

Common Surgical Procedures in Mice and Rats

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<https://research.utsa.edu/compliance/larc/training.html>

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Procedures in this Presentation

- Partial and total splenectomy (dorsal and ventral approach)
- Ovariectomy
- Ovariohysterectomy
- Orchietomy (scrotal and abdominal approach)
- Scrotal and abdominal vasectomy
- Adrenalectomy
- Nephrectomy
- Ureter ligation

2

Procedures in this Presentation

- Partial hepatectomy in mice (with and without gall bladder removal)
- Subdiaphragmatic vagotomy
- Pilonplasty secondary to subdiaphragmatic vagotomy
- Removal of sciatic nerve section
- Common carotid catheterization
- External jugular vein catheterization
- Femoral artery catheterization
- Femoral vein catheterization

3

Disclaimers

1. Images and videos associated with this presentation are for the purpose of demonstration of surgical techniques and are not intended to teach or demonstrate aseptic technique
2. As author of this presentation, I firmly advocate that survival surgery in rodents should be performed with meticulous attention to aseptic technique
3. Procedures shown in this presentation conducted as terminal procedures in anesthetized animals, under a protocol approved by the animal ethics committee at my institution

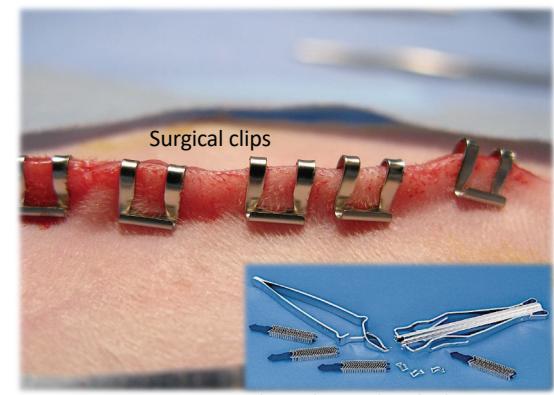
4

Before beginning...

Skin Closure

5

Surgical clips



Clips, clip applier & clip remover

6

Non absorbable, monofilament suture



Silk is not an appropriate suture for skin closure

7

Silk on Skin



- It is easy to handle making it the suture of choice of many investigators... however...
- Should not be used for skin closure for following reasons:
 - Produces undue local reaction and inflammatory response
 - It is braided and through its wicking action serves as a fomite to introduce microorganisms into the wound
 - These properties result in potential clinical and subclinical infections
 - As such, it is not consistent with sound principles of veterinary medicine

8

Approaching the Spleen

Dorsal & Ventral Approach

9

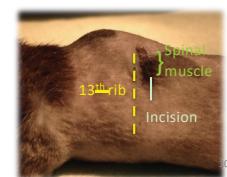
Dorsal Approach

~1 cm skin incision

On animal's left side

Parallel to 13th rib

Dorsal extreme
beginning just below
the spinal muscle



10

Separate (no need to cut)
abdominal muscle fibers with
tips of sharp scissors (iris
scissors)



Spleen is seen below opening



Exteriorize spleen



11

Ventral Approach

- Make a 1-2 cm mid ventral skin incision with its extreme cranial end at the level of the stomach
- Abdomen is entered through the linea alba
- Spleen is identified below and to the left of the linea alba
- Exteriorize spleen



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Partial Splenectomy (Biopsy)

Via a Ventral or Dorsal Approach

13

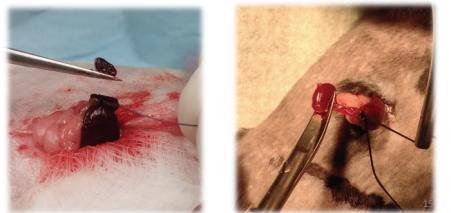
Spleen is accessed via a dorsal or ventral approach

14

1. Tie a ligature around the spleen arm to stem bleeding

Do not include splenic vessels into ligature

2. Excise splenic tissue distal to ligature



15

Partial Splenectomy (Biopsy) in the Mouse – Video

Ventral Approach
3:08

Video – Partial Splenectomy



17

Total Splenectomy

Via a Ventral or Dorsal Approach

18

Spleen is accessed via a dorsal or ventral approach

19

1. Exteriorize spleen
2. Cut gastro-splenic ligament with scissors or cautery to separate spleen from stomach
3. Identify, isolate and ligate splenic vessels.
In tiny mice careful cauterization of vessels without a ligature may be performed with caution



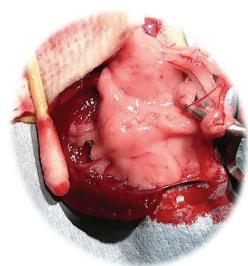
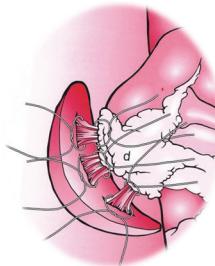
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Cauterize or transect blood vessels distal to ligature



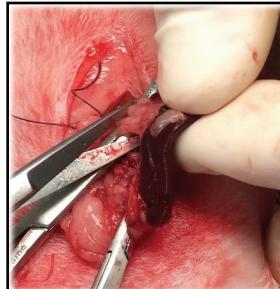
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In rats (especially large ones) isolate individual blood vessel bundles and ligate



22

Isolating blood vessels



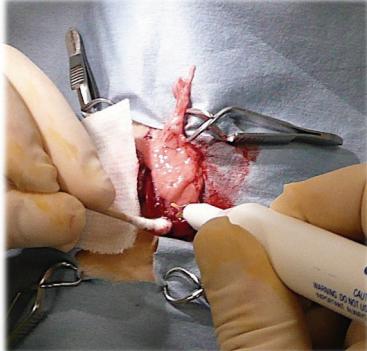
Ligate



& Cut

23

24



Cautery alone works well in smaller animals (mice)

25

- Muscle layer is closed with absorbable suture
- Skin is closed with
 - Monofilament, non-absorbable suture in an interrupted fashion, or
 - Surgical Clips

Surgical skin glue (cyanoacrylate) may be applied to close small skin incisions or to reinforce (and provide a microbial barrier) larger incisions

26

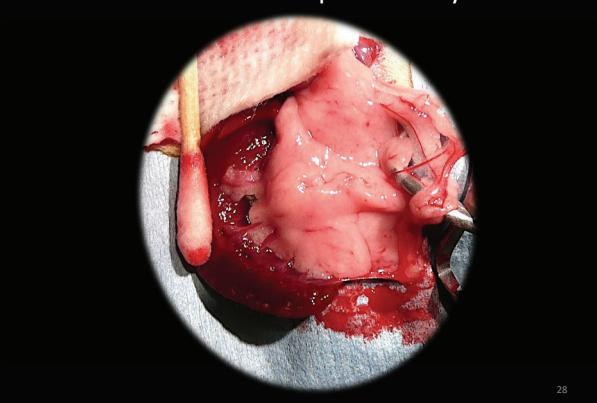
26

Total Splenectomy in the Mouse Video

Ventral Approach
3:28

27

Video – Total Splenectomy

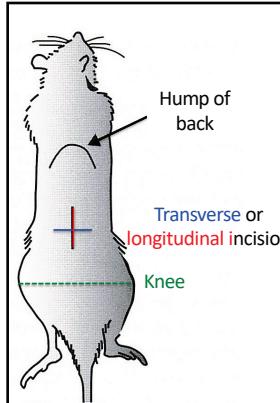


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28

Ovariectomy

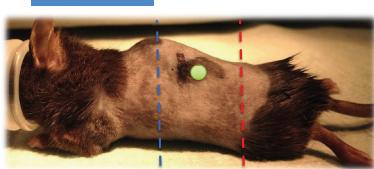
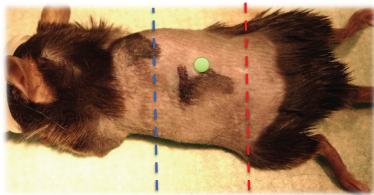
29



- Make a **transverse** or **longitudinal** dorsal incision ~ half distance between hump of back and level of **knee** with animal in ventral recumbency
- **Transverse** incision allows easier bilateral access to both ovaries through same incision

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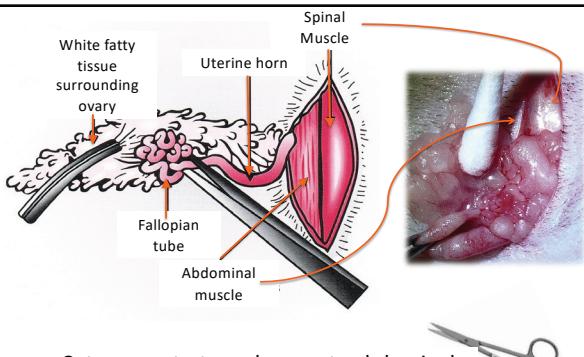


Hump of back
Not as obvious
in heavy animals

Ovary

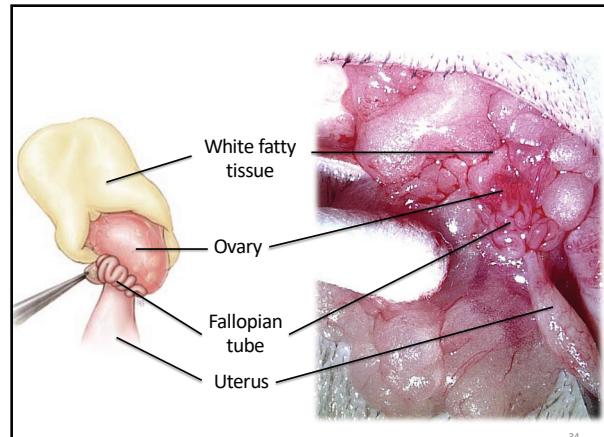
knee

32

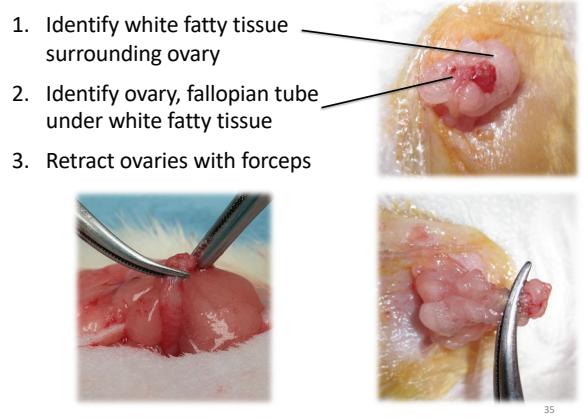


Cut or penetrate and separate abdominal wall muscle fibers with tips of sharp scissors (iris scissors) to locate ovary

33



34



35

Cut or cauterize to remove ovary
With rats (specially if large) may place a ligature to minimize bleeding



36

35

Alternative to Traditional Cautery

1. Submerge tips of hemostatic forceps in a hot bead sterilizer
 2. Remove and immediately clamp tissue



37

37

- Muscle layer is closed with absorbable suture
 - Skin is closed with
 - Monofilament, non-absorbable suture in an interrupted fashion, or
 - Surgical Clips

Surgical skin glue (cyanoacrylate) may be applied to close small skin incisions or to reinforce (and provide a microbial barrier) larger incisions

