STANDARD OPERATING PROCEDURE

ENVIRONMENTAL ENRICHMENT FOR FISH USED IN RESEARCH AND TEACHING

1.0 PURPOSE:

1.1 This standard operating procedure (SOP) describes the methods for environmental enrichment of fish.

1.2 These methods are intended to improve the well-being of these animals by increasing species-specific behaviors and reducing maladaptive behaviors.

1.3 This SOP is part of the UGA Environmental Enrichment Program that fully complies with the requirements of the National Research Council, *Guide for the Care and Use of Laboratory Animal*, ed8 available at http://grants.nih.gov/grants/olaw/Guide-for-the-Care-and-Use-of-Laboratory-Animals.pdf and the Animal Welfare Act and Regulations: Public Law 99-198 – The Improved Standards for Laboratory Animal Act available at http://www.nal.usda.gov/awic/legislat/awa.htm .

2.0 STANDARDS:

2.1 Natural Behavior:

The term "fish" most precisely describes any non-tetrapod craniate (an animal with a skull and in most cases a backbone) that has gills throughout life and whose limbs, if any, are in the shape of fins. Fish are not a single clade but a paraphyletic collection of taxa. (Helfmann et al., 1997; Lundberg, 1995) Traditional classification divides fish into three extant classes. However, only the Superclass *Osteichthyes* or "bony fish" are currently used in the University of Georgia laboratory animal-research environment. The needs of captive fish, including their environmental enrichment needs, vary extensively. For this reason, this SOP will focus on the needs of current species housed at UGA, which include guppies (*Poecilia reticulate*), zebrafish (*Danio rerio*), medaka (*Oryzias latipes*), mummichog (*Fundulus heteroclitus*), catfish (*Siluriformes*) and sturgeon (*Acipenseridae*). Guppies are found in a wide variety of habitats and alter their feeding and shoaling habits accordingly. Zebrafish and medaka are found naturally in shallow, slow-moving water, usually in the shade. The areas typically have vegetation and a silty substrate, such as a rice-field (Spence, 2006; Davis et al., 2002) They are both omnivorous. Zebrafish feed both on material sinking and material floating. Medaka require a variety of food sources for adequate growth and production (Davis et al., 2002) Mummichog are marsh or tidal species most often found in mud substrate or grass. They are well-adapted to surface feeding but may feed at other levels as well. They are very hardy in a variety of temperature and salinity ranges. Catfish are a very diverse group or ray-finned fish. They live on every continent except Antarctica. Most catfish are bottom feeders. Most catfish feed through suction or gulping rather than biting or cutting prey (Bruton, 1996). Sturgeon are one of the oldest families of bony fish in existence. They are native to subtropical, temperate and sub-Artic rivers, lakes and coastlines of Eurasia and North America. Most sturgeons are bottom feeders, migrating from their coastline habitat to freshwater to breed and spawn. They prefer sandy substrates with dark, deep flowing water. (Allen et al, 2007) All species of fish have optimum stocking densities that are a function of fish size, number, tank size and husbandry, such as feeding amount and amount of available enrichment. Overstocking will increase competition, aggression and stress.

2.2 Environmental enrichment must be evaluated by taking into account the following:

2.2.1 The Natural Behavior and needs of fish (see above)

2.2.2 Social Enrichment – Housing of compatible co specifics offers a high level of enrichment. Every effort will be made socially house social species. If social housing is not possible, animals should be housed in a manner that allows for as much tactile, auditory, visual or olfactory contact as possible. Social housing is a recognized and important part of the Environment Enrichment Program but should not be viewed as the sole means of meeting the enrichment needs of animals.

2.2.3 Physical Enrichment (devices, toys, etc) – Physical enrichment can be an important part of the Environmental Enrichment Program. However the selection of physical enrichment should take into account the safety of the device, its ability to stimulate and maintain the animal's interest and its impact on the research being conducted. Physical enrichment should be carefully monitored to assess its impact of the goals of increasing natural behaviors.

2.2.4 Activity/Food Enrichment – Activity/food enrichment can be an important part of the Environmental Enrichment Program. However, the selection of activity/food enrichment should take into account the health of the animal, the limitations of its confines and its impact on the research being conducted. Any activity/food enrichment should be planned in consultation with the Attending Veterinarian (AV) and the Principal Investigator (PI).

2.3 The enrichment program is carried out by University Research Animal Resources (URAR). Specific needs and requirements should be communicated to the Assistant Director of the Animal Resources (AR) Unit.

2.4 Unless specifically justified by the PI in the Animal Use Proposal (AUP), all animals will receive enrichment. It is recognized that animal enrichment can be a research variable. In caring for the psychological well-being of animals, it is important to recognize limitations and use a balanced approach in providing the best possible care and allowing for the expression of species-typical behavior within a functioning research environment.

2.5 Abnormal Behaviors:

The Environmental Enrichment Program is a dynamic process. Ongoing evaluation is a necessary component to meeting the goal of more species-specific natural behaviors. University Research Animal Resources (URAR) will regularly monitor all enrichment, in part, by looking for stereotypical behaviors that might indicate animal stress or maladaptation to the laboratory environment.

Abnormal behaviors in fish include:

- Listlessness
- Changes in feeding habits
- Flashing (scraping against objects in the tank)
- Isolation from other fish
- Gulping air at the surface
- Altered appearance of skin or fins
- Opercular flaring (exaggerated opening and closing of gill covers)

When these behaviors are observed, URAR will evaluate the need for additional environmental enrichment. All changes to enrichment will be approved by the AV and the PI. Enrichment changes will be made for all animals on study, in order to minimize research variability, even if all of the animals are not showing the stereotypical behavior.

3.0 PROCEDURES:

3.1 Social Enrichment – Fish will be group housed whenever possible but in low enough densities to reduce population stress. If group housing is not possible, animals will be housed in a manner that allows for as much visual contact as possible.

3.2 Physical Enrichment - in order of preference

- 3.2.1 Zebrafish and Medaka: plastic or natural plants or structures for hiding and depositing of eggs
- 3.2.2 Mummichog and Catfish: dense substrate that allows for hiding and low light levels or appropriate alternative
- 3.2.3 Sturgeon: deep, dark flowing water or appropriate alternative

3.3 Activity/Food Enrichment - in order of preference

- 3.3.1 Guppy: microalgae based diet (Dagar, 2010)
- 3.3.2 Zebrafish, Medakea, Mummichog and Catfish: varied diet of commercial fish food and live prey
- 3.3.3 Sturgeon: live prey diet that encourages foraging in the upper water column

4.0 RECORDS:

The Animal Care Staff will log provision of enrichment daily according to their facility specific documentation records.

5.0 DEFINITIONS AND REFERENCES:

5.1 Definitions:

- 5.1.1 Animal Use Proposal (AUP): a detailed written description of the procedures involving the use of animals in a research or instructional project.
- 5.1.2 Attending Veterinarian (AV): the veterinarian responsible for the health and well-being of all laboratory animals used at the institution
- 5.1.3 Enrichment: a method of providing animals with the opportunity to behave as they do in the wild, playing, foraging, grooming, and interacting in other ways with one another.
- 5.1.4 Principal Investigator (PI): the scientist who plans and coordinates all phases of the research or instructional work and the protocol.
- 5.1.5 Standard Operating Procedure (SOP): a set of standardized instructions for dealing with routine laboratory procedures

5.2 References:

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Bruton, Michael N. (1996) *Alternative life-history of catfishes.* Aquatic Living Resource
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