

Aug 12, 2019 Version 2

U Mass - Hind Limb Ischemia V.2

DOI

dx.doi.org/10.17504/protocols.io.56jg9cn



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DOI: dx.doi.org/10.17504/protocols.io.56jg9cn

External link: <https://mmpc.org/shared/document.aspx?id=324&docType=Protocol>

Protocol Citation: Mark Kelly, Timothy P. Fitzgibbons 2019. U Mass - Hind Limb Ischemia. **protocols.io**
<https://dx.doi.org/10.17504/protocols.io.56jg9cn>

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Protocol status: Working

We use this protocol and it's working

Created: August 05, 2019

Last Modified: August 12, 2019

Protocol Integer ID: 26539

Keywords: Hind Limb Ischemia, peripheral artery disease, vascular remodeling



Abstract

Summary:

This is a mouse model of hind limb ischemia, a technique involving an interruption in the arterial blood supply to tissue in the hind limb. This model is used to study peripheral artery disease and vascular remodeling.

Materials

MATERIALS

⊗ Ketamine **Patterson Veterinary Catalog #07-803-6637**

⊗ Xylazine **Patterson Veterinary Catalog #07-869-6707**

⊗ Isoflurane 1-3% **Patterson Veterinary Catalog #07-893-1389**

⊗ Buprenorphine **Patterson Veterinary Catalog #07-891-9756**

⊗ Meloxicam **Patterson Veterinary Catalog #07-893-1368**

⊗ 6-0 Prolene suture **Patterson Veterinary Catalog #07-824-3204**

⊗ 7-0 Silk suture **Patterson Veterinary Catalog #07-824-1501**

1 Expected procedure duration:

30 minutes

2 Adequacy or depth of anesthesia is monitored by:

Respiratory Rate and Toe Pinch

3 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 throughout the procedures, as well as toe/ear pinches.

Deviations from expected behavior Should be noted.

4 Anesthesia Regimen:

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

5 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).

6 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, 6-0 Prolene, 7-0 silk suture, 1 ml-syringes, 0.9% NaCl, Sterile gloves, Povidone-iodine, 70% ETOH

7 Surgical Procedure:

(1). Anesthetize the mice and ensure depth of anesthesia with a toe pinch.

(2). Place the mice supine on the heating pad.

(3). Remove the hair from 0.5 cm above the elbow to 0.5 cm below the knee joint of surfaces to be joined.

- (4). Prep the surgical field with 70% isopropanol as well as Betadine solution and drape the mice.
- (5). Apply ointment to animal's eyes.
- (6). Confirm depth of anesthesia with a toe pinch.
- (7). Make an incision in the skin from the medial thigh towards the knee.
- (8). Blunt dissect away subcutaneous fat tissue to reveal the underlying femoral artery.
- (9). Pierce the membranous femoral sheath to expose the neurovascular bundle. Dissect and separate the femoral artery from the femoral vein and nerve from the inguinal ligament to the epigastrica.
- (10). Place 2 7-0 silk sutures around femoral artery 2mm apart and ligate. Divide the femoral artery between the 2 ligating sutures.
- (11). Close the skin incision using 6-0 Prolene suture.
- (11). Injection 0.5 ml of 0.9% NaCl subcutaneously to each mouse to prevent dehydration.

8 **Surgical Procedure (alternate):** Severe ischemic model

- (1). Anesthetize the mice and ensure depth of anesthesia with a toe pinch.
- (2). Place the mice supine on the heating pad.
- (3). Remove the hair from 0.5 cm above the elbow to 0.5 cm below the knee joint of surfaces to be joined.
- (4). Prep the surgical field with 70% isopropanol as well as Betadine solution and drape the mice.
- (5). Apply ointment to animal's eyes.
- (6). Confirm depth of anesthesia with a toe pinch.
- (7). Make an incision in the skin from the medial thigh towards the knee.
- (8). Blunt dissect away subcutaneous fat tissue to reveal the underlying femoral artery.

(9). Pierce the membranous femoral sheath to expose the neurovascular bundle. Dissect and separate the femoral artery, femoral vein and nerve from the inguinal ligament to the sapheno-popliteal bifurcation.

(10). Ligate the femoral artery using 7-0 silk sutures between the superficial epigastric artery and the sapheno-popliteal bifurcation.

(11). Close the skin incision using 6-0 Prolene suture.

(11). Injection 0.5 ml of 0.9% NaCl subcutaneously to each mouse to prevent dehydration.

9 **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

10 **Post-procedure Monitoring:**

Mice are monitored 2x daily for the first 5 days post the surgery. Thereafter, mice are monitored at least 3x per week.