This document describes standard operating procedures for in vivo bioluminescence imaging including animal transport, anesthesia and biocontainment.

1) IVIS imaging device training contact:
   
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2) Investigator Responsibilities:
   Procedures involving live animals must be covered by an IACUC approved protocol that includes the following information:
   
a) An explanation of the purpose of the study  
b) The number of animals  
c) Imaging frequency and the expected number of imaging sessions  
d) A description of other procedures performed on these animals either prior or after imaging (such as administration of luciferin)  
e) The location of the imaging device is in HEB 043 and you must sign out the device using the on-line calendar.  
   i) Room - DLAR HEB043 (Procedure room with Faxitron/X-Ray & IVIS/imager)  
f) Animals that are to be imaged should be fed a diet free of alfalfa and alfalfa byproducts as this can negatively influence the image. DLAR carries two alfalfa free diets, Teklad 2916 (Irradiated standard diet) and Teklad 2919 (Irradiated 19% protein breeder diet).

3) Transport of Animals
   a) Animals on study may be transported from neighboring rooms on a cart in their home cage/s. If hazardous the sealed cage must be sprayed with disinfectant prior to removing from the animal room and transporting to the IVIS system.  
b) Contact the DLAR office (383-4310) to coordinate transport from other facilities in different buildings.

4) General Procedures:
   The following general protocol will be followed for imaging. The details are limited to procedures as individual experiments are described in each investigator’s approved protocol (list or expand on those described below to reflect your usage):
   
a) Animals may be injected with pathogen-free bioluminescent or fluorescent cell lines, dyes OR injected with a vehicle as a control (when appropriate for optical imaging). Handling and use of any BSL2 cell line(s) must be approved on the PIs IBC protocol with a standard operating procedure prior to use.
b) Animals will be induced using the clean plexiglass induction box for anesthetization via inhaled isoflurane. Once anesthetized the animals will be carefully relocated from the induction box to the heated stage with nose cones inside the IVIS systems.

c) Alternative anesthetics may be used following veterinary consultation.

d) For bioluminescent optical imaging, 150 mg/kg D-Luciferin will be given to each mouse via intraperitoneal injection, with a maximum volume of 1 ml.

e) Imaging will be done efficiently minimize the time under anesthesia and typical would take 5-20 minutes.

f) Following the imaging session animals are returned to their home cage and observed until they can move about the cage.

g) Animals that display clinical signs of illness which place them at risk for anesthetic death, will not be imaged.

h) Active anesthetic scavenging will be employed to minimize personnel exposure to anesthetic gases.

5) Record Keeping:

a) A permanent written log listing the date, name of personnel using the equipment, the PI and their IACUC number will be maintained with the imaging device. You should always keep additional record of the process in your lab notebook.

6) Working with Hazardous Animals in the IVIS:

a) Only Chemical Hazardous levels 2 or lower and Biosafety Level 2 or lower are permitted.

b) The lab must have taken Hazard Training through DLAR

c) Hazardous animal cages must be sprayed off with disinfectant in their designated hazard room prior to being transported to the IVIS room 043.

(d) Hazardous cages must only be opened in the Biological Safety Cabinet “BSC”.

e) All work surfaces and equipment (counter tops, BSC, imaging chamber, anesthetic equipment) inside and outside the camera must be disinfected before and after each imaging session.

f) Use paper towels sprayed with disinfectant to clean the inside of the IVIS chamber. Never sprayed directly into imaging chamber!

(g) Biohazardous waste will be place in a biohazard container located within the room.

h) Any spills will be handled as potential biohazardous.

i) Approved Disinfectants such as isopropyl alcohol will be available in the procedure area.

ii) Do not use Peroxigard® as this can negatively influence the imaging of your animals.

j) All personnel working with infected animals will wear the approved personal protective equipment. Procedures that have the potential to aerosolize infectious agents will be conducted within a biosafety cabinet “BSC” as much as possible, but the imaging is outside of the BSC which may require respiratory protection such as an N-95 mask or PAPr.
Animal Handling Procedures
Bioluminescence Imaging

a) Each person using the IVIS imaging system must be specifically trained and authorized to use the equipment.

b) Personnel using the IVIS camera must record their use in the log book including: Date, Name of user, PI name, IACUC number and name of infectious agent (if applicable).

c) Personnel must wear proper posted PPE while working with animals.

d) Animals must be transported to and from animal rooms on a cart in microisolator cages.

e) Animals will be returned to their home cage or a separate enclosed container and observed continuously during recovery from anesthesia.

f) All surfaces and equipment (counter tops, the biosafety cabinet, imaging chamber, and anesthetic equipment) must be disinfected between groups of animals.

g) Approved Disinfectants such as isopropyl alcohol will be available in the procedure area. Use paper towels sprayed with disinfectant to clean the inside of the IVIS chamber. Never sprayed directly into imaging chamber!
   i) Do not use Peroxigard® as this can negatively influence the imaging of your animals.

h) Dispose of contaminated biohazardous materials in the biohazard container located within the room.

i) Wash hands after handling animals.