

**Stony Brook University
Institutional Animal Care and Use Committee (IACUC)**

PRODUCTION OF SUBCUTANEOUS TUMORS IN RODENTS

A. Anesthesia

1) Anesthesia may be used for subcutaneous tumor inoculation but is not required.

2) Anesthetic Agents

Mice → Isoflurane (Drop method or vaporizer)
Ketamine (40-120 mg/kg) and Xylazine (10 mg/kg), IP
Pentobarbital (50 mg/kg), IP

Rats → Isoflurane (Drop method or vaporizer)
Ketamine (60-75 mg/kg) and Xylazine 5-13 mg/kg), IP
Pentobarbital (40-60 mg/kg), IP

3) The use of these and any other anesthetic agents must be listed in the IACUC application.

B. Anesthesia Monitoring

During the injection procedure the following parameters must be monitored at a minimum of 5 minute intervals:

- Respiratory rate
- Response to noxious stimulus (ie toe pinch)
- Spontaneous movement

C. Anesthesia Recovery Monitoring

1) During recovery from anesthesia, the following parameters must be monitored at a minimum of 15 minute intervals until the animal is ambulatory.

- Respiratory rate
- Movement
- Ability to maintain sternal recumbancy

2) To protect animals from hypothermia they should never be placed on metal surfaces. Animals should be placed on a water re-circulating heating blanket, or covered well, to conserve body temperature.

3) It is estimated that animals will recover within 30-60 minutes.

D. Parameters for Tumor Production

1) The maximum tumor size is 2.0 cm in the largest diameter. However, tumors may need to be harvested before this maximum size is reached depending on tumor location and growth pattern. Important factors for early tumor harvesting include the ability of the

animal to move around the cage normally, reach food and water, and evidence of tumor infection or necrosis.

- 2) No more than two tumors may be inoculated per animal.
- 3) There is potential for contamination of transplantable tumors and tissue cell lines with a variety of murine pathogens (e.g. LCM, MHV, MVM). Therefore, all tumor and tissue cell lines are required to undergo IMPAC testing or MAP/RAP testing before final IACUC approval will be granted. This requirement also pertains to animals that have been inoculated with this tissue at other facilities and are being transferred to the DLAR. Contact the DLAR Veterinary staff for testing information.

E. Tumor Inoculation Procedure

- 1) A 23-25 gauge needle is preferred for tumor cell inoculation.
- 2) The back and flank are the approved sites for tumor placement. Any other location requires scientific justification in the IACUC application.

F. Analgesia

None required

G. Adverse Effects

Potential adverse effects of subcutaneous tumor growth include:

- erythema of the tumor or surrounding skin
- ulceration, infection, bleeding and/or necrosis of the tumor
- metastasis of the tumor leading to systemic signs such as weight loss, respiratory distress, diarrhea, lethargy, abdominal distension, unkempt appearance
- self-mutilation due to pruritis (itching)
- limited ability to move and/or reach food and water

H. Clinical Monitoring and Management

- 1) Post inoculation, animals must be monitored 3 times weekly until a palpable tumor nodule is present.
- 2) Once a nodule is present, the animals should be monitored daily for the potential adverse effects listed above.
- 3) If tumor growth is rapid, daily monitoring should include weekends and holidays.

I. Early Endpoints

- 1) Animals must be euthanized if the tumor becomes ulcerated, infected, necrotic, or bleeds.
- 2) If an animal, due to the size or location of tumor, cannot move normally or cannot easily reach food and water, they must be euthanized.