



Bile Diversion Procedure in mice

Version: 1

Replaced by version: No previous versions

Edited by: Vance L. Albaugh, MD, PhD

Summary:

This is the protocol for conducting the bile diversion procedure in the mouse. In this procedure the common bile duct is ligated proximal to the confluence of the common bile and the pancreatic ducts and an anastomosis is created at a prespecified point along the gastrointestinal tract to re-establish drainage of the biliary tree.

Reagents and Materials:

Reagent/Material	Vendor	Stock Number
6-0 polyglactin suture	Arosurgical	XS-G618R11-U
4-0 nylon suture	Arosurgical	CC13A06N-45
9-0 nylon suture	Arosurgical	TK-091038
Cotton swabs	Fisher Scientific	19-072333

The remaining materials that are necessary for any surgical procedure in mice are sterile, high quality surgical instruments for microsurgery. A dissecting microscope (at least 10x magnification) is also highly recommended, however, surgical loupes or other magnification aid could be substituted. The individual performing the operations should be well-trained in small animal surgical and mouse anatomy.

Protocol:

Preoperative Care

1. All animals must be singly-housed.
2. Preoperative pain medications should be administered:
 - a. Ketoprofen (5 mg/kg)
 - b. Buprenorphine (0.05 mg/kg)
 - c. LRS are given at the end of surgery and a second dose is given 24 hours later.
3. Ensure adequacy of anesthesia.
4. Place mouse on surgical board/surgical field and immobilize gently.
5. Prep and drape animal sterilely.

Operation

1. Begin the procedure by making a midline laparotomy incision with sharp surgical scissors. Be sure to stay on the linea alba and away from the rectus abdominus muscles.
2. Using cotton swabs, gently sweep the intestinal contents away from the gallbladder fossa, exposing the common bile duct and pancreas/pancreatic ducts.
3. Using a 9-0 nylon suture, ligate the common bile duct proximal to the pancreatic duct. Do not intentionally ligate any of the blood supply to the gallbladder or surrounding structures.

IMPORTANT: When ligating the common bile duct it is critical to leave the pancreatic duct uninterrupted. If the pancreatic duct is ligated inadvertently or damage (possibly creating a stricture), then this will impede the flow of pancreatic fluids and cause a state of chronic pancreatitis in the animal.

4. Once the common bile duct is ligated, gently replace the intestines and then identify the intestinal segment to which the anastomosis between the gallbladder and the intestine will be created.
5. On the antimesenteric surface of the bowel to where the anastomosis will be made, make a 1-2mm enterotomy.
6. Next, on the gallbladder (which should be filling with bile secondary to ligation of the common bile duct) make a ~2mm cut with surgical microscissors transversely on the gallbladder.

IMPORTANT: When cutting the gallbladder do not intentionally cut through any of the obvious arterial supply to the gallbladder proper. The incision should be transverse and be below the fundus of the gallbladder.

7. Juxtapose the gallbladder and the bowel to be connected. In order to improve visualization, it may sometimes help to use sterile pieces of cotton to move the gallbladder and liver more caudally to bring it into the surgical field. If visualization is adequate though, then this is unnecessary.
8. Using the 9-0 nylon suture and microsurgical needle holders, create the anastomosis moving from the left to right side of the mouse. All knots should be on the outside of the anastomosis. The anastomosis should be done in a continuous-running fashion, with the back row portion of the anastomosis completed prior to the front facing portion.
9. At the conclusion of the procedure, the gallbladder may be somewhat distended secondary to bile (as there will be some tissue swelling at the site of the anastomosis). If this occurs, it means that the anastomosis is likely sealed and does not leak. Alternatively, one may need to test for leaks by gently pressing on the infundibulum of the gallbladder in order to distend the fundus.
10. If leaks are detected, they can be sealed/fixated using a simple, interrupted 9-0 nylon suture. However, given the extreme delicacy of the gallbladder tissues, extreme care should be taken in order to avoid tearing any gallbladder tissue.
11. After the anastomosis between the intestine and gallbladder is completed, gently irrigate the abdominal cavity with warmed, sterile saline.
12. Next, close the abdomen in a simple, two-layer, interrupted fashion. The first interrupted layer should be a simple, interrupted muscle layer with 6-0 polyglactin suture. Using good technique suture the connective tissue and not the muscle proper. The second layer is a skin layer also done in a simple, interrupted fashion with 4-0 nylon suture.

Postoperative Care

1. All mice receive 0.5ml of warmed, sterile saline following the procedure before being placed in the recovery cages.
2. All mouse cages are kept partially on a veterinary-approved heating pad for 5 to 7 days postoperatively.
3. Mice are monitored until recovered from the procedure, which typically takes 7-14 days depending on the procedure. General behavior (i.e. bright/alert/responsive vs. depressed/obtunded) is monitored.
4. Weight and food intake is measured and recorded weekly.
5. Pain medication is administered per protocol: buprenorphine 0.5 mg/kg q12h x 2 doses (including preoperative dose), ketoprofen 5 mg/kg once at 24 hours postoperative.
6. Additional pain medication may be needed depending on postoperative recovery.