38

38

Mouse Ovariectomy Video

1:55

39

Video – Ovariectomy

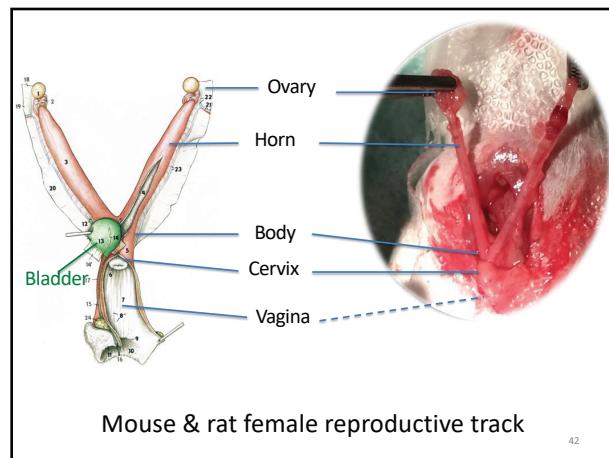


40

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Ovariectomy

41



Mouse & rat female reproductive track

42

42

Linea alba

1. Make a ventral midline skin incision with its extreme caudal end at level of the pubis (bladder)
2. To enter abdomen, linea alba is incised in same direction as skin incision



43

- Exteriorize uterus with its surrounding fat & tissues
 - If necessary move bladder to the side or empty it with a syringe & needle
- Identify uterine body, horns and ovaries

44



- Ovaries, uterine horns and uterine body are dissected free to separate from other tissues. In large rats, a ligature distal to ovary minimizes bleeding
- Cautery is useful to dissect tissues away from uterus

45

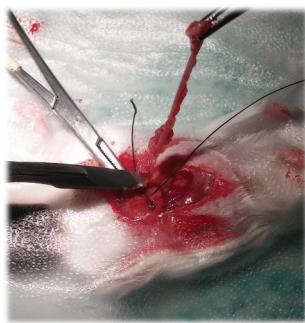


Clamp uterine body or
cervix with hemostatic
forceps



Place a ligature distal to
hemostatic forceps

46



Remove (excise) uterus along with ovaries proximal to
hemostats with cautery, surgical blade or scissors

47

- Muscle layer is closed with absorbable suture
- Skin is closed with
 - Monofilament, non-absorbable suture in an interrupted fashion, or
 - Surgical Clips

Surgical skin glue (cyanoacrylate) may be applied to
close small skin incisions or to reinforce (and
provide a microbial barrier) larger incisions

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Mouse Ovariectomy
Video

1:58

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Video – Ovariectomy



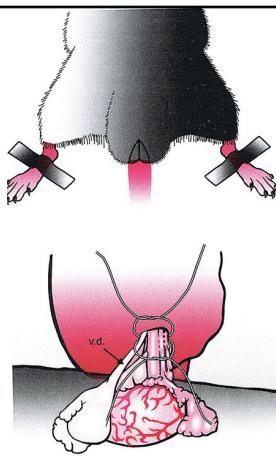
50

Scrotal Orchietomy

51

- Rats & mice have open (loose) inguinal canals. This allows free movement of testes between scrotum and abdomen
- During scrotal castration, testes may need to be forced into scrotum. This may be done by exerting pressure on testes towards the scrotum in the caudal abdomen with fingers or Q-tips

52



53



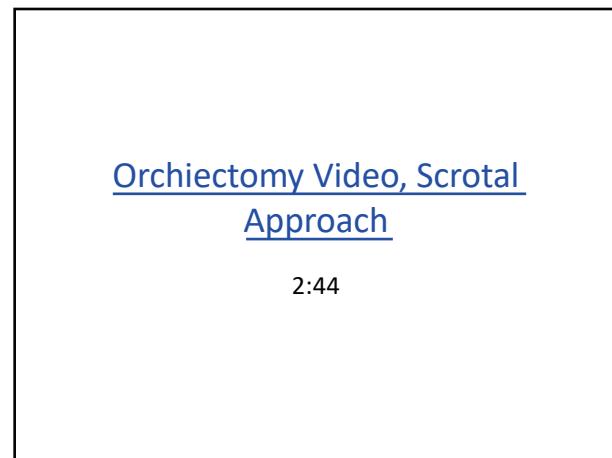
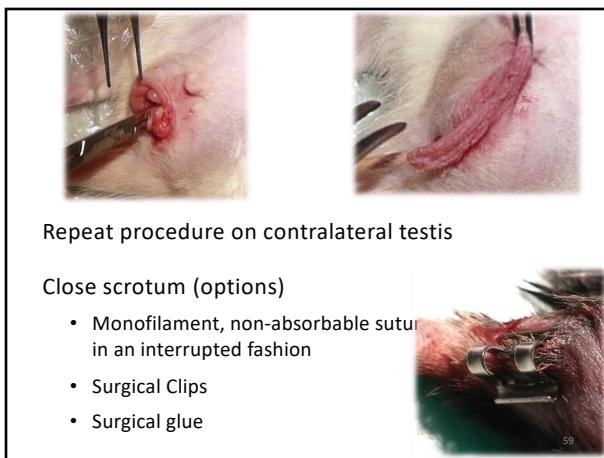
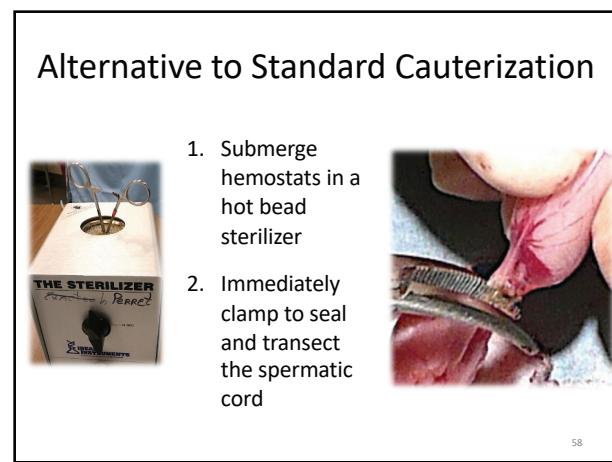
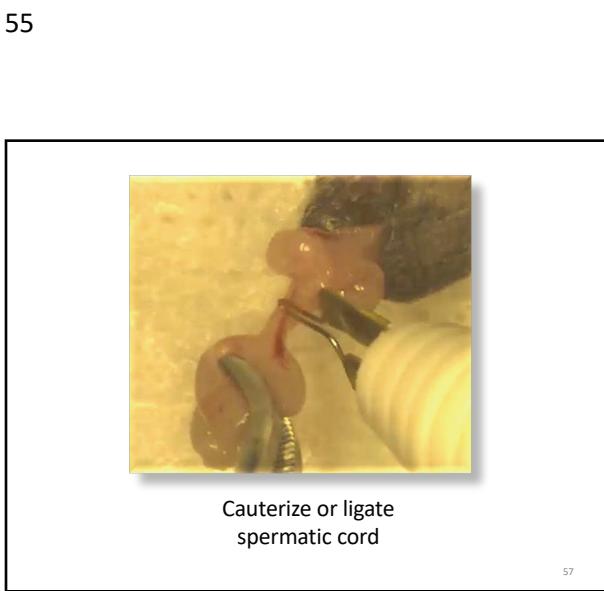
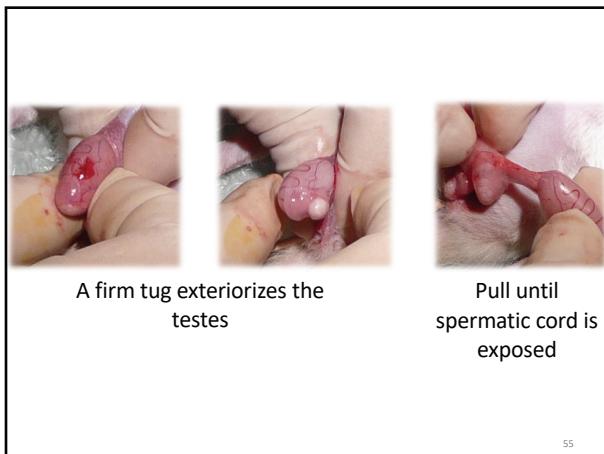
Incise scrotum
on midline

Incise parietal tunica,
avoiding cutting the vaginal tunica, which is
intimately associated with &
adhered to the testes

In mice, use scissors

54

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Video – Scrotal Orchiectomy



61

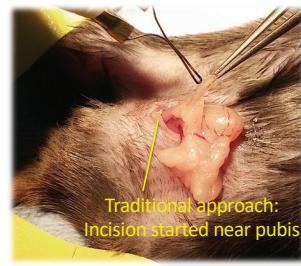
Abdominal Orchiectomy

62



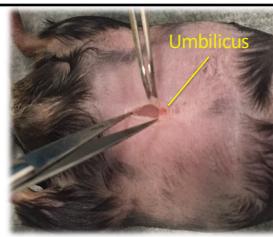
- Rats and mice have open inguinal canals, allowing free movement of testes between scrotum and abdomen
- Testis may need to be pushed from scrotum to lower abdomen towards. This may be done by exerting pressure on scrotum towards abdomen with fingers or Q-tips

63



- The traditional abdominal castration makes a ventral midline incision close to the pubis, which can damage seminal vesicles while testis are being searched
- Suggested is a refinement to this technique in next slide

64



- A refined approach consists of making a ventral midline skin and muscle incision, starting at umbilicus & continuing caudally for a few millimeters
- Testis are pulled out through the incision and removal is performed as described under the scrotal orchiectomy

65



Exteriorize
whitish fat
that is related
to the testis



Pull this fat
along with
testis

66



Clamp below the testis ensuring no testicular tissue is trapped in the clamp



Cauterize between the testis & clamp

67

- Replace excess tissue back into abdomen
- Muscle layer is closed with absorbable suture
- Skin is closed with
 - Monofilament, non-absorbable suture in an interrupted fashion, or
 - Surgical Clips
 - Surgical skin glue (cyanoacrylate), which provides a microbial barrier

68

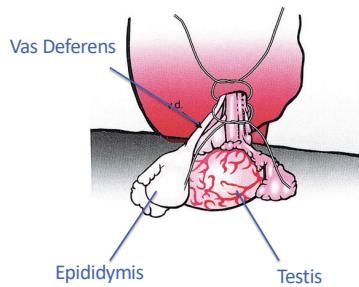
Vasectomy – Vas Deferens

Abdominal or Scrotal

69

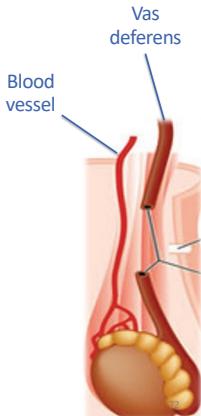
Testis is exteriorized through the incision and closure as described under scrotal and abdominal orchietomy

70

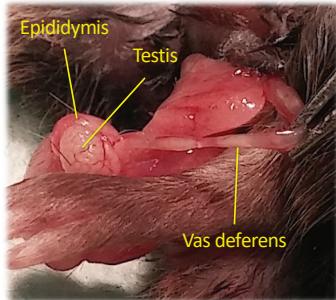
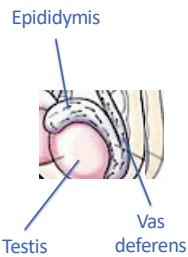


71

- Vas deferens is relatively massive and easily visualized as a white/bright tubular structure with a blood vessel running alongside it
- Vas is also recognized as continuation of the epididymis
- Dissect out/isolate the vas deferens



72



Vas deferens is relatively massive and easily visualized as a white/bright tubular structure associated with the epididymis

73

Excise ~0.5 cm section of each vas deferens with a blade, scissors or cautery with or without suture ligation



