any dead animals are reported to the PI and the ACS Facilities Manager or Veterinarian.

SANITATION/AQUARIUM CHANGES/WATER QUALITY MONITORING

I. The water levels are checked daily in each aquarium containing frogs.
   1. Frogs may be housed in individual glass or plastic aquaria on the benchtop, in the trout tank, or in the Aquatic Ecosystem.
   2. If the Aquatic Ecosystem is broken, house frogs individually or in the trout tank (but, be careful about what age classes are combined as adult frogs will eat juvenile frogs).
   3. Water depth should be at least 3 inches for aquaria, if less than 3 inches add dechlorinated water to reach the correct depth (unless a shallower depth is purposely used for an experiment).
   4. For the Aquatic Ecosystems set up, the water should be at the line marked 'max water level'. (please see Supplement 1 and the end of the document for more on the Aquatic Ecosystem).

J. The Aquatic Ecosystem’s recirculating tank’s water is changed weekly.
   1. Use DI water containing 0.3 g Instant Ocean per liter.

K. Water in individual tanks is changed on Monday, Wednesday, and Friday.
   1. Use DI water containing 0.3 g Instant Ocean per liter.
   2. Dirty tanks are washed with white vinegar and DI water and left to air dry.

L. Use the vacuum siphon to clean the debris from the Aquatic Ecosystem tanks, the trout tank if in use, on Monday, Wednesday and Friday.

M. The filters should be cleaned on the Aquatic Ecosystem every two weeks.

N. For the trout tank and the Aquatic Ecosystem, as you change the water, wipe down the sides of aquarium to remove any deposits.

O. Water Quality testing is conducted once weekly.
   1. Parameters include at least: water temperature, pH, NH₄, NO₂, NO₃
   2. Testing is done with commercially available kits (e.g., Tetra EasyStrips, API water drop kits) following the manufacturer’s instructions.

OCCUPIED ANIMAL ROOMS SANITATION
A. Trash is removed daily.
B. Floors are swept and mopped with disinfectant mixed according to the manufacture’s suggestions at least once a week.
C. Door frames and vents are dusted once a week
D. Total room sanitation occurs monthly
E. The Facility and clean aquaria will be sampled within 24 hours of sanitation procedures on a quarterly basis using the SafeStep microbiological contamination monitoring system.

HEALTH CARE

Everyone with access to the animal facility is responsible for informing the University Veterinarian when an animal becomes ill or a change in behavior is noted. Seriously ill animals should be reported IMMEDIATELY to the veterinarian. When an investigator, technician, or animal care personnel requires veterinary assistance, they should:

A. Complete the “Animal Treatment/Observation Form” in the Notebook. Indicate the date, room number, animal number/cage ID, and problem observed, and ensure that the name (or initials) of the person making the report is recorded.

B. Contact the University Veterinarian or the ACS Facility Manager at:

   Dr. Tiffanie Brooks, ACS Attending Veterinarian  
   806-834-8588 Office 
   806-239-2120 Cell 

   Dr. Paul Stonum, ACS Clinical Veterinarian  
   806-834-7373 Office 
   660-562-4425 Cell 

   Sydnee Woodman, ACS Facilities Manager  
   806-834-2872 Office 
   602-758-0670 Cell 

C. Provide all the above information to the individual contacted above, who will give advice and authorization for the action(s) that should be taken.
Supplemental Document 1 – Aquatic Ecosystem (aka Frog Apartments)

- Water lines from pump
- Drain lines to sump
- Valves controlling flow to each tank. Should be mostly closed to reduce stress on frogs.
- Water valves controlling flow from pump.
Water Valves

1. Regulates flow to pump. Should always be mostly open to prevent damage to pump.
2. Used to drain sump.
3. Regulates direct flow to tanks. Should always be mostly open in case filters clog.
4. Controls flow thru filters
5. Used to back flush filters.
6. Used to drain filters. Can also be used to black flush individual filters.
7. Used to drain filters. Can also be used to black flush individual filters.
8. Closed when filters are removed for cleaning.
9. Can be connected to sump to regulate flow rate to tanks.
10. Closed when back flushing filters.
11. Used to connect with additional tanks when needed.
12. Used to connect with additional tanks when needed.
13. Used to connect with additional tanks when needed.

Water changes and Back Flushing

1. Connect drain hose to 5.
2. Open 5.
3. Open 3 completely.
5. Do not let water level in sump get below minimum line.
7. Close 5.

D. Weekly duties (filter cleaning) expanded
   1. Attach the green hose from the trash can in the back of the room to the bottom valve of the filter chamber on the right with the pumps still going. Turn the valve vertical (to where the white line is facing you). Drain for about 5 minutes. Turn the valve back to horizontal, unscrew the hose, and repeat with the chamber on the left. Return the hose to the trash can in the back.

E. Monthly duties (filter cleaning) expanded
   1. Do the step from section 6.6
   2. Turn off the pumps.
3. Turn the white knob on top of the lid to let out the air from the chamber. Next, remove the lid on top of the filter chamber. To do this, you will need to get a hammer or wrench and tap on the protrusions on the side to loosen the lid before unscrewing the lid.

4. Take out the filters and put into bucket. KEEP THE FILTERS IN THE SAME ORDER.
5. Suck out the dirty water
6. Clean the filters with a little bit of bleach and a lot of water. All of the bleach should be completely gone from the filters before returning the filters back to the filter chamber.

7. Screw the lid back on along with the white knob on top to seal the lid. Lightly tap the lid with the wrench/hammer to fully tighten. Turn the pump back on.