Research and Innovation Division

Research Animal Standard Operating Procedure

SOP# 103

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	Superovulation of mice	
Species	Mouse	
ACEC	**Reference**	SOP#103 – May23 - Superovulation of mice
	Author	Jenny Smart
	Version	1.2
	Date approved	26 May 2023
	Date for review	26 May 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

Appropriate administration of gonadotrophins in female mice can induce them to ovulate a greater than usual number of oocytes at predictable time (superovulation).

This document describes the procedures used to superovulate female mice to produce oocytes for use in research protocols.

Description of procedure

EQUIPMENT:

1. Sterile syringe (0.3- 1.0mls)
2. Sterile hypodermic needle 30-25G
3. Antiseptic such as chlorhexidine in 70% ethanol in water
4. Cotton gauze swabs
5. Sharps container
6. Pregnant Mare Serum Gonadotrophin (PMSG)- Folligon
7. human Chorionic Gonadotrophin B (hCG)- Chorulon
8. watchmaker's forceps
9. fine scissors

PROCEDURE:

1. Follow ACEC Approved **SOP#78- Injection, intraperitoneal- rodents**
2. For each female mouse to be superovulated:
    a. Inject Gonadotrophin (PMSG) intraperitoneally at 5-7.5IU/ 20g mouse
    b. 48 hours later inject Chorionic Gonadotrophin (hCG) intraperitoneally at 5IU/ 20g mouse
    c. Euthanase mouse 12-15 hrs following second injection by inhalation of carbon dioxide following **SOP#74- Euthanasia using Carbon Dioxide Gas- Rodents and small birds**
3. Dissect reproductive tract and collect upper uterine horn and oviduct with fine scissors. Remove to petri dish containing appropriate media with fine forceps.
4. View oviduct using stereomicroscope and puncture oviduct with watchmakers forceps, releasing cumulus oocyte complexes (COCs) into media. Transfer COCs to fresh droplet of media with fine pipette.

Substances administered

Drug name (**generic** name, not trade name)	Dose rate (**mg/kg body weight**)	**Volume**	**Route**	Timing of administration, and frequency	Purpose
Pregnant Mare Serum Gonadotrophin (PMSG)-	5-7.5IU/ 20g	50ul	IP	Initial injection	Follicular recruitment
Chorionic Gonadotrophin (hCG)	5IU/ 20g	50ul	IP	48-56 hours after PMSG injection	ovulation

References

Administration of Gonadotropis for Supoerovulation in Mice
Authors: Richard Behringer, Marina Gertsenstein, Kristina Vintersten, Andras Nagy,
Cold Spring Harbor Laboratory Press, 2018, pp 24-27

https://www.jax.org/news-and-insights/1998/july/superovulation-technique

ACEC Chair