

**TITLE:** Xenopus Husbandry

**SOP Category:** Aquatics

**RUAC SOP #:** 5.08

**Page:** 1 of 4

**Effective Date:** 04/21/2025

**Approval:**

*David C. Rein DVM*

**Revisions:** 5/24/2022; 10/25/2024

**SCOPE:** To describe procedures for proper care and husbandry of Xenopus housed in the facility.

**OBJECTIVE:** The objective of the procedures outlined is to ensure that Xenopus care and housing is consistent with regulatory guidelines.

## PROCEDURES:

### Housing

Tanks must be constructed of non-porous material that can be cleaned and disinfected regularly and allow for daily observation of the animals. Tanks must provide a safe environment and not be constructed of material that may cause injury to the frogs. Frogs must be housed such that their primary enclosure meets their general needs based on species needs, behavior, and goals of the study. Generally, Xenopus should be grouped housed, however, aggression, illness or project-related reasons may warrant individual housing.

### Density

- *Xenopus laevis*
  - Adult frogs should have at least 2 liters of water per adult frog.
  - Floor space must be taken into account and water depth for adult frogs must be at least 15 cm of water, although 30-50 cm is preferred.
  - Frogs must be able to fully submerge and be adequately covered.
- *Xenopus tropicalis*
  - Adult *X. tropicalis* should be housed at a stocking density of 1-2 frogs per liter of water.
  - Floor space must be considered and water depth for adult frogs must be at least 15 cm of water.
  - CMR houses Xenopus in a rat size cage -tank (approx. 18 ½" x 10" x 8") We house 2 adults per cage.



## **Exceptions**

Exceptions to these guidelines may be granted under the following conditions:

- Any deviations from the housing density guidelines provided above, including single housing, must be reviewed, and approved by IACUC in the animal use protocol.
- However, single housing may be approved via veterinary exception for health or welfare concerns.

## **Physical environment**

- Temperature
  - Heating and air in frog rooms should be controlled in a manner that supports species-specific needs. Optimal room temperatures should be between (19-25°C/66-77°F).
  - Room temperature must be recorded on the room log sheet and close to the temperature of the tank water to prevent having to adjust water temperatures as a result of fluctuations in room temperatures.
- Percent Relative Humidity (%RH)
  - Humidity at the room level does not directly impact Xenopus however high levels of humidity can be detrimental to electronic equipment. Regular monitoring of the HVAC system is important and is best performed at the room level. Humidity should be between 30-70%.
- Ventilation.
  - Recommended range is 10-15 air changes/hour.
- Illumination
  - A lighting regime of between 12 and 14 hours of light, and 12- and 10-hours dark is recommended.
- Feeding
  - Frogs are fed palatable, non- contaminated, and nutritionally adequate food on Monday, Wednesday, and Friday, typically 3 times per week a week (with a minimum gap of around 48 hours) or according to specific requirements as described in an approved protocol.
  - Change water 2-3 hours after feeding.
  - Feed must be kept in properly labeled vermin-controlled container. Ours is kept in the refrigerator. It should be discarded after one year, when refrigerated. If food is kept in the room, then it's to be discarded 6 months after being opened. These were recommendations from the manufacturer Nasco. This shelf life is extended indefinitely if feed is kept frozen.

## **Sanitation**

**Clean tanks at least once weekly.**

- Standing water tanks.

## **TITLE: Xenopus Husbandry**

**SOP #: 5.08**

**Page 3 of 4**

- Transfer frogs to a clean tank containing RO water. Use dedicated nets and accessories. Drain the water from the dirty tank.
- If the tank is hand cleaned, use a clean sponge or brush to aid in removing deposited debris. For disinfection, food grade hypochlorite at 200mk/70L (0.28% solution), 3% bleach, Betadine scrub, or other approved disinfectant should be used.
- It is critical that the tanks are thoroughly rinsed clean of any residual chemicals before placing frogs into the tank.

### **Daily Procedures**

- Sweep/ mop floors to remove dust, dirt, and excess water.
- Be sure aeration pump is working properly in each tank and in the storage tank in room.

### **Biweekly**

- Wipe down shelves used for housing when containers are removed for washing.

### **Twice a year**

- Disinfect and sanitize room and scrub brushes, sponges, enrichment devices, holding containers and floors.

### **Observations and other procedures**

#### **• Daily Room Log**

- Observe each animal and check for health issues. Signs to look for include red or other discoloration of the skin, failure to feed properly, weight loss, open cuts or abrasions, bloating, and lethargy.
- Check and record water temperature. Temperature should be maintained at 17-24°C (*X. laevis*) and 24-28°C (*C. tropicalis*).
- Check that each tank is individually identified and has a total frog count.
- Document parameters listed above in addition to room activities (ex: feeding, animal number, and room temperature).

#### **• Weekly Procedures**

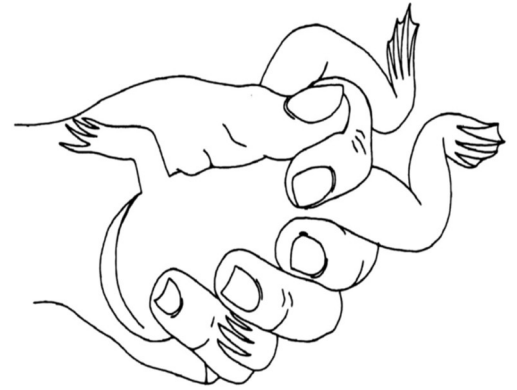
- Check and record water quality, pH (6.5-8.5)

#### **• Water Quality and Temperature**

- Water should be dechlorinated and chloramine free water with 1 adult frog per 2 liters of water and 5-10cm in depth.
- If water quality values are out of the normal range, contact your supervisor or manager to correct the issue. Document tank checks on room log sheet.
- Suggested water quality should be kept in the following ranges for both *X. laevis* and *X. tropicalis* unless specified to maintain optimal growth and performance:
  - Hardness (CaCO<sub>3</sub>) 175-300 mg/L (*X. laevis*) and 100-300 mg/L (*X. tropicalis*)
  - Alkalinity (CaCO<sub>3</sub>) 50-200 mg/L
  - Ammonia (NH<sub>3</sub>) <0.02 mg/liter
  - Nitrite (NO<sub>2</sub>) <0.5mg/liter
  - Nitrate (NO<sub>3</sub>) <50mg/liter

- **Handling**

- Handle frogs with clean, soap-free hands or smooth latex gloves. Powdered or textured gloves abrade the frog's skin.
- Pick up the frog by placing one hand across its back with a forefinger between the animal's hind legs and wrapping the rest of the hand around the animal's middle.
- Use the other hand to cover the frog's eyes, to calm and physically restrain it. Escaped frogs dehydrate quickly and die with hours.



- **Enrichment**

- All frogs must have a refuge to serve as environmental enrichment such as a PVC pipe cut lengthwise. Ensure that there are not any sharp edges which may cause abrasions on the frog's skin, other forms of enrichment are acceptable providing they are nonporous, do not harm the frogs, and can be cleaned and disinfected.

- **Identification and records**

- Each tank should have an RFID cage card and frog count should be noted.