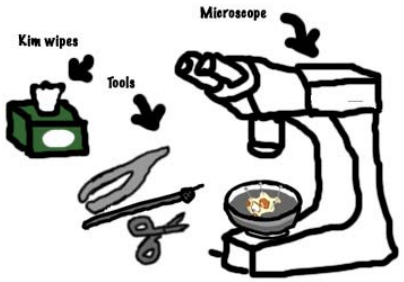
Cancer Borealis: Fine Dissection
By Gabrielle Gutierrez for the Marder Lab, Brandeis University

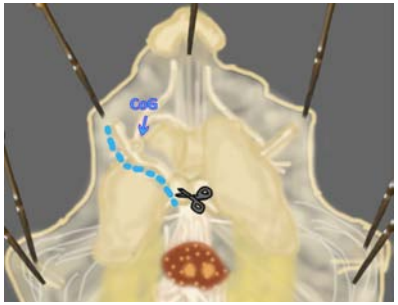
For the fine dissection you will need your microdissection tools, a dissecting microscope, and some kimwipes. Make a sylgard-coated dish with fine wire pins ahead of time.



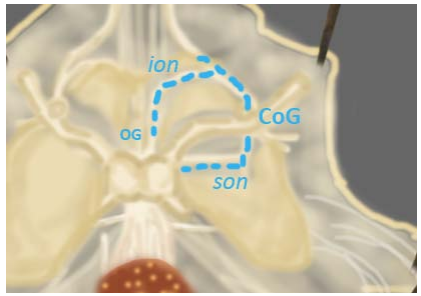
Re-pin the prep to make sure it is taut in the dish. Pin the flaps by the lip. Gather the extra tissue on either side of the STG and pin that down too.

Trim the hypodermis leaving only a patch with the two large dots on it. The STG is under there, so be careful.

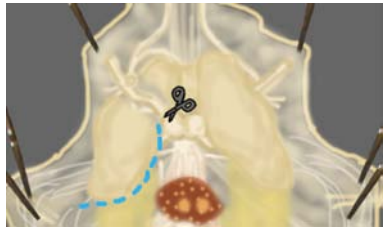
Locate the brain and follow the thick processes exiting it toward the lip. Those are the CoGs.



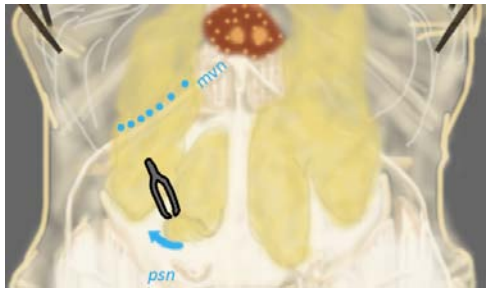
Follow the processes leaving the CoGs, the *ion*s and *son*s, clearing tissue away from them till you