74

Vasectomy Video in the Mouse

2:15

75

Video – Vasectomía del Ratón



76

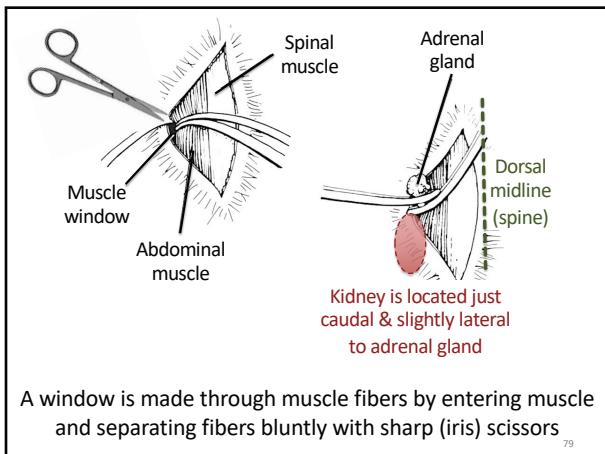
Adrenalectomy

77

Make a transverse incision on the dorsal skin or 2 incisions on each side of the spine immediately lateral to the spinal muscles. The incision(s) is(are) approximately 4 mm caudal to the highest point of the hump



78

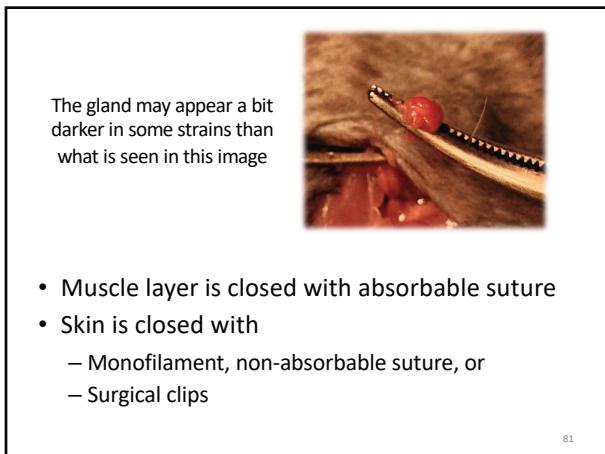


79

- Adrenal gland is located cranial and medial to kidney, embedded in fatty tissue
- Be careful not to penetrate the diaphragm at this point, which will result in immediate death
- Remove the gland intact with forceps without needing to cut



80



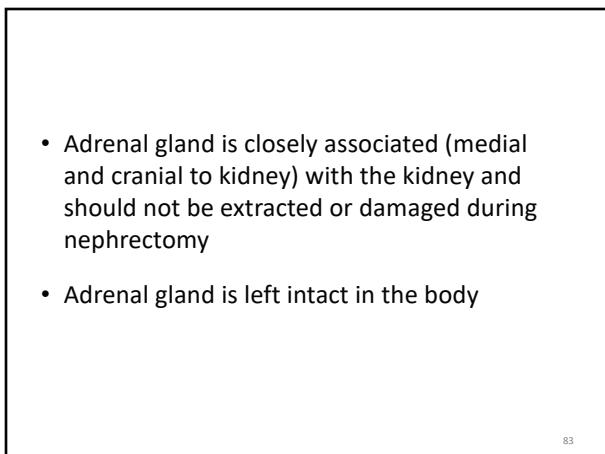
81

- Muscle layer is closed with absorbable suture
- Skin is closed with
 - Monofilament, non-absorbable suture, or
 - Surgical clips

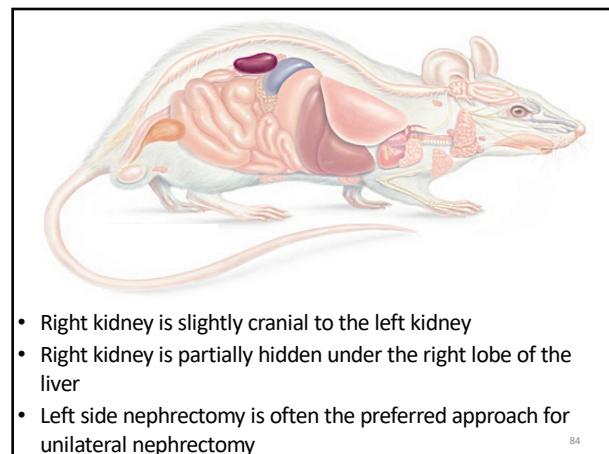
81

Nephrectomy

82

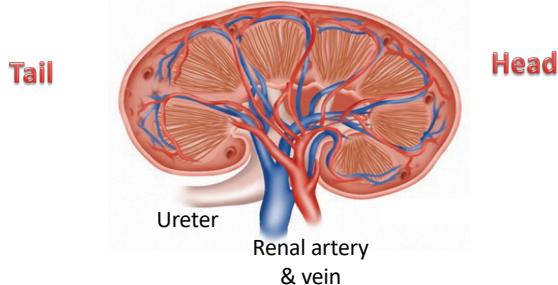


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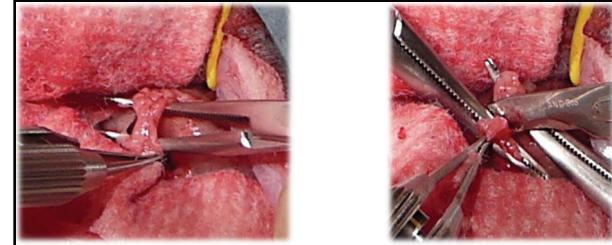


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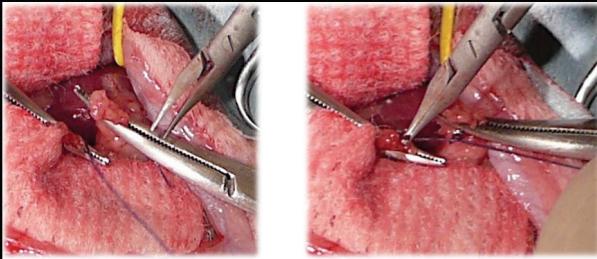


85



Renal vessels and ureter are isolated

86



Ligate with 1 or 2 sutures and remove kidney

Second ligature more important the larger the animal is

87

- Muscle layer is closed with absorbable suture
- Skin is closed with
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 - Surgical Clips

Surgical skin glue (cyanoacrylate) may be applied to close small skin incisions or to reinforce (and provide a microbial barrier) larger incisions

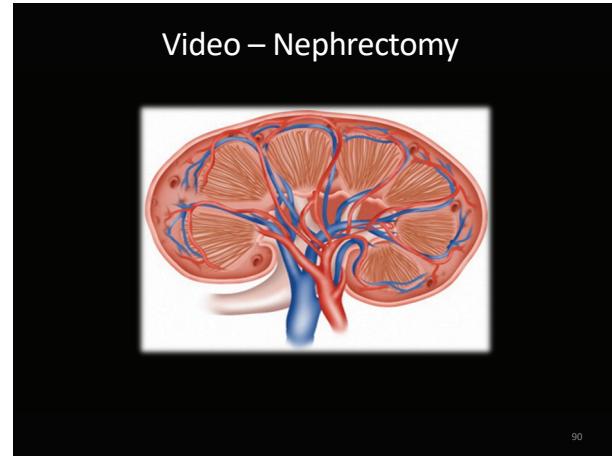
88

88

Nephrectomy Video

2:29

89



90

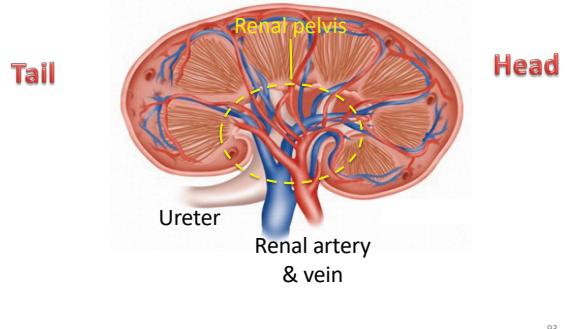
Ureter Ligation

To simulate unilateral obstructive nephropathy

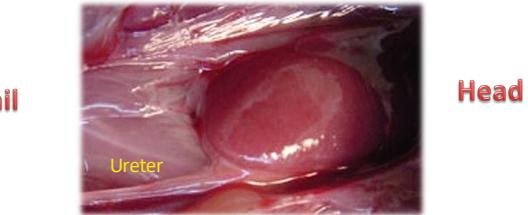
91

The kidney is exteriorized as described under nephrectomy session

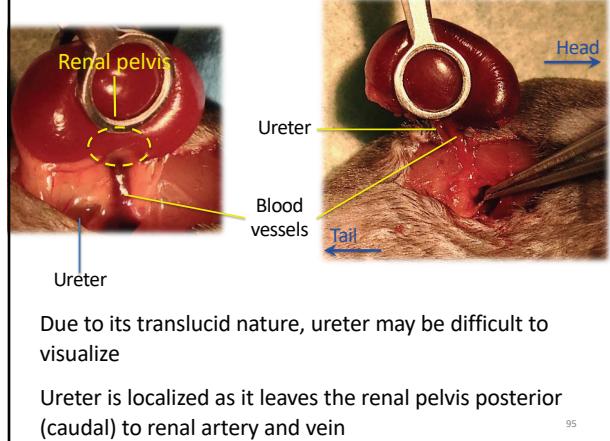
92



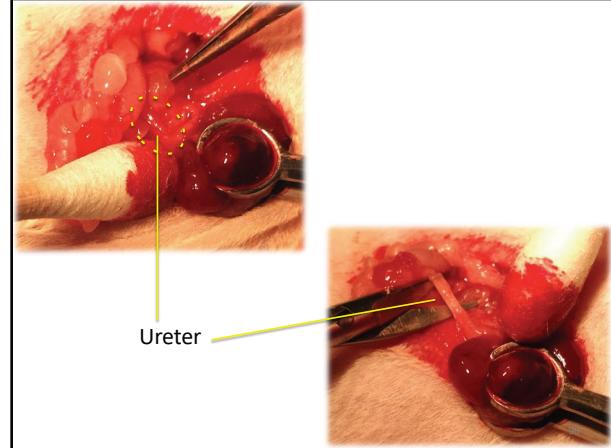
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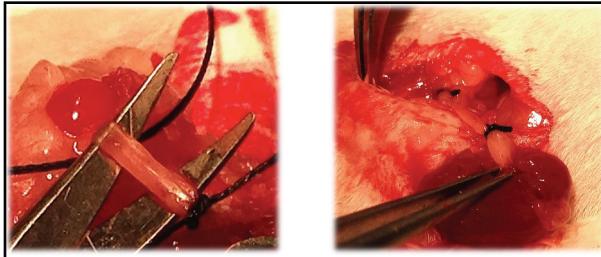
94



95



96



Ureter is ligated with 1 or 2 ligatures or with vessel clamps (for temporary occlusion) to simulate obstructive nephropathy

97

- Muscle layer is closed with absorbable suture
- Skin is closed with
 - Monofilament, non-absorbable suture in an interrupted fashion, or
 - Surgical Clips

Surgical skin glue (cyanoacrylate) may be applied to close small skin incisions or to reinforce (and provide a microbial barrier) larger incisions

