Institutional Animal Care and Use Committee		UNTHSC
Title: Guidelines for the Use of Chicken/Avian Embryos		
Document #: 060	**Version #:** 01	
Approved by IACUC Date: February 25, 2025		

A. BACKGROUND INFORMATION

a. According to U.S. regulatory agencies, avian embryos are not considered live animals. However, research activities using embryonated avian eggs may require oversight by the IACUC under certain circumstances. The scientific community upholds a general consensus that embryos greater than 75% of embryonic development may be sufficiently developed to experience pain, with 66% being the earliest stage of embryonic development where pain can possibly be felt. Additionally, if embryos of egg-laying species hatch, intentionally or unintentionally, they are live vertebrate animals and thus require oversight by the IACUC.

b. Domestic chicken embryos, which hatch in approximately 21 days, are considered the model species. With a 21-day incubation, pain can possibly be felt as early as day 13 of embryonic development. If other avian species are used, the guidelines should be adjusted based on relative time to hatch for that species.

c. OLAW expects assured institutions to have policies and procedures in place that address the care or euthanasia of animals that hatch unexpectedly. Euthanasia techniques should be consistent with the AVMA Guidelines, unless the deviation from these guidelines is justified in the protocol for scientific or medical reasons.

B. RESPONSIBILITIES

a. It is the responsibility of all investigators, staff, students and animal care staff using animals in research or teaching at the University of North Texas Health Science Center to follow this procedure.

C. PROCEDURES

a. In the event of intentional or unintentional hatching, the bird is now considered a live vertebrate animal and proper euthanasia procedures should be followed.

b. **Euthanasia prior to 66% development:**

Embryos can be euthanized via hypothermia by placing the eggs in a -20°C freezer for greater than four hours.

c. **Euthanasia at 66% development and beyond:**

Embryos at or beyond 66% development should be euthanized using humane methods. Recommended methods are CO_2 exposure, anesthetic agent exposure or decapitation. Embryos being euthanized by CO_2 should be exposed to 90% CO_2 for a minimum of 20 minutes.

d. All laboratory animals must be euthanized in a timely manner to prevent/ alleviate animal suffering, either as described in the approved protocol according to experimental endpoints, or as soon as necessary if established criteria for humane endpoints are reached. All IACUC approved protocols must include a description of the methods for euthanizing the animals, and how death will be confirmed, especially in animals receiving anesthetics or CO_2 prior to euthanasia. Refer to IACUC SOP 006 Euthanasia Guidelines.
e. Proper disposal of embryos must follow IACUC SOP 016 Carcass Disposal, including bringing them to DLAM staff for proper disposal whether they are euthanized in the lab or animal facility.

D. REFERENCES

a. Garcia-Austt E., Jr. Development of Electrical Activity in Cerebral Hemispheres of the Chick Embryo. Proc. Soc. Exp. Biol. Med. 1954;86:348–352. doi: 10.3181/00379727-86-21095.
b. Embryonated Avian Eggs in Research and Teaching | Illinois Research Animal Care and Safety
c. FAQs | OLAW
d. Policies & Guidelines UNC Charlotte - Division of Research
e. AMVA Guidelines for Euthanasia of Animals: 2020 Edition
f. IACUC SOP 006 Euthanasia Guidelines
g. IACUC SOP 016 Carcass Disposal