Electroporation-Mediated Gene Transfer to Rodent Embryonic Tissue IACUC Standard Procedure Effective Date: May 2024



Description of procedure:

Electroporation is a safe and efficient method to transfer naked plasmid DNA into the cells of various tissues. The major applications of the technology include gene function analysis, gene therapy, DNA vaccination, etc. Design requirements differ considerably for these applications.

Suggested Supplies:

- Electroporation System
- Beveled needle, micropipette or microinjection capillary tube
- Sterile gel
- Vevo 770 ultrasound system (or similar)
- Fiber-optic trans-illuminator
- Sterile surgical equipment
- Warm sterile saline
- Suture material
- Plasmid solution

Procedure Steps:

- 1. The animal is anesthetized following <u>UCSF Rodent Anesthesia Guidelines</u>. Anesthetic and analgesic agents are administered as per Section I Agents in the IACUC approved protocol. Peri-operative analgesia is required.
- 2. The surgeon will follow IACUC Guidelines for Rodent Survival Surgery.
- 3. Electroporation Procedure:

Surgical Procedure:

The abdomen of a pregnant (12.5-16.5 d postcoitum) female will be prepared according to IACUC <u>Guidelines for Rodent Survival Surgery</u>. A ventral midline incision will then be made in the skin and peritoneum, approximately 1.5cm to 2cm in length, to expose the uterine horns. Care should be taken to maintain asepsis and keep the externalized uterus and surrounding tissues moist with warm sterile saline. Embryos/ fetus can be trans-illuminated through the uterine wall using a fiber optic light to visualize recipient tissues. Other modalities for visualization of the targeted area can be used such as ultrasound. After the electroporation procedure is completed, the uterine horns will be returned to the peritoneal cavity, and the incision will be closed in two layers.

Electroporation:

A plasmid solution is delivered to the targeted site by trans-uterine injection using a small-bore hypodermic needle or beveled glass micropipette or microinjection capillary tube. Immediately after plasmid delivery, short electrical pulses are applied. Pulses are delivered by two pre-gelled electrodes placed on each side of the target tissue/organ.

Depending on the tissue, organ, and/or age of the animals, these parameters may need to be optimized: number of pulses, tduration and strength of each pulse, and interval between pulses.

Protocol requirements:

- In the "Procedures" section G of the RIO protocol application, indicate the experiments involving electroporation.
- In the "Agents" section (I) of the RIO protocol application, list the appropriate anesthetic and analgesic agents required for this procedure.
- In the "Adverse Effects" section (J) of the RIO protocol application, list infection, pain, and embryo mortality

References:

- Huang CC, Carcagno A. Electroporation of Postimplantation Mouse Embryos In Utero. Cold Spring Harb Protoc. 2018 Feb 1;2018(2)
- Jove: In Utero Intra-ventricular Injection and Electroporation of E16 Rat Embryos <u>http://www.jove.com/Details.php?ID=236</u>