98

98

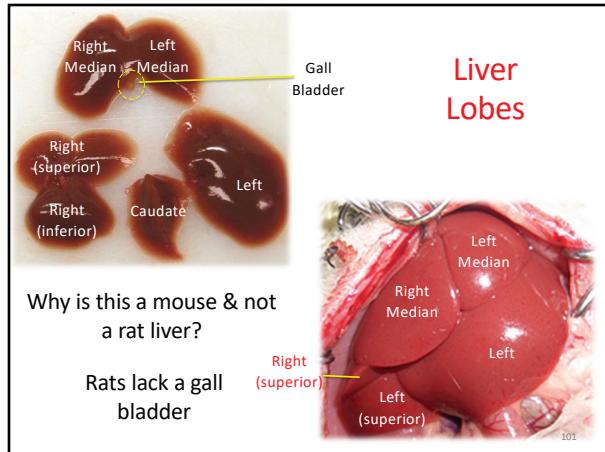
$\frac{2}{3}$ Partial Hepatectomy in Mice

Two Methods

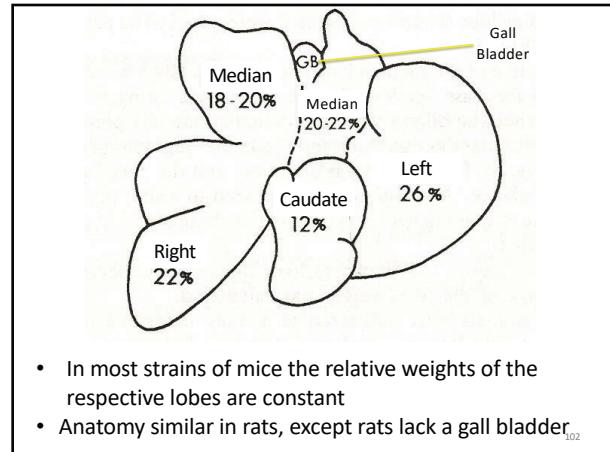
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Liver Anatomy

100



101



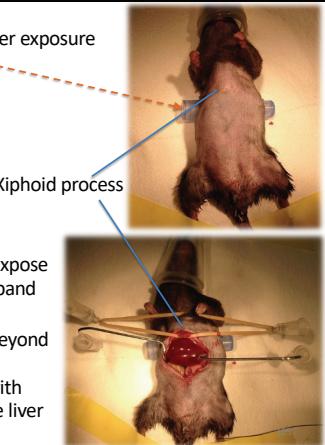
102

- Mammals can survive removal of up to 75% of total liver mass
 - If > 75% is removed, remaining liver mass is not sufficient to maintain critical levels of blood glucose
 - Removal of <1/3 will not elicit a generalized liver proliferative response
 - Therefore, median and left lobe hepatectomy is considered ideal and results in removal of ~ 64-68% of total liver mass, a partial hepatectomy technique known as **2/3 Hepatectomy**
 - After resection of 2/3 of the liver, remaining hepatocytes undergo one or two replication rounds without complications related to hypoglycemia
 - **2/3 hepatectomy** is therefore a preferred method for studying dynamics of liver regeneration

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Surgical Technique

104

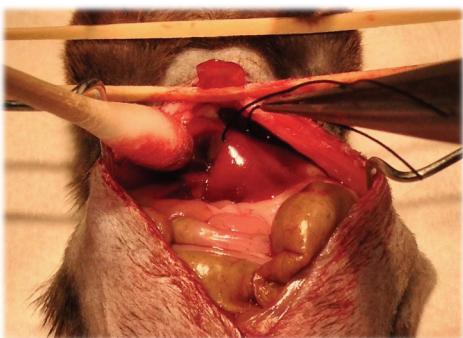


105



Alternative Xyphoid Lifting

106



Q-tips soaked in saline are useful tools for moving lobes

107



Before placing ligatures at the base of the lobes, cut various ligaments as illustrated here in the middle lobe

108

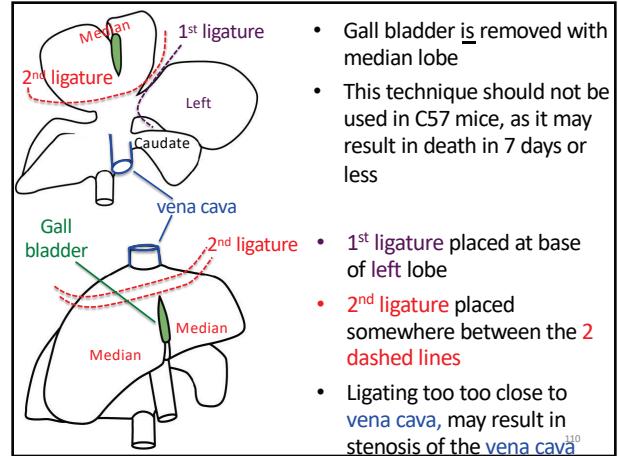
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108

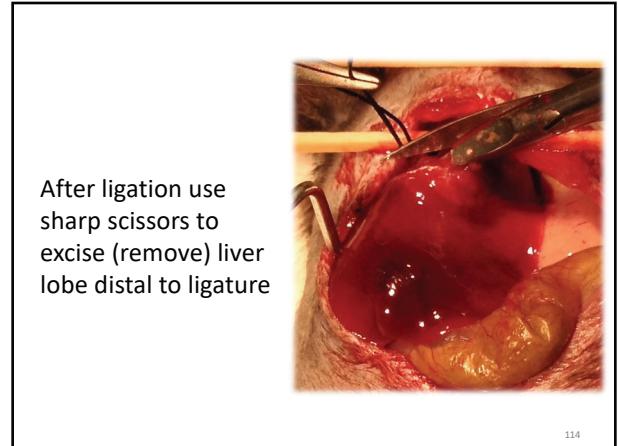
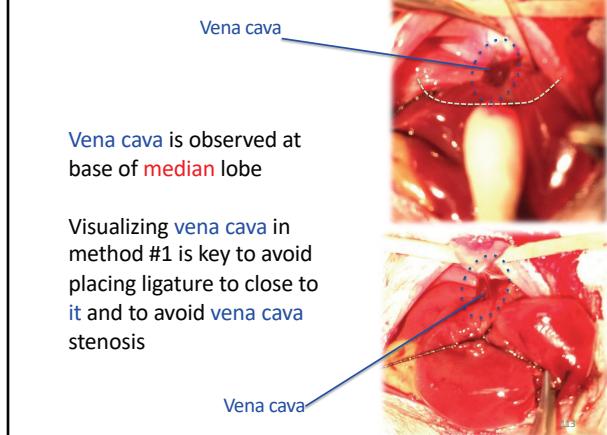
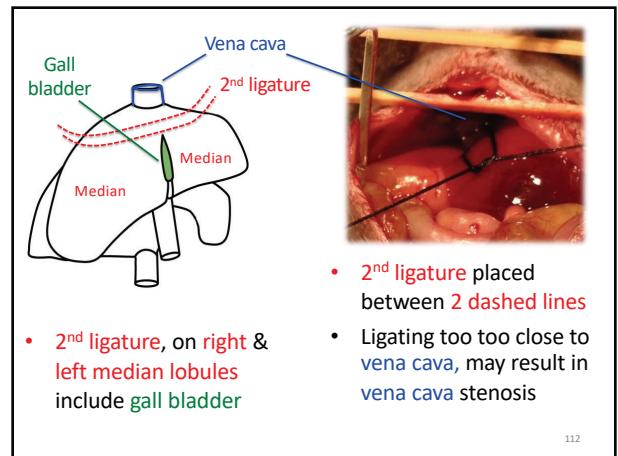
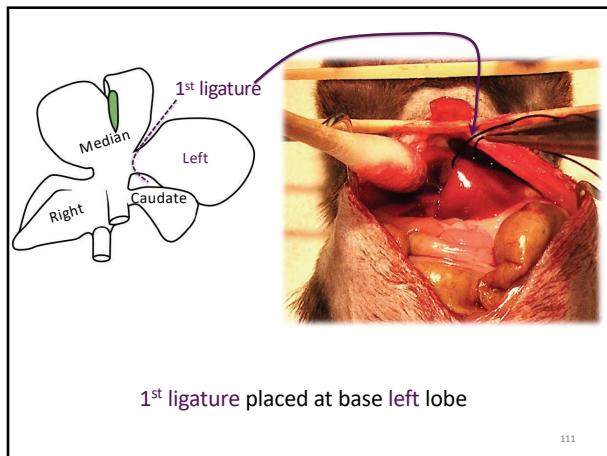
Partial Hepatectomy – Method #1

**Two ligating sutures
Gall Bladder Removed**

109



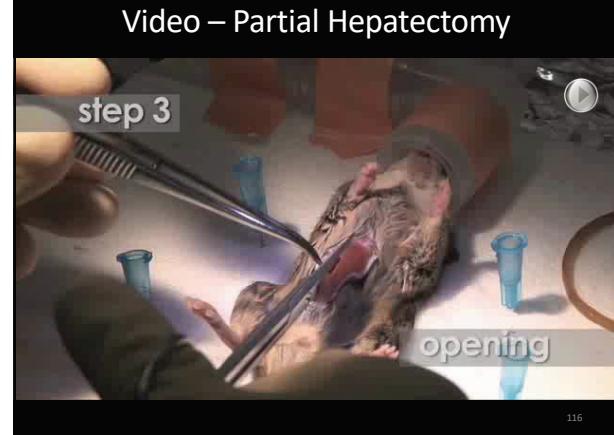
110



Partial Hepatectomy Video
Method #1, Gall Bladder Removed

3:48

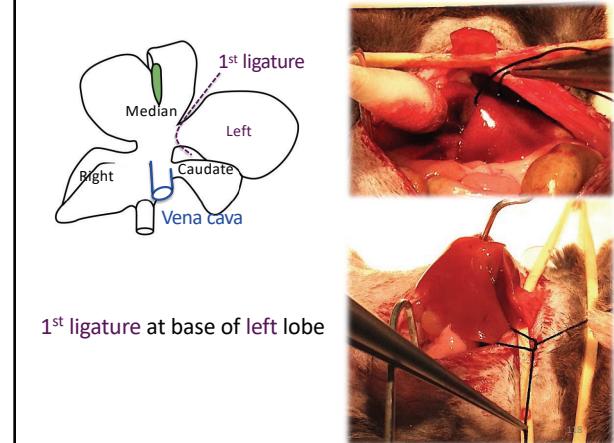
115



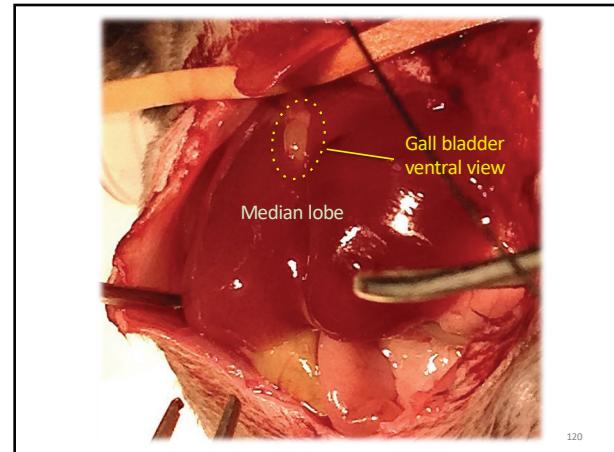
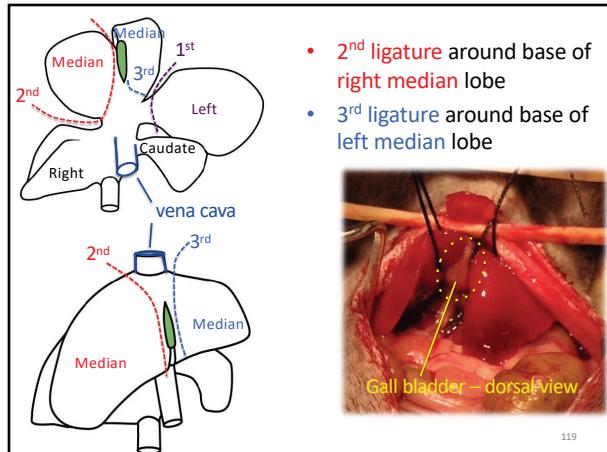
Partial Hepatectomy – Method #2

