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Subdiaphragmatic Vagotomy

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Anesthesia Induction and Surgical Preparation of the Mouse

1 Place the mouse into the induction chamber chamber and slide the lid all the way closed. Turn the appropriate stopcock valves to deliver the appropriate amount of isoflurane and O2.

2 As the mouse transitions from a state of awakeness to anesthetized, you should observe a slower respiratory rate and rhythm, alongside a loss of balance and slowed movement. After around 3 minutes, the mouse will become recumbent and non-responsive, at which point you may remove the mouse from the induction chamber.

3 Place the mouse in dorsal recumbency on the shaving station pad. Place the nose cone over the mouse's nose, and redirect the flow of Isoflurane and O2 by adjusting the stopcock valves. Access to the induction chamber should be closed and access to the nose cone opened. Note: Mice are obligate nasal breathers and cannot breathe through the mouth. Therefore, it is not essential to include the mouth in the nose cone. If necessary, you may secure the position of the nose by creating a "strap" out of suture that runs just under the mouse's front central incisors and ties to the nose cone.

4 Apply ample amounts of sterile eye lubricant to each of the mouse's eyes. Take care not to wipe off all the lubricant upon inserting the mouse's head back into the nose cone.

5 Using a charged shaver, remove the hair on the mouse's underside starting between the hypogastrium and umbilical region, and shaving rostrally to the xiphoid process. Brush off any excess hair with a q-tip.

6 Keeping the mouse's head inside the nose cone, transfer the mouse to the surgical area beneath the scope. Roll pieces of tape to make small "cuffs" and insert the mouse's forelimbs into the tunnel, sticking the tape to the absorbent pad underneath so that the mouse's arms rest out to the side, roughly perpendicular to the body. Secure the mouse's tail with an additional piece of tape.

7 Stop and monitor the mouse for depth of anesthesia. Respirations should remain rhythmic and slowed, even when noxious stimulus is applied (toe-pinch). If a fast respiratory rate, spontaneous movement or movement in response to toe pinch is observed, the depth of anesthesia is too lite and the percent of isofluorane should be increased. If the respiratory rate becomes markedly slower or the animal begins to gasp, the depth of anesthesia is too deep and the percent of isoflurane should be decreased.

8 When the depth of anesthesia is deemed appropriate, clean the incision site first with alcohol and then chlorhexidine. Always prep from "clean to dirty" areas, starting in the center of the animal and wiping in a spiral to the outside. Be careful not to accidentally transfer microorganisms from the periphery back to your target incision site. Never "double dip" a contaminated cloth wipe back into the antiseptic solution and do not "back track" over an area that has already been prepped. You should always allow the prepping solution to dry completely before draping.

9 Form a drape using a pre-cut, autoclaved paper towel and secure it over the animal in a way where you can still monitor the animal's breathing.

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Sub-diaphragmatic Vagotomy and Closure

10 Using a pair of forceps, grasp the mouse's skin along the midline directly above the umbilical region. Using a sharp pair of scissors, make a midline incision from the umbilicus to approximately 0.5cm below the xiphoid process. The first incision should be no more than ~1.5cm in length for males or ~1.25cm for females, and should pass through the skin, subcutaneous tissue and fascia. Further separate the skin from the abdominal cavity lining by placing the closed tip of the scissors between the layers and gently opening up the blades.Be sure to angle the scissors up and away from the body cavity.

11 Make a second incision, also from the umbilicus to approximately 0.5cm below the xiphoid process, passing through the linea alba, transversalis fascia and peritoneum lining. Angle the scissors up and away from the viscera as you do this. (Note: The branches of the superior epigastric artery supply blood to the rectus abdominus muscles and may be cut, producing a little blood. This is okay.)

12 Insert the head of a Q-tip into the abdominal cavity and gently push the viscera (mainly the intestines) down into the hypogastric region. Lift the left and median lobes of the liver to expose the edge of the stomach. When you visualize the insertion of the esophagus at the stomach, gently hook the esophagus and guide (without pulling too much) the stomach medially until the esophagus is more aligned with the incision and sitting in your line of view.

13 Follow the esophagus up to the esophageal hiatus, where the esophagus passes through the diaphragm. Using sharpened 45 degree angled forceps, carefully grab the vagus nerve as close as you can to the diaphragm. You may need to pull away a thin fascial sheath before you can grab the actual nerve. (Note: the descending vagus nerve leaves the vagus nerve plexus on the thoracic esophagus, down through the esophageal hiatus passing through the diaphragm, and wraps counterclockwise around the abdominal esophagus. Though the vagus nerve will appear large compared to the blood vessels enervating the esophagus, you may just barely visualize it from the ventral position. If you have difficulty finding the vagus nerve, you may gently roll the esophagus over by rotating the hook, and therefore the esophagus, clockwise.)

14 Steadily "peel" the vagus nerve off the esophagus and use a pair of micro scissors to cut the vagal insertion at the base of the esophagus.

15 Once you are confident the vagus nerve has been resected, restore the stomach, liver lobes, and intestines to their original position.

16 Briefly examine the mouse's internal organ orientation and check that all organs are intact and in their correct place.

17 Begin closure. If your incision is 1cm or less, you may use one cruciate suture if the closure is deemed secure and appropriately placed. If the incision is greater than 1cm, perform a line of simple interrupted sutures spaced approximately 2-3mm apart.

18 Once the linea alba is securely closed, use the scissors to free the skin from any remaining fascia. You will want the skin quite loose for staple closure. Taking forceps in both hands, grab both seams of skin and bring them together, raising the skin upward like a tent. Have an assistant wield the stapler and place a staple between where your forceps are holding the skin. There should be a space about half a staple wide between each staple. You may need to use a forceps or needle driver to tighten the staples. If there is skin left over after the last staple, consider passing a line of suture over the pucker to keep the skin sealed together.

Anesthesia Recovery

19 Prior to placing the animal in the recovery cage, check the temperature of the heating pad underneath. The animal should be provided a means to move away from the heat source once awake, and this can be accomplished by supplying heat to one half of the cage only. Use a clean paper towel for a small barrier of material between the animal and the heated part of the cage. Bedding should never be used.

20 Place the animal in the recovery cage either in ventral or lateral recumbency. Monitor how long it takes for the mouse to awaken. While the mouse is recovering from anesthesia, someone must stayin the procedure room to monitor the mouse until it has completely recovered.

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21 Once the animal is able to walk around the cage freely and appears to have recovered and an appropriate level of alertness and responsivity, it may be returned to its home cage.

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