

Research Animal Standard Operating Procedures (SOP) must meet the following criteria:

1. Describe procedures or activities involving research animal(s) common to more than one research project.
2. Support the handling and or performance or undertaking of a procedure(s), involving an animal, in the same way on each occasion it is performed.
3. Describe a procedure or activity involving a research animal(s) undertaken by more than one person; and
4. Describe a procedure or activity involving a research animal(s) that will be undertaken in more than one location.

Name of Procedure	Intramuscular injection in small rodents	
Species	Rat and mouse	
ACEC	Reference	SOP#77- Aug 23 - Injection, intramuscular-rodents
	Author	Jenny Smart
	Version	1.4
	Date approved	25 August 2023
	Date for review	31 August 2026
	Procedure classification 1. Observation involving minor interference 2. Animal unconscious without recovery 3. Minor conscious intervention 4. Minor surgery with recovery 5. Major surgery with recovery 6. Minor physiological challenge 7. Major physiological challenge	3
Ethical considerations	1. Respect for animals must underpin all decisions and actions involving the care and use of animals for scientific purposes. 2. The procedure must be performed according to current best practice to support the wellbeing of the animal. 3. Persons performing this procedure must be competent in the procedure or be under the direct supervision of someone who is competent.	

Details

Purpose

To describe the procedure for safely performing an intramuscular injection in rats and mice with a minimum of stress to the injected animal.

Description of procedure

EQUIPMENT

1. Sterile syringe of sufficient size to contain the volume of injectate.
2. Sterile hypodermic needle. The needle should be of the smallest gauge possible through which the injectate can pass. Viscous solutions will require a larger gauge needle. In general use a 25G needle for rats and a 29G needle for mice.
3. Antiseptic such as chlorhexidine in 70% ethanol in water
4. Cotton gauze swabs
5. Sharps container
6. Injection solution

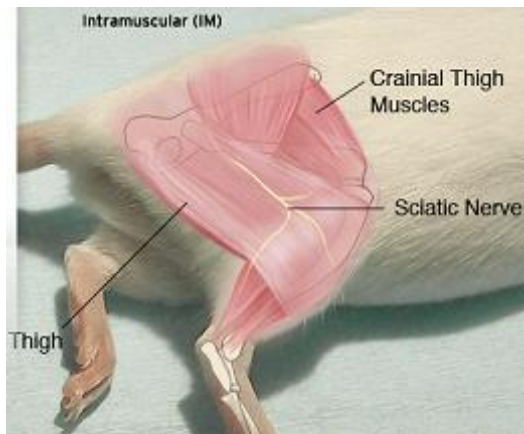
NOTE:

1. Injections should be carried out in a quiet area away from other animals.
2. Animals should be acclimatised to handling before attempting injections. Use the acclimatisation period between delivery of the animal to the research holding facility and the first injection to condition the animal to the catching and restraint techniques used for the injection.
3. The maximum volume injected via the intramuscular route should be no more than:

Species	Mouse	Rat
Maximum Volume for adult animal	0.05 ml	0.1 ml/ site

PROCEDURE

1. Attach the needle to the syringe, apply antiseptic to the rubber diaphragm of the bottle of injectate, carefully uncap the needle and insert it through the rubber diaphragm.
2. Invert the bottle of injectate and draw up the calculated volume of injectate into the syringe.
3. Put the needle and syringe to one side. DO NOT recap needle (to avoid needle stick injuries). Avoid contaminating the sterile needle by resting the needle hub or top of syringe against the needle cap so that the needle is elevated.
4. The preferred muscle group for injection in both species is the quadriceps muscle group in front of the femur (cranial thigh muscles). The sciatic nerve runs through the caudal thigh muscles; hence this muscle group should be avoided as sciatic nerve injury, which is very painful, can result if the caudal thigh muscles are used.



5. Catch and restrain the animal to be injected as follows:
 - a. **Mice**- use tunnel or cup handling to remove the mouse from the home cage and place into a restrainer or mouse bag providing access to the hind legs or use an assistant to restrain the mouse by scruffing and immobilising the hind limb.
 - b. **Rats**- grasp the rat gently around the upper body and place the rat into a restrainer or rat bag providing access to the hind legs or use an assistant to restrain the rat by grasping the upper body in one hand and immobilising the hind limb in the other.
6. Prepare the site of injection by using a cotton gauze swab soaked with antiseptic. Apply the antiseptic to the injection site by wiping against the lay of the hair so that the antiseptic reaches the skin.
7. Ensure that the hind limb is immobilised. Intramuscular injections are painful and movement of the hind limb during injection will result in muscle damage. In small rodents the femur can be grasped between the finger and thumb of the non-dominant hand. This immobilises the leg and pushes the cranial thigh muscle into a suitable position for injection.
8. Holding the needle perpendicular to the skin, insert the needle into the middle of the muscle mass.
9. Draw back on the hub of the needle to ensure that the needle has not penetrated a blood vessel. If blood is seen in the hub of the needle, withdraw the needle and start the injection process again.
10. If the needle is correctly sited, inject slowly and remove the needle smoothly.
11. Place needle and syringe into sharps container.
12. Release the animal back into its cage and observe for any signs of abnormal behaviour.

Images of Intramuscular Injections

MOUSE



RAT



References

1. Refining procedures for the administration of substances” Report of the BVA/WF/FRAME/RSPCA/UFAW Joint Working Group on Refinement. *Laboratory Animals* (2001) 35, 1 41
<https://journals.sagepub.com/doi/pdf/10.1258/0023677011911345>
2. Basic Bi methodology for laboratory mice
http://www.theodora.com/rodent_laboratory/injections.html
3. <https://researchanimaltraining.com/articles/intravenous-injection-in-the-mouse/>
4. This site was previously known as Procedures With Care. It links out from the NC3Rs site

Note: Photos also courtesy of these sites

Research and Innovation Division
Research Animal Standard Operating Procedure
SOP#77



ACEC Chair