Three ligating sutures
Gall Bladder Left in Place

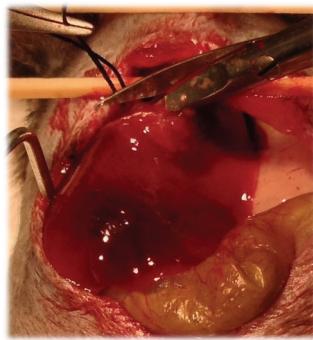
117



118



After ligation, distal to ligatures, lobes are excised with sharp scissors



121

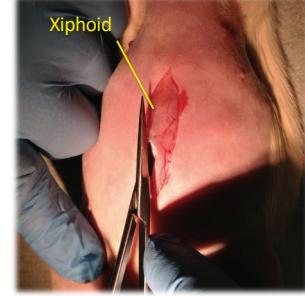
- Muscle layer (linea alba) is closed with absorbable suture
 - Skin is closed with
 - Monofilament, non-absorbable suture in an interrupted fashion, or
 - Surgical Clips
- Surgical skin glue (cyanoacrylate) may be applied to reinforce closure (and provide a microbial barrier)



122

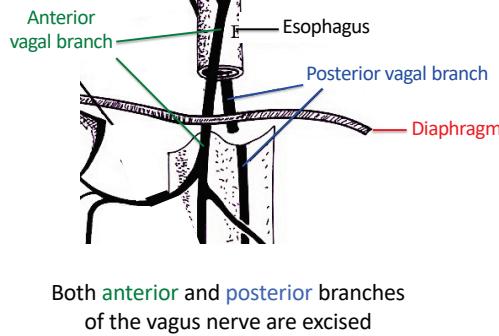
Subdiaphragmatic Vagotomy

122

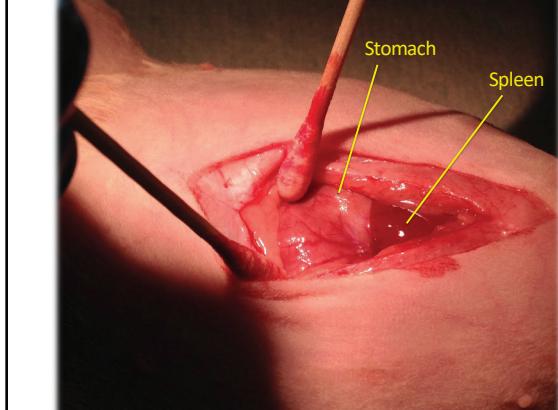


124

- A mid-ventral skin incision is made with its extreme cranial end at the level of xiphoid process
- Abdomen is entered through linea alba in same direction as skin incision



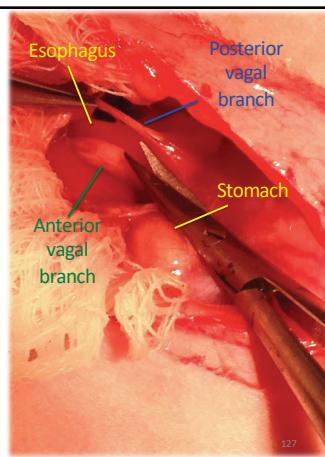
125



126

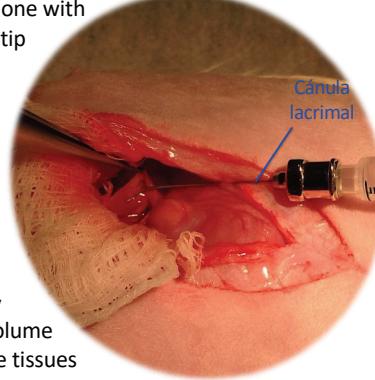
125

- Esophagus & both branches of vagus (**anterior & posterior**) are identified caudal to diaphragm
- With delicate blunt dissection separate both vagal branches away from esophagus



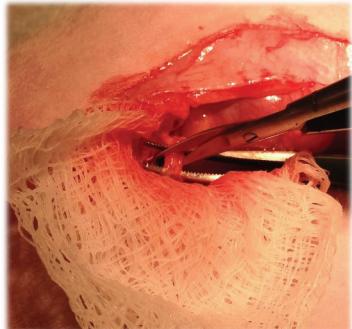
127

- Dissection/separation of vagus branches from esophagus can be done with micro-scissors, fine tip forceps or by hydrodissection
- Hydrodissection is done by inserting a lacrimal cannula attached to a 1 ml syringe between esophagus & vagus branch, followed by injection of small volume of saline to separate tissues



128

Each vagal branch is individually cut or ligated depending on study objectives
Vagotomy in mice requires high magnification



129

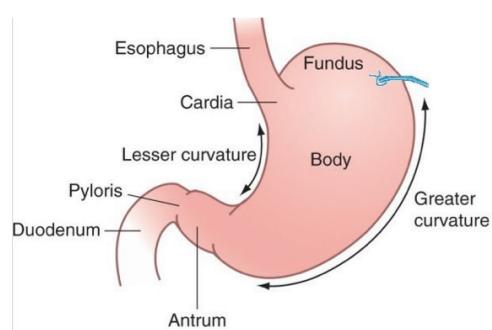
Pyloroplasty as Adjunct Surgery to Subdiaphragmatic Vagotomy

130

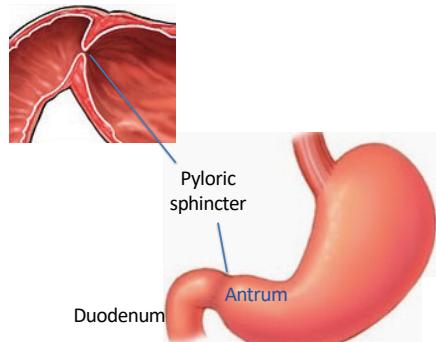
The Problem and the Solution

- The pyloric sphincter controls output of food from the stomach into the duodenum
- Problem: Vagotomy results in pylorus inability to allow stomach emptying into duodenum
- Solution: Pyloroplasty widens the pylorus to allow passage of food into the duodenum

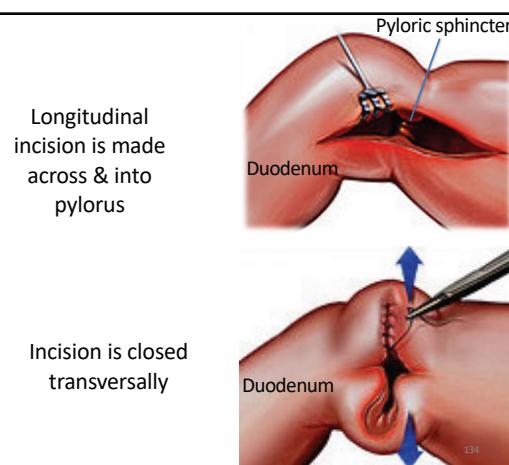
131



132



133



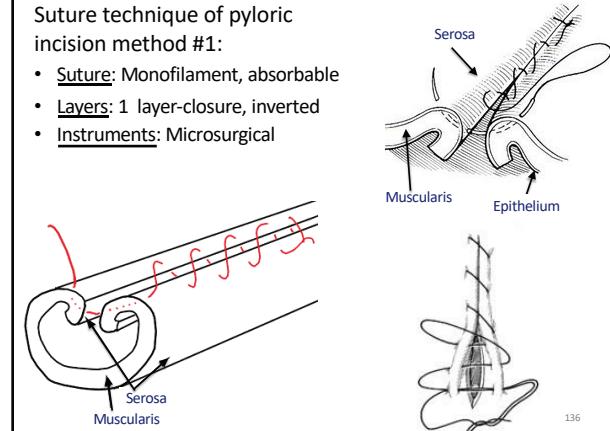
134



135

Suture technique of pyloric incision method #1:

- Suture: Monofilament, absorbable
- Layers: 1 layer-closure, inverted
- Instruments: Microsurgical



136

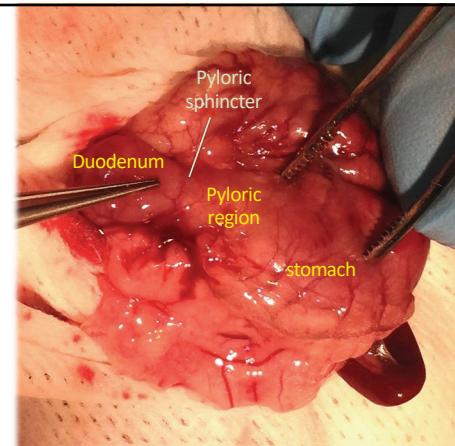
Suture technique of pyloric incision method #2 (preferred):

- Suture: Monofilament, absorbable
- Layers: 1 layer-closure, inverted
- Instruments: Microsurgical

