Title: Fish Husbandry
SOP Number: 001
PURPOSE: To provide the highest quality of care for fish housed in Texas Tech animal facilities, and to prevent the development of issues that could compromise animal health or research.

ROOM MAINTENANCE

A. Daily

1. The Principle Investigators, students or staff will observe all tanks/aquaria daily.
   a. Check the room for operational deficiencies.
      1. Report any problems to the PI, Unit Manager or Building Work Order Coordinator.
   b. Observe all fish for evidence of illness or change in behavior and complete the "DAILY OBSERVATION FORM."
      1. Report sick fish or other animal health problems and treatment on the "TREATMENT/OBSERVATION FORM", and report to the ACS Staff.
   c. Remove dead fish and document in "Daily Observation" book.
   d. Check temperature in tanks/aquaria.
   e. Feed accordingly.
   f. Mop up any water on the floors.

B. Weekly

1. Check water parameters and record in notebook:
   a. Nitrites: should always remain at 0 ppm, reduce if elevated.
   b. Ammonia: should always remain at 0 ppm, reduce if elevated.
   c. pH: should be in the range of 6 - 9 pH units and adjusted according to optimal conditions for the species being held.
   d. Dissolved Oxygen should be >4.5 mg/L.

C. New tanks and aquariums

1. Initial record of various water quality parameters should be made every other day until stabilized (4-6 weeks) then weekly as above regarding the natural quality of the water source prior to its use in an artificial aquatic system to document both the high alkalinity and pH values. The following water quality parameters should be made on new aquariums:
   a. pH
   b. Alkalinity
   c. Hardness
   d. Chloramines
   e. Dissolved oxygen
2. Life support systems must be set up and acclimated prior to receiving new fish. Life support systems typically fall into three categories: closed recirculating, flow-

through or static. The water may be fresh, brackish or salt and is maintained at specified temperatures depending the species.

Closed Recirculating

1. The biological filtration system for a closed recirculating system should be “primed” by the addition of either an approved hardy disease-free fish or ammonium chloride (5 mg/L/day). Since the detoxifying capacity of a biological filter adapts to its nutrient supply, the capacity of a seeded or “seasoned” biofilter should be considered prior to the introduction of the entire fish population.
2. Water parameters (e.g. ammonia and nitrite levels) of the system should initially be monitored every other day until they stabilize to safe levels.
3. Considering the naturally high pH levels of water available at Texas Tech University and the tendency of ammonia to exist in its more toxic form in alkaline waters, measurement of the nitrogenous waste products is critical in newly established aquatic systems.
4. The testing interval can be lengthened to weekly checks for pH, ammonia, nitrites and salinity (for marine systems) once these parameters measure within the safe range.

Flow-through

1. Water is constantly replaced and may use large volumes of water.
2. Required water turnover rate (tank volumes per day) may vary depending on fish density. It must be set at a value capable of maintaining water quality parameters within acceptable values.

Static

1. Water is stationary and periodically replaced (partially), and may use mechanical devices to move and aerate water.
2. Required frequency of replacement depends partly on fish density and must be set at rate that maintains water quality parameters within acceptable values. Typically every other day to weekly.

D. Water supply

1. If a municipal water supply is used the water must be dechlorinated. All water should be analyzed prior to use.
2. Use a Daily Observation Form and the Required Water Analysis chart to record data. Check water quality prior to use and adjust accordingly.

E. Personnel Safety

1. Use exam gloves when handling fish and wash hands with anti-microbial hand soap (unless chemicals within antimicrobial soaps may interfere with ongoing or future research in the laboratory).
2. Periodically check all electrical cords, fixtures, and outlets.

F. Health Care

It is everyone's responsibility to inform the ACS Staff 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:

1. Complete the "TREATMENT/OBSERVATION FORM" in the Notebook. Indicate the date, animal number/cage ID, problem observed and name or initials of the person making the report.
2. Contact Animal Care Services Veterinarian or ACS Manager:

   Attending Veterinarian.
   806-834-8588 Office
   806-239-2120 Cell Phone

   Facilities Manager.
   806-834-3437 Office
   254-913-5156 Cell Phone
3. Provide all the above information to the individual contacted above, who will give advice and authorization for the action(s) that should be taken.

AQUATIC HEALTH RECOMMENDATIONS

A. Disease Transmission Prevention

1. All fish populations intended for research purposes are initially quarantined and the initial health status determined prior to starting the research project.
2. Health checks need to be conducted upon arrival and 1 week later to alert a researcher to any potential health problems.
3. Monitoring and recording water quality parameters and fish feeding/ behavioral patterns at regular intervals need to be conducted for the duration of a research project.
4. Suspect fish can be submitted to the Texas Veterinary Medical Diagnostic Lab in College Station, TX or Amarillo, TX for diagnostic evaluation or evaluated on-site using diagnostic techniques.
5. Dip nets should be assigned to a specific aquarium or disinfected prior to use between different aquaria. A "dip net" bucket can be used for this purpose (e.g. 10 ml household bleach to 1 L water as a 1 hour dip). Bleach must be mixed fresh daily or as needed. Nets then need to be rinsed thoroughly after this treatment to remove chlorine residues prior to netting fish. Since chlorine is highly toxic to fish, sodium thiosulfate (or other dechlorinator) should be on-hand for emergency dechlorination purposes.
6. Transport stress can result in the occurrence of secondary infections in fish by aquatic opportunistic pathogens. Quarantine and acclimation of new fish populations for 2-4 weeks prior to initiation of a study is recommended to prevent the interference such an event would cause during a research project. A physical examination (e.g. gill biopsy, etc.) of a sample set from a new population may also be conducted to give an indication of the initial health status of a fish population upon its arrival to the facility.

7. The presence of multiple aquaria in large open recirculating systems increases the likelihood of disease transmission by potential pathogens. Disinfection of water exiting the biofiltration system by ozonation or UV-radiation should be considered as a deterrent against disease transfer between aquaria in these systems.

B. Handling and Restraint

1. Variation in euthanasia dosages for Tricaine Methanesulphonate (MS-222) was cited in different proposals and may reflect species variation noted in different references. The standard dosage reported by DeTolla in Guidelines for the Care and Use of Fish in Research is 500 mg/L.
2. The anesthetic dosage of this compound is reported to be between 100-200 mg/L. The anesthetic effect of MS-222 is affected by both the dosage and duration of exposure. Since it acts as a respiratory depressant prior to causing cardiac arrest, the opercular movements of fish can be used to determine the plane of anesthesia. Fish should be removed from the anesthetic-treated water as opercular movements slow and response to stimuli ceases. If opercular movements cease, the fish should be placed in untreated water and forcibly moved open-mouthed through the waters until respiration recovers.
3. All methods of immobilization should be tested on the study species (if possible) prior to their actual utilization in a study due to species variation that is encountered in response to various techniques.