



Coronary Artery Ligation

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Summary:

This is a mouse model of acute myocardial infarction. This protocol is suitable for studying mouse models thought to be relevant to coronary ischemia or the development of congestive heart failure.

Reagents and Materials:

Reagent/Material	Vendor	Stock Number
Ketamine	Patterson veterinary	07-803-6637
Xylazine	Patterson veterinary	07-869-6707
Isoflurane 1-3%	Patterson veterinary	07-893-1389
Buprenorphine	Patterson veterinary	07-891-9756
Meloxicam	Patterson veterinary	07-893-1368
5-0 Proline suture	esutures	8860H
7-0 Proline suture	esutures	M860

Protocol:

WARNING HAZARDOUS CONDITION WARNED AGAINST. This comment describes a hazardous condition to which the technician may be exposed in the performance of this protocol. It also contains directions on how to avoid or minimize the danger. Warnings are always and only used for personnel safety, and precedes the first step that will expose

Expected procedure duration:

20-40 minutes

Adequacy or depth of anesthesia is monitored by:

Respiratory Rate and Toe Pinch

Frequency of anesthesia depth assessment:

At the start of surgical procedure, a toe or ear pinch can be used to assess the depth of anesthesia. Visual monitoring should be performed thought-out the procedures, as well as toe/ear pinches.

Deviations from expected behavior Should be noted.

Anesthesia Regimen:

Ketamine (80-100mg/kg), Xylazine (5-20mg/kg) or Isoflurane 1-3%

Pre-surgical Analgesics:

Approximately 30 minutes prior to undergoing the surgical procedure, mice receive an S.C. injection of Buprenorphine (0.05mg/kg) and Meloxicam (5mg/kg).

Surgical prep:

Aseptic technique will be maintained by:

Clipping/shaving fur around incision site, Sterile Instruments.

Isoflurane, heating pad, forceps, scissors, needle driver, eye ointment, 5-0 Proline, 7-0 Proline suture, 20g iv catheter sheath, PE 90 tubing, 1 ml-syringes, 0.9% NaCl, Sterile gloves, Povidone-iodine, 70% ETOH

Myocardial Ischemia/Infarction Procedure:

1. Anesthetize the mouse and ensure depth of anesthesia with a toe pinch.
2. Shave the left thoracic area of the mouse.
3. The mouse is intubated using a 22-G angiocatheter sheath and placed on a rodent ventilator with the left chest up, secure feet to surgical area with tape.
4. Prep the surgical field with 70% isopropanol as well as Betadine solution and drape.
5. A left side thoracotomy is performed between the 3rd and 4th intercostal space and opened approximately 6-7 mm with a small retractor. Use caution to avoid injury to the lung.
6. After making an opening the pericardium, the left anterior descending coronary artery is identified using a microscope. A 7-0 Proline suture is placed just under the descending coronary artery without entering the left ventricle. This can be tied permanently to ligate the LAD for an acute ischemia model or alternatively, the 7-0 proline suture is secured over a 3-4 mm piece of PE-90 tubing to ligate the LAD for 30 minutes, and then the suture is removed to facilitate reperfusion of the heart for an ischemia reperfusion model. The suture is placed 2 mm below the left atrium for a "large" infarction; 3 mm below the left atrium for a "medium" infarction; or 4 mm below the left atrium for a small infarction.
7. Two separate 6-0 Proline sutures are place to close the rib cage
8. A 20g iv catheter sheath is left between the sutures to evacuate air after closure is complete.
9. The dermis is sutured closed with 6-0 Proline, Tissue clips or glued with surgical glue
10. Air is removed from thoracic cavity via the sheath and a 1-mL syringe. Gently pinching the skin around the sheath slowly draw back on the syringe while withdrawing the sheath.
11. Animal is weaned off anesthesia and extubated when spontaneous breathing resumes.
12. Injection 0.5 ml of 0.9% NaCl subcutaneously to each mouse to prevent dehydration.

Post-procedure Analgesics:

Buprenorphine (0.05mg/kg) every 12 hours, for 72 hours post-op.

Meloxicam (5mg/kg) every 24 hours, for 72 hours post-op

Post-procedure Monitoring:

Mice are monitored 2x daily for the first 72 hours after the surgery. Thereafter, mice are monitored at least 3x per week.