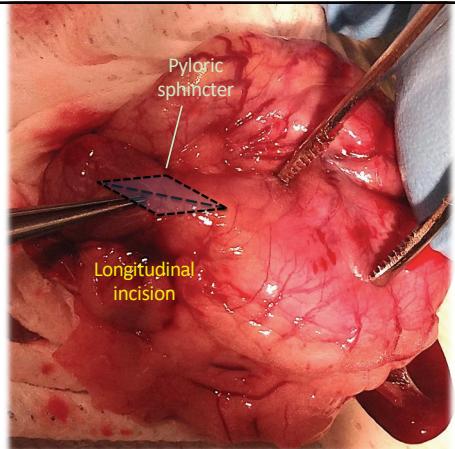
Horizontal mattress pattern



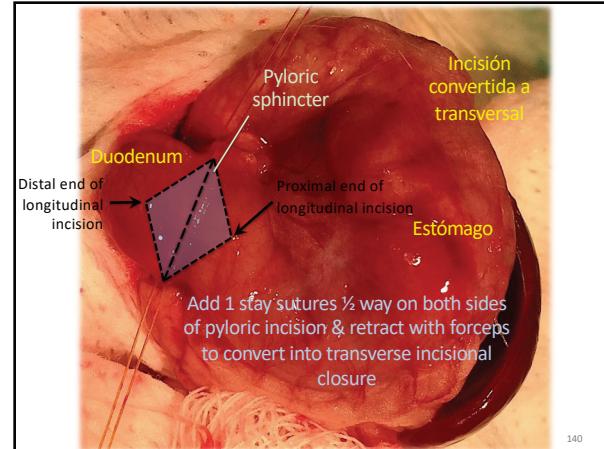
137



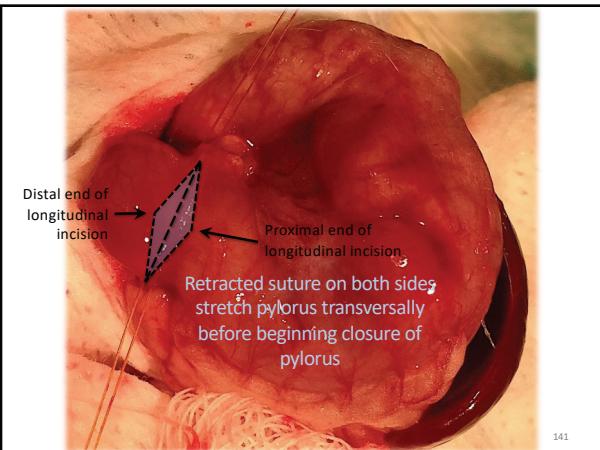
138



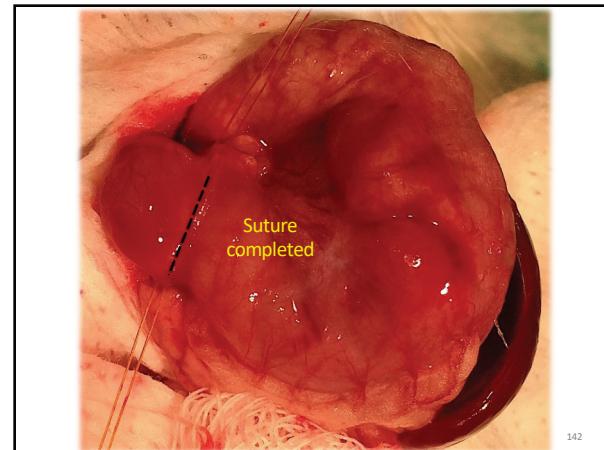
139



140

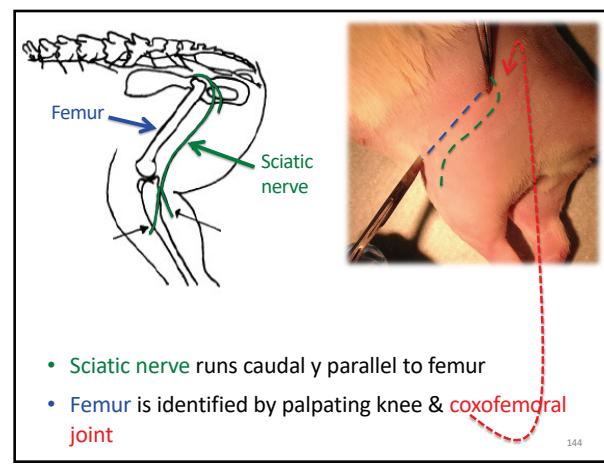


141



142

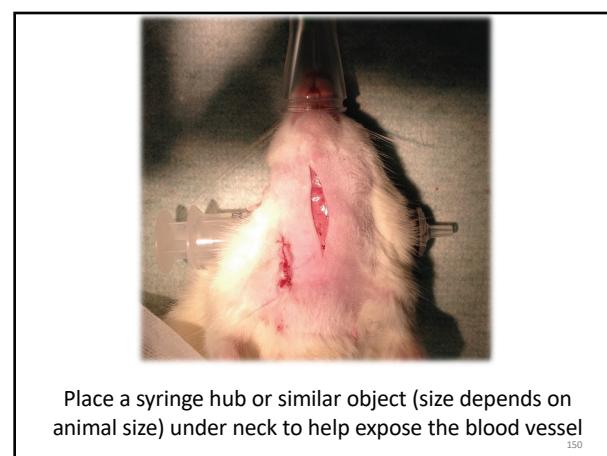
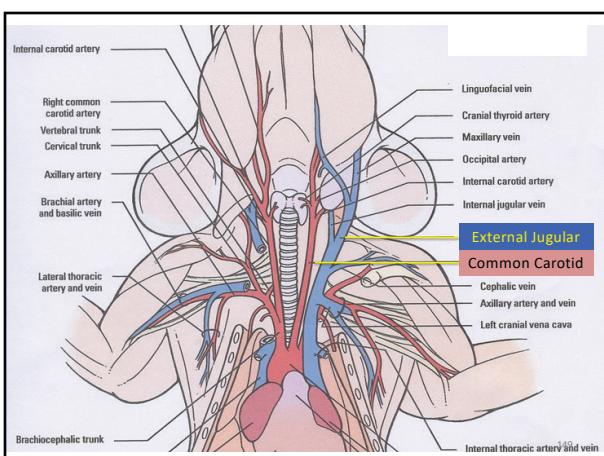
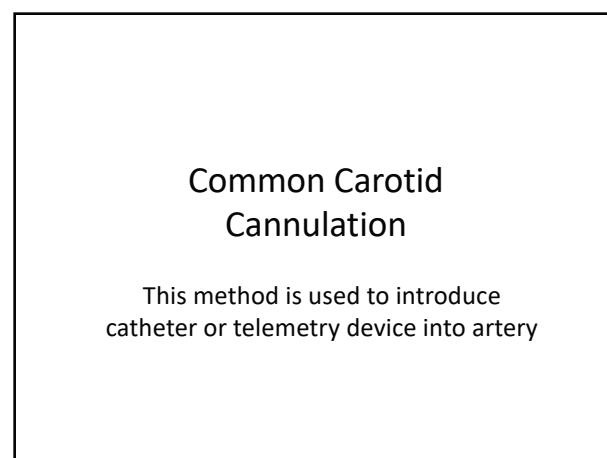
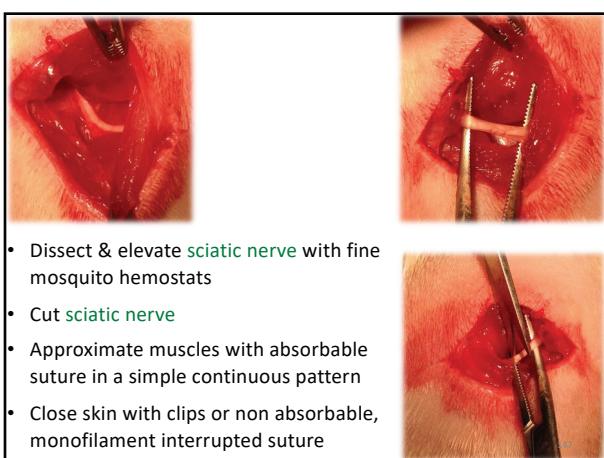
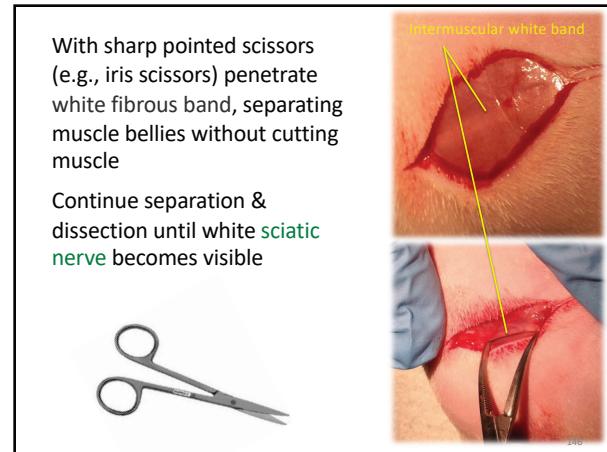
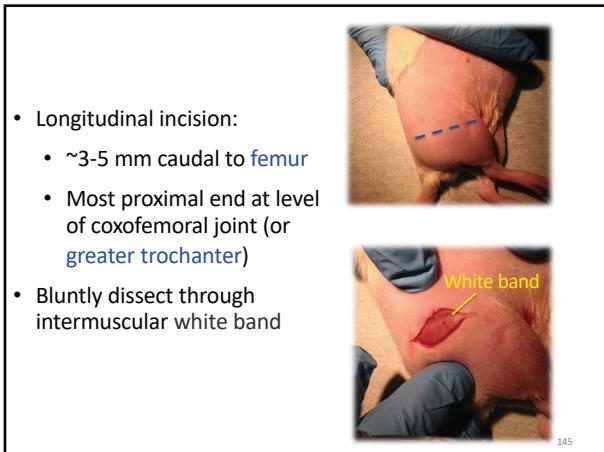
Sciatic Nerve Excision

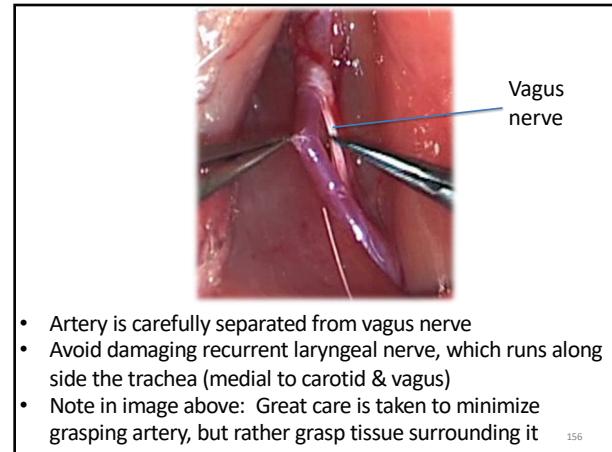
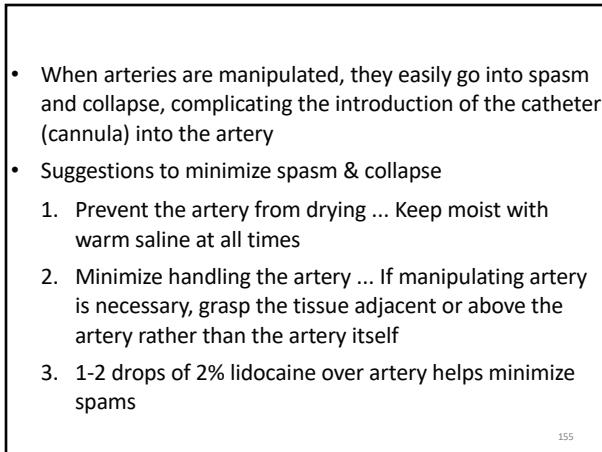
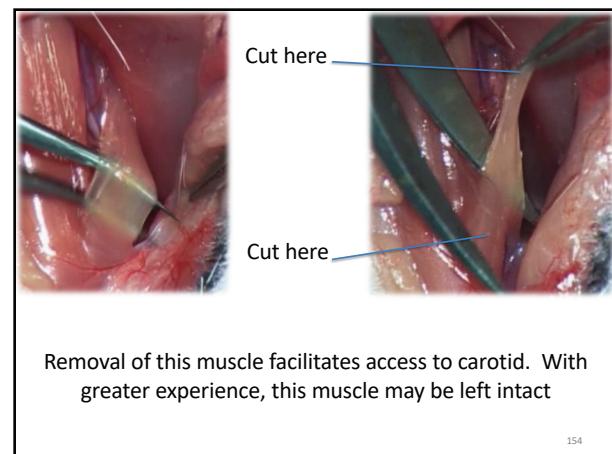
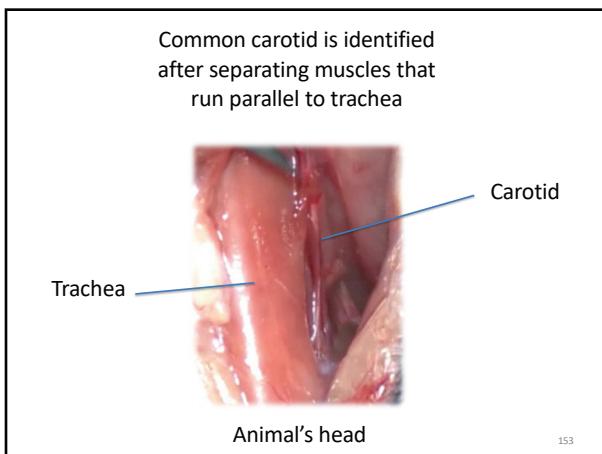
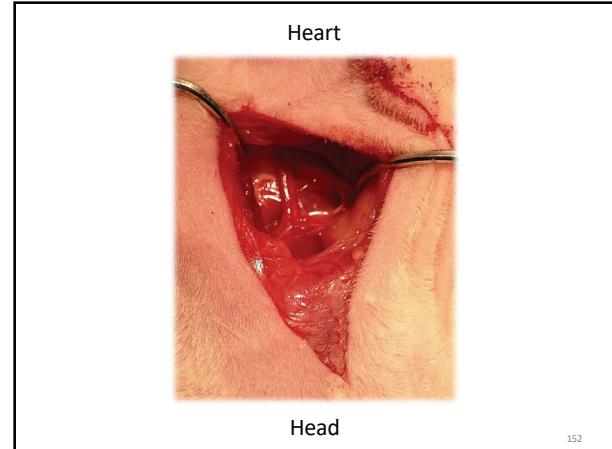
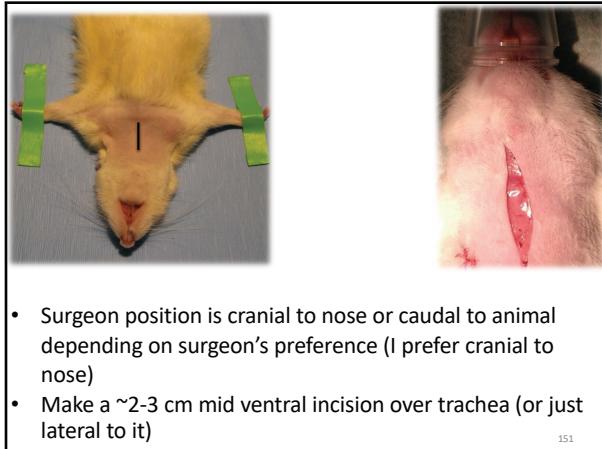


