Vezina Lab Mouse Kidney Capsule Implant Protocol

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Aug 18, 2020

1 Works for me This protocol is published without a DOI.

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PROTOCOL CITATION

Chad Vezina 2020. Vezina Lab Mouse Kidney Capsule Implant Protocol. **protocols.io** https://protocols.io/view/vezina-lab-mouse-kidney-capsule-implant-protocol-bjv6kn9e

LICENSE

CREATED

Aug 18, 2020

LAST MODIFIED

Aug 18, 2020

PROTOCOL INTEGER ID

40606

Two days prior to experiment

1 Collect following supplies and ensure that isoflurane and buprenorphine are still in date):

- Betadine scrub, 32 oz (*RARC pharmacy; **Check Expiration Date**)
- Surgical drape, non-sterile, 38.5 in X 100 yd (VWR 34300-532)
- Procedure face masks, 2 Ply with Earloops, 50/bpx (*RARC pharmacy)
- Sterile gloves, 50/Box (*RARC pharmacy; **Check Expiration Date**)
- Sterile wooden applicators with cotton tips (Fisher 14-959-91)
- Isoflurane (FORANE), 250 mL (*RARC pharmacy; **Check Expiration Date**)
- Buprenorphine, 1 mL of a 0.3 mg/mL solution (Can be ordered through vet school, contact James Budde, jbudde@svm.vetmed.wisc.edu; **Check Expiration Date**)
- Ketoprofen, 100 mg/mL solution X 50 mL (*RARC pharmacy; **Check Expiration Date**)
- Sterile saline solution (for diluting buprenorphine and/or Ketoprofen to an appropriate working solution), 0.9%, 250 mL (*RARC pharmacy; **Check Expiration Date**)
- Sterile empty serum vial (for diluted buprenorphine and/or ketoprofen), 10 mL X 25/box (*RARC pharmacy)
- 1 mL syringe, 100/box (*RARC pharmacy)
- 26 G X 1/2 in length syringe needle, 100/Box (Fisher 14-826-15)
- Suture, Vicryl 4-0, absorbable polyglactin 910,6/pk or 36/box (*RARC Pharmacy; **Check Expiration Date**)
- Artificial tears, 1/8 oz tube (*RARC pharmacy)
- Mouse clippers (WAHL brand, Walmart)
- 2-count Hemostat, straight, serrated tip (Fisher 08-907)
- Blunt-pointed dissecting forceps, 5 inch (Fisher 08-890)
- Sharp pointed dissection scissors, 4.5 inch (Fisher 08-940)
- Gauze, 2 X 2 in, 12-ply, 200/bag (*RARC pharmacy)

2. One day prior to experiment

2 Autoclave surgical equipment: Place a 4.5 inch scissors, two hemostats, and one dissecting scissors into an autoclave bag. Seal bag and complete autoclave cycle.

3 To prepare 0.02 mg/mL Buprenorphine working solution from a 0.3 mg/mL stock solution, into a sterile septated bottle,

add 0.33 mL of Buprenorphine stock (0.3 mg/mL) to 4.67 mL saline. Remember to mark down the use of Buprenorphine stock in the controlled substances log. The expiration date should be listed on the bottle, and is the earliest date of either the buprenorphine expiration date or the saline expiration date.

4 Review Vezina Lab Animal Care and use protocol sections that pertain to castration and analgesia.

5 Send the following email message to svmvets@rarc.wisc.edu: To whom it may concern: we will be performing kidney capsule implants on a batch of [insert number] mice on [state date of surgery] in accordance with our approved animal protocol. The mice will be housed in AHABs room B23A/B and their cage cards will be marked with a label stating the date of the surgery. We anticipate some bleeding at the surgical site that should clear within a few days of the surgery."

Day of experiment

6 Clear items from surgery area.

7 Treat bench surface with 10% bleach or sporicidin for 30 min.

8 Mop bleach from bench surface and spray with 100% ethanol and air dry.

9 Bring animals upstairs.

10 15 minutes prior to surgery, inject animals with 0.1 mg/kg buprenorphine (volume = 5 microliters of 0.02 mg/mL buprenorphine per gram of mouse).

11 Lay down sterile drape.

12 Enter surgery start time and other information into surgical log.

13 Scrub hands and gown up.

14 Arrange tools in surgical space.

15 Set isoflurane vaporizer and turn it on (set mixer to 2% and flow rate to 1 liter/minute).

16 Induce mouse anesthesia and pinch toe to confirm unresponsiveness.

17 Add artificial tears to mouse.

18 Use betaine scrub to prep back area.

19 Use scissors to make a midline skin incision over the vertebral column, just caudal to the hindlegs.

20 Use scissors to cut through body wall above the right kidney.

21 Use forceps to exteriorize kidney.

22 Gently lift kidney capsule with forceps until a small tear forms.

23 While holding the kidney capsule with one forceps, use another forceps to transfer implant tissue under the kidney capsule.

24 A maximum of three implant tissues can be added per kidney. (maximize distance between implants by adding one to the cranial kidney surface, one to the medial, one to the caudal kidney surface).

25 Record the animal ID, the position and identity of each implanted tissue into the surgical log

26 Interiorize the kidney

27 Close the body wall incision with a single suture.

28 Repeat this procedure for left kidney

29 Close the midline skin incision with a single suture.

30 Remove mouse from vaporizer and return to cage. House at a density of one mouse per cage until wound is healed. Watch closely until coordination returns.

31 Enter surgery end time into surgical log.

Daily for 1 week after surgery

32 Inspect mice and record date, time, and inspector into surgical log.

33 Grounds for early euthanasia: mice will be euthanized if the body score becomes less than 2, if animal movement is compromised, or if animal becomes unresponsive to mild stimulus. If mouse has these symptoms, (1) euthanize mouse, (2) record the euthanasia in the surgical log, (3) alert RARC staff that an adverse event occurred (email svmvets@rarc.wisc.edu and explain what happened).

34 If an infection develops (1) email svmvets@rarc.wisc.eduv and request antibiotics (2) record the event in the surgical log.