



Institutional Animal Care & Use Committee

Zebrafish and Xenopus Larvae Policy

I. Purpose

This document provides guidance to personnel working with larval forms of Zebrafish and Xenopus to determine if IACUC review is required.

II. Scope

This policy applies to all personnel who are working with Zebrafish and Xenopus.

III. Guidance

A. Zebrafish

Current OLAW interpretation of PHS policy considers aquatic species as "live, vertebrate animals" at hatching. Although this is an imprecise stage for zebrafish it can be approximated at 72 hours post fertilization. As such the following guidelines for all research, teaching and testing zebrafish have been established.

1. 0-3 days post fertilization(dpf), a description of activities in an IACUC protocol is not required.
2. 4+ dpf, a description of activities in an IACUC protocol is required. Since early stages (4-7dpf) do not feel pain and distress, the larvae may be classified as Cat. C. An estimate of the number of larval zebrafish(4-7dpf) should be included in the justification.
3. The pain and distress categorization of zebrafish ≥ 8 dpf should be determined by the investigator based on the specific procedures described in the protocol.

B. Xenopus

The hatching process for most Xenopus begins at about 2 dpf (50 hours post fertilization). The hatching process is completed over the next 48 hours and they begin to feed(4 dpf). This corresponds approximately with Nieuwkoop and Faber developmental stage 45. As such the following guidelines for all research, teaching and testing Xenopus have been established.

1. 0-4 dpf, and 0-45 developmental stage, a description of activities in an IACUC protocol is not required.
2. 5+ dpf and 46+ developmental stage, a description of activities in an IACUC protocol is required. Since early stages (5-23 dpf), developmental stages 46-52 do not feel pain and distress, the larvae may be classified as Cat. C. An estimate of the number of larval Xenopus should be included in the justification.

IACUC Approval Date: 09/19/2018

Review Date: 10/19/2022

Issue Date: 10/24/2022

3. The pain and distress categorization of *Xenopus* ≥ 24 dpf and \geq developmental stage 53 should be determined by the investigator based on the specific procedures described in the protocol.

References

1. University of Oregon (2008) Final Report to OLAW on Euthanasia of Zebrafish.
2. National Institutes of Health (2009) Final Report to OLAW on Euthanasia of Zebrafish.
3. https://olaw.nih.gov/sites/default/files/zeb_2016_1344.pdf a. Guidelines for Use of Zebrafish in the NIH Intramural Research Program
4. M. Babošová, P. Vašeková, J. I. Porhajašová & J. Noskovič (2018) Influence of temperature on reproduction and length of metamorphosis in *Xenopus laevis* (Amphibia: Anura), *The European Zoological Journal*, 85:1, 151-158, <https://www.tandfonline.com/doi/full/10.1080/24750263.2018.1450456>
5. Xenbase <https://www.xenbase.org/anatomy/alldev.do>
6. Carlana Ramlochansingh, Francisco Branoner, Boris P. Chagnaud, Hans Straka
 - a. Efficacy of Tricaine Methanesulfonate (MS-222) as an Anesthetic Agent for Blocking Sensory-Motor Responses in *Xenopus laevis* Tadpoles; Published: July 1, 2014
 - b. <https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0101606>