144

143

144





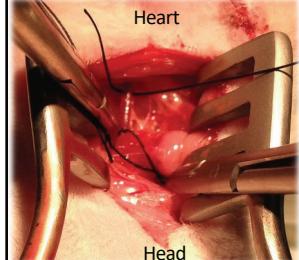
Heart



Head

157

Heart

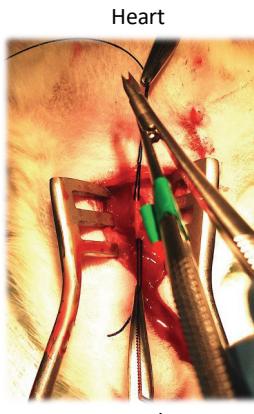


Head

- Place 2 ligatures distal & proximal to where catheter will be introduced. 3 ligatures can be placed for added security
- Distal suture (towards head) is tightened to ligate artery
- Proximal suture (towards heart) is kept loose

158

With forceps place tension on proximal suture (closest to heart) to kink artery
This avoids excessive bleeding when artery is entered (incised)



Head

159

Heart



Alternative to kinking:

Artery can be occluded with vascular clamps proximal (closer to heart) to artery incision point



Head

160

- ~50% of artery's diameter is incised
- Arterial incision should be made close to distal suture (closest to head)



Head

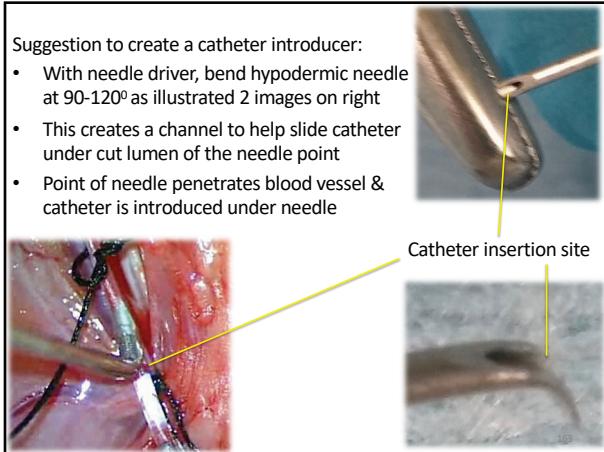
161

Suggestion to create a catheter introducer

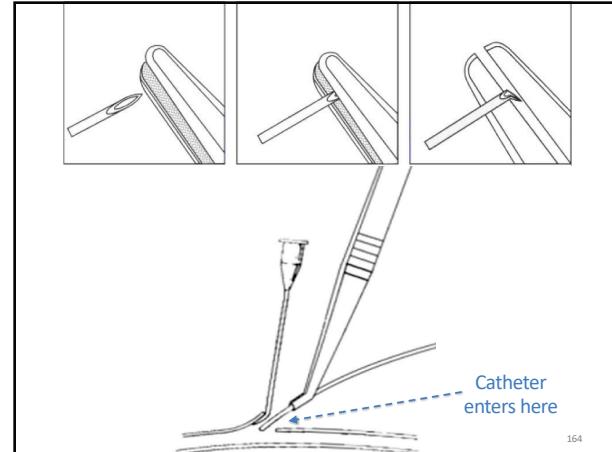
162

161

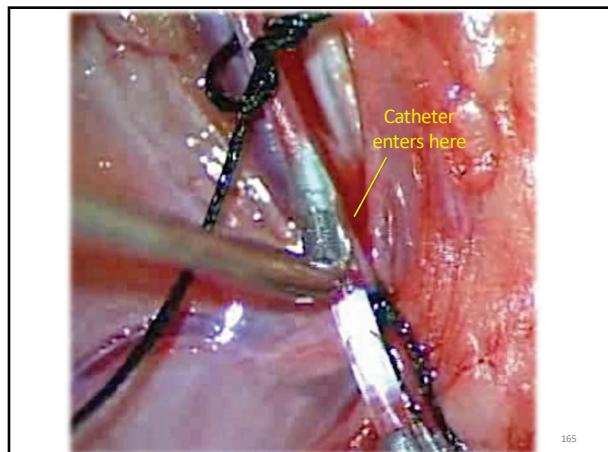
162



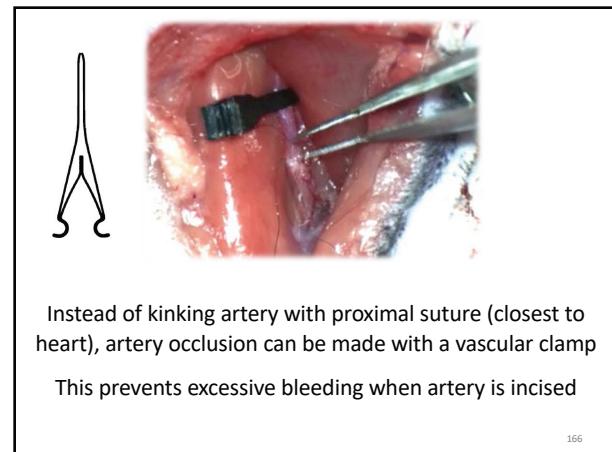
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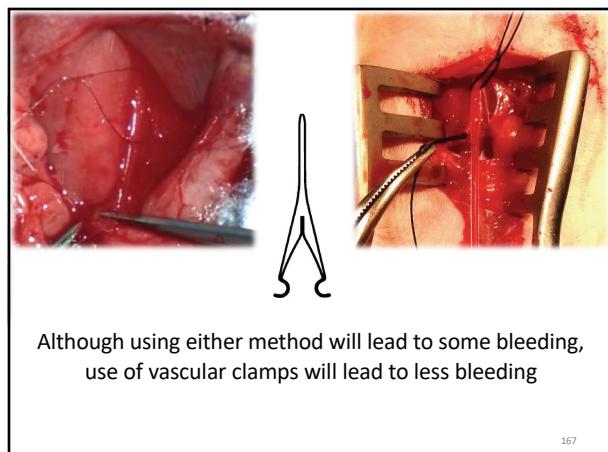
164



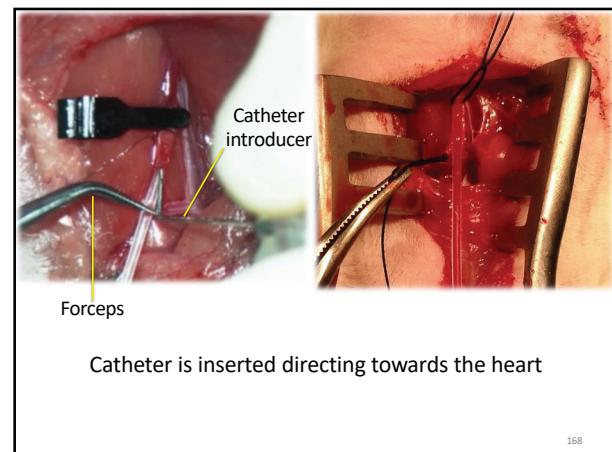
165



166



167



168

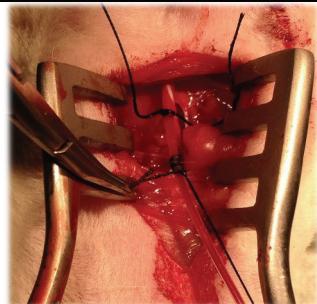


Suggestion: Catheter forceps (with internal grooves) are useful tools to grasp & introduce catheter into artery or vein, while minimizing damage to catheter

169



170



- After introducing catheter, tie proximal suture over artery and catheter
- Tie distal suture around exteriorized catheter

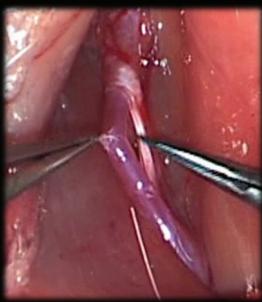
171

Common Carotid Artery Catheterization Video

4:24

172

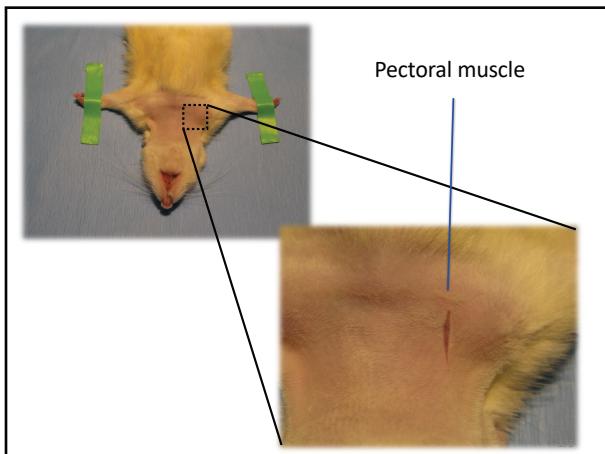
Common Carotid Artery Catheterization Video



