A. PURPOSE

To describe the guidelines on performing rodent irradiation and proper care following irradiation.

B. RESPONSIBILITY

1. The **PI** is responsible for ensuring the training of research staff that will be performing this SOP. It is the responsibility of laboratory personnel using this procedure to read, understand, receive training for, and agree to follow the procedure described in this SOP.

C. DEFINITIONS AND/OR ACRONYMS

1. **Irradiation:** A type of ionizing irradiation, in which the source may be 137Cesium, 60Cobalt or high energy x-rays. At the University of Arizona we currently only have X-ray irradiators.
2. **Gray (Gy):** The SI unit of absorbed radiation dose due to ionizing radiation. 1Gy = 100cGy = 100rad
3. **Fraction of Dose:** The total irradiation dose is split into two or more equal parts separated by a time interval; to minimize morbidity and mortality.
4. **TBI:** Total Body Irradiation
5. **BMT:** Bone Marrow Transplant

D. CONSIDERATIONS

1. The dose of irradiation should take into consideration the strain and age of the rodent as well as the purpose of the experiment.
    - **i.** Different rodent strains have varying sensitivity to radiation. BALB/C mice are more sensitive to irradiation compared to C57B/6 mice.
        1. Relative radiosensitivity of mouse strains: 129S ≤ SJL ≤ C3H ≤ C57BL/6 << BALB/c << scid
    - **ii.** Resistance to irradiation:
        1. Neonatal: relatively resistant
        2. Weaning (20-30 days): most sensitive
        3. Young adulthood (3-4 months): Most Resistant
        4. Aged (6 months +): declining resistance throughout age
    - **iii.** Doses will vary by irradiator type and radiation source.
    - **iv.** Fraction doses should be considered, if appropriate, to reduce morbidity and mortality.
    - **v.** Consider giving antibiotic treatment prior to irradiation. UAC veterinary staff can provide recommendations if needed.

E. Rodent Irradiation Best Practices

1. Ensure irradiators have been properly calibrated and users are trained.
2. Validate specific sources of radiation and dosage rates for biological responses and models.

F. PROCEDURE

1. **Policy for IACUC Protocols**
    - **i.** Irradiation must be listed and described in detail in the IACUC protocol, including planned dose or dose range and monitoring plan.

1. Details must include who will be performing the irradiation. If a core will be performing the procedure, describe whether animals will be transferred to the Core protocol or if core personnel will be listed on the PI's protocol.
    a. If you will be using a core, contact them ahead of time to help coordinate and plan the experiment.
    b. IACUC approval must be shared with the core prior to starting the experiment.
2. The location of irradiation is listed on the IACUC protocol.

ii. Irradiation must be scientifically justified in the protocol.
iii. Unless published literature references are available for the specific dose and strain, a pilot study to determine the best dose is recommended if the PI is starting a new study or using a new strain of rodent.
iv. Irradiation listed on an IACUC protocol must be approved by RLSS prior to starting any experiment.

2. **Irradiation**
    i. Dose is dependent on many factors, including strain of rodent, age, and whether it is targeted or whole-body irradiation. See the table below of published doses of irradiation for various rat and mouse strains.
    ii. Doses of 800-1300 cGy (7-13 Gy) are myeloablative in mice.
    iii. Single exposure
        1. Mice irradiated with 8-10 Gy (exact dose is strain dependent).
        2. If a higher dose is requested, scientific justification is required in addition to literature references. If this is an initial experiment, a pilot study is required.
    iv. Split exposure
        1. The total dose will be delivered in two or more sessions, separated by at least 3 hours.
    v. Reduced exposure
        1. Some strains, such as SCID mice, are highly sensitive to irradiation and should receive a total dose of 3-5 Gy.
    vi. Cages of irradiated rodents should be identified with the following information:
        a. Date of irradiation
        b. Dose of irradiation
            i. List the information either on the cage card or another notecard.
    vii. Sterilized caging, food, and water is recommended for irradiated rodents.
3. **Post Procedure Monitoring Policy**
    i. Animals exposed to irradiation must be monitored and findings documented daily for the first 5 days, then at least once weekly. Once the PI has established a protocol and experience with a particular strain of rodent and dose, the monitoring can decrease to 3 times per week.
        1. If animals experience morbidity or mortality, then daily checks are required.
    ii. Wet or softened food on the floor of the cage and/or napa nectar is recommended as whole body irradiation can cause dental damage.
    iii. Giving isotonic fluids (IP or SC) is recommended after irradiation and again at 24 hours.

iv. Possible Clinical Signs

1. Weight loss, lethargy. Hunched posture, rough coat, skin burns, anemia, infection, intestinal bleeding, graying of hair (black haired rodents), development of secondary neoplasia, and damage to incisors.
2. When clinical signs or complications are observed, UAC veterinary staff should be notified immediately to decide a course of action.

Mouse Irradiation Dosage Table (2x indicates fractionated doses are given 3 hours apart)

Mouse Strain	Protocol	Dose (Gy)	Response to Dose	Reference
C57BL	TBI and BMT	2 x 5.5	Survival	The Mouse in Biomedical Research. Vol 3 Fox J.G et al. Eds 2nd Ed 2007. Academic Press.
C57BL	Hematopoietic ablation	3.5-6, 2 x 5.5	Survival	
C57BL/6	Complete myeloablation	9-11	Lethal	Taconic
	Fractionated Dose	6 + 6 within 3-4 hrs	Survival	
	Partial myeloablation	3.5-6	Sublethal	
BALB/c	Complete myeloablation	7-8	Lethal	Taconic
	Partial myeloablation	6 or less	Sublethal	
Scid	Immunosuppression	0.5-2.5		Taconic
C57BL,BALB/c, B6CF1	LD50/6, LD50/30	0.8, 1.4 (Source 60Co)	F1 hybrid is similar to response of C57BL/6 and different from BALB/c	Hanson et. al. 1987. Comparison of intestine and bone marrow radio sensitivity of the BALB/c and the C57BL/6 mouse strains and their B6CF1 offspring. Radiat Res 110:340-352.
FVB/N, C57BL, Rag1-/-iNOS, FVB/N Tie2-GFP	TBI and BMT	10-22 Gy (Source 137Cs)	Germ free mice were much less sensitive to irradiation enteritis than conventional mice	Crawford, PA and Gordon, JI Microbial regulation of intestinal radiosensitivity. PNAS 102(37); Sept. 13, 2005.

Rat Irradiation Dosage Table

Rat Strain	Protocol	Dose (Gy)	Response to Dose	Reference
Wistar, M, 100g	TBI	14.4	Rats begin to die on day 5 post exposure	Kassayova, E. et.al. 1999. Two-Phase Response of Rat Pineal melatonin to Lethal Whole-Body Irradiation with Gamma Rays. Physiol. Res. 48:227-230.
Wistar, M, 100g	TBI	9.6	Rats begin to die on day 10 post exposure	
Wistar, M, 100g	TBI	4.8		

G. REFERENCES, MATERIALS, AND/OR ADDITIONAL INFORMATION

1. Experimental Mouse Shared Resource (EMRS): https://cancercenter.arizona.edu/researchers/shared-resources/experimental-mouse
2. Research Laboratory and Safety Service (RLSS): https://research.arizona.edu/compliance/RLSS