

University of Maryland College Park Animal Care and Use Standard

Animal Accounting for Animal Study Protocols and Census

Purpose: This standard defines the age at which various species must be counted as "animals".

Background: The Animal Welfare Regulations, Public Health Service Policy on Humane Care and Use of Laboratory Animals (PHS Policy), the Guide for the Care and Use of Laboratory Animals, and the Guide for the Care and Use of Agricultural Animals in Research and Teaching apply to vertebrate animals used in research, testing and teaching. These regulatory and oversight documents require that proposals to conduct activities involving animals must contain the species, approximate number of animals to be used and a justification for that number. The Institutional Animal Care and Use Committee (IACUC) evaluates the species, total number of animals requested and rationale for that number during protocol review. The IACUC also submits annual reports to regulatory and oversight bodies that include the total number of animals used in research, testing and teaching throughout the year. This standard provides guidance on when to count animals for animal study protocols (ASPs) and the animal census.

Standards: Most animal species are accounted for at birth, but egg-laying species present some unique challenges, particularly when animal activities propose the use of embryonated eggs or larvae. The UMD IACUC follows the Office of Laboratory Animal Welfare's guidance on egg-laying species when counting animals for research, testing or teaching. Although avian and other egg-laying vertebrate species develop backbones prior to hatching (breaking free of the egg matrix), OLAW interprets the PHS Policy as applicable to the offspring of egg-laying species only after hatching. The egg-laying adult animal is covered by the Policy. Once hatched, larval forms of fish and amphibians have vertebrae and are covered by the PHS Policy. The IACUC recognizes that obtaining an exact count of large numbers of individual animals (e.g., zebrafish larvae and adults) is difficult; therefore, the IACUC will rely on experienced investigators to arrive at a reasonable estimate of the numbers of fish at hatch and older that either have been or will be used. Hatch times that deviate from standard (see below) as a result of variable environmental conditions are permissible but must be described in the protocol and are subject to IACUC approval.

Methodology: Animal use activities involve any animal bred or housed for the intended purpose of research, testing, teaching or exhibition. The following guidelines should be used when estimating animal numbers for ASPs and the animal census:

- Warm-blooded vertebrate animals (all species) are counted at birth (altricial neonatal rodents may be counted at the first post-partem cage change to reduce the potential for cannibalism, etc.);
- **Birds** are counted at hatch;
- **Manipulated embryonated avian eggs** are counted after they reach 80% of their average incubation period (e.g., 17 days post fertilization for chickens);
- **Reptiles** are counted at hatch;
- **Manipulated embryonated reptile eggs** are counted after they reach 80% of their average incubation period;
- **Fish/Amphibians** are counted at birth (viviparous) or hatch (oviparous), when they transition from embryo to protruding mouth larval stage. This may vary by species.

A.3. Animal Accounting for ASPs and Census (Formerly D.1.a.) Approved: 05/24/16 Reviewed/Revised: 10/16/20;

- o Zebrafish are counted at hatch, generally 72 hours post-fertilization (hpf).
- o Xenopus are counted at hatch, generally 72 hpf.