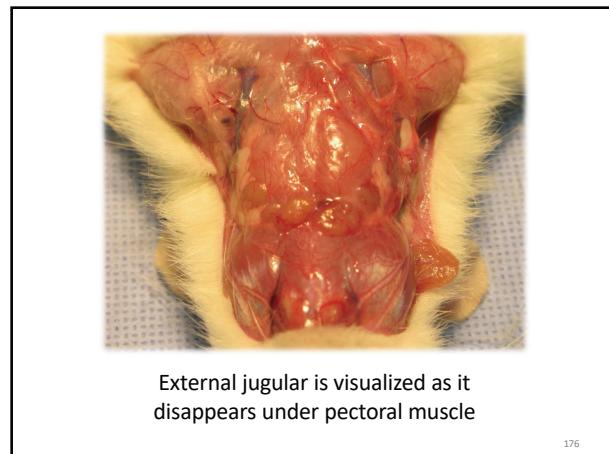
173

External Jugular Vein Catheterization

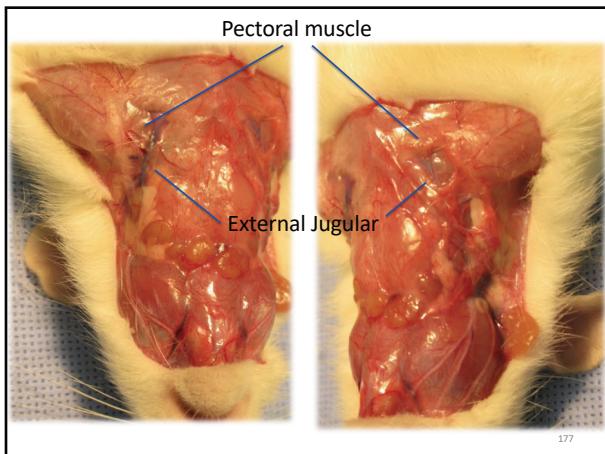
174



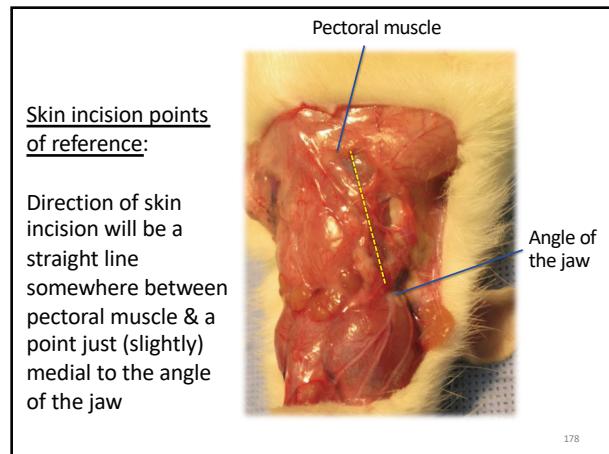
175



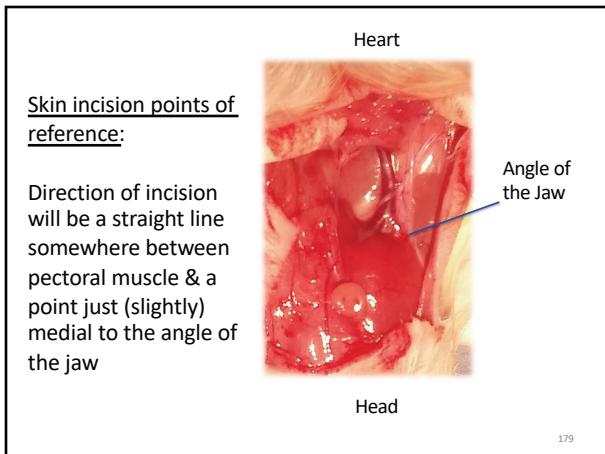
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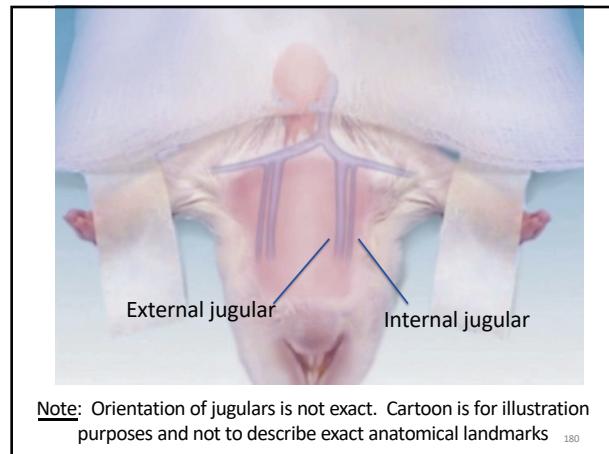
177



178



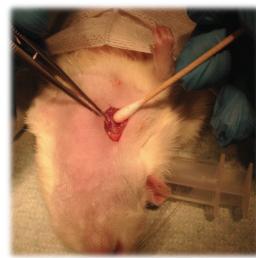
179



180

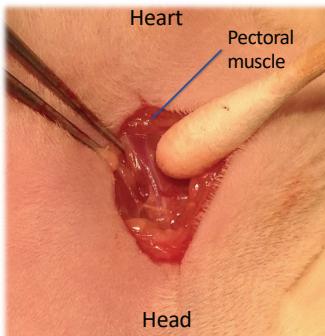


Place a syringe hub or similar object (size depends on animal size) under neck to help expose the blood vessel
181

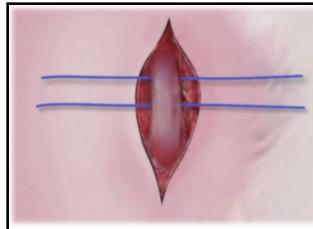


- Surgeon position is cranial to nose
- Make a 1.5-2.5 cm incision
- Incision started at level of pectoral muscle & directed towards angle of jaw (or slightly medial to it)

182



Once located, dissect external jugular free of surrounding tissue
183



Place 2 sutures under vein

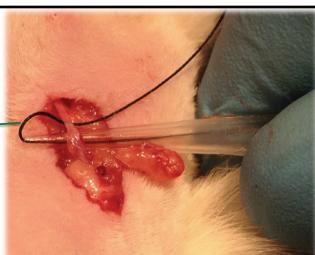


Cut here

184

Suggestion for passing suture under blood vessel

Grasp suture here – (this is the “other” side)



- Insert suture through hub of small/sterile pipette
- Force tip of pipette under vessel while rotating pipette back & forth about its longitudinal axis
- Once through the “other” side, grasp suture with forceps
- While grasping suture on “other” side, remove pipette & leave suture in place

185

Caudal suture (heart)

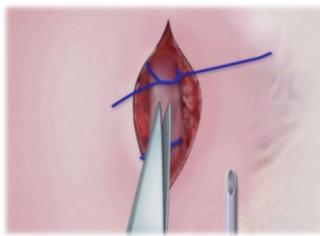
Cranial suture (head)

Caudal suture (heart)

Cranial suture (head)

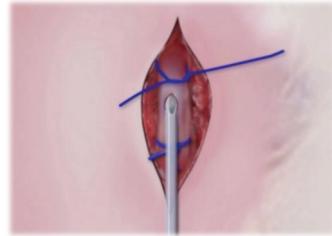
- Most cranial (towards head) ligature is tied and knotted
- Most caudal (towards heart) ligature is left loose without tying (for now)

186



~25% of vein's diameter is incised

187



Catheter is introduced directed towards (in the direction of) the heart

188



- After introducing catheter, tie caudal (towards heart) suture over vein and catheter
- Tie cranial (towards head) suture around exteriorized catheter

189

External Jugular Catheterization Video

6:41

190

External Jugular Catheterization Video



191

Femoral Artery and Vein Catheterization

192

- Femoral Vein & Artery catheterization is performed following the same principles discussed earlier in this presentation for the common carotid and external jugular sections
- The difference is how to approach & manipulate the femoral vessels. See following slides for description of these differences
- Femoral Vein, Artery & Nerve run adjacent and parallel to each other. Their orientation from posterior to anterior they run in this order, VAN (Vein, Artery & Nerve)

193

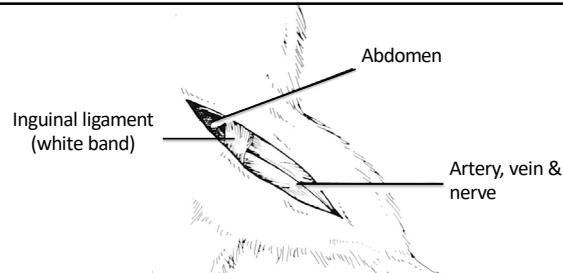


- Incision is made longitudinal or perpendicular to the femoral furrow according to the surgeon's preference
- Incision is made on the medial surface of the thigh over the areas traversed by the femoral Vein, Artery & Nerve, with its most extreme cranial end (if incision is longitudinal) at the level of the inguinal ligament

194

Immediately after making the incision, a body of fat (fat pad) is encountered, which is retracted away or excised to visualize femoral Vein, Artery & Nerve lying right underneath this body of fat

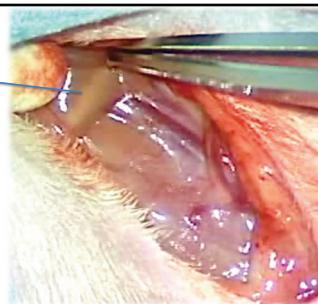
195



- Incision should expose inguinal area, evident by visualization of a white band of tissue (inguinal ligament)
- Vein, Artery & Nerve disappear proximally under the inguinal ligament as they enter into the abdominal cavity

196

Inguinal ligament
(white band)



- Incision should expose inguinal area, evident by visualizing a white band of tissue (inguinal ligament)
- Vein, Artery & Nerve disappear proximally under the inguinal ligament as they enter the abdominal cavity

197



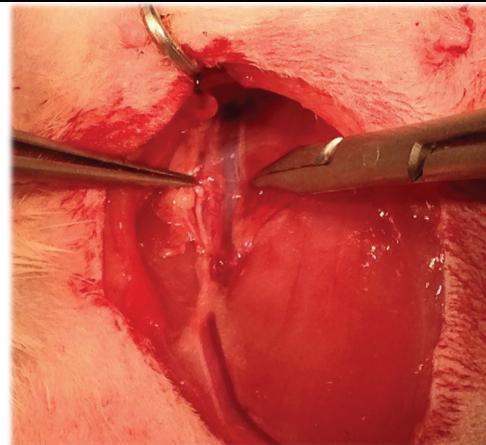
Separation of Vein, Artery & Nerve can be done with micro-scissors, fine tip forceps or hydrodissection

198

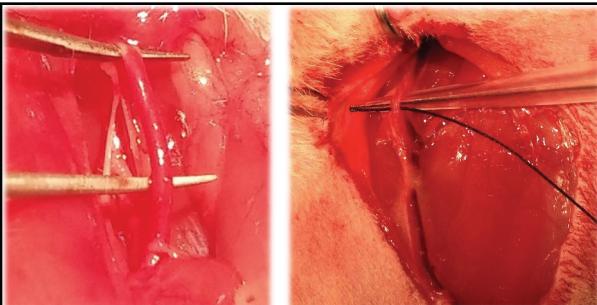


Hydrodissection is performed by inserting a lacrimal cannula attached to a 1 ml syringe between vessels, followed by injection of small volume of saline to separate structures

199



200



Femoral Vein or Artery catheterization is performed following the same techniques described earlier in this presentation in the common carotid and external jugular sections

201

- Subcutaneous tissue is approximated with absorbable suture in a continuous pattern
- Skin is closed with
 - Monofilament, non-absorbable suture in an interrupted fashion, or
 - Surgical Clips

Surgical skin glue (cyanoacrylate) may be applied to reinforce (and provide a microbial barrier) the incision

202

202

Femoral Artery Catheterization in the Rat Video

8:04

203

Femoral Artery Catheterization in the Rat Video



204

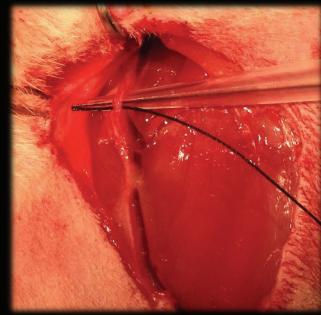
204

Femoral Vein Catheterization in
the Rat Video

4:01

205

Femoral Vein Catheterization in the Rat
Video



206

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Training Resources:
<https://research.utsa.edu/compliance/larc/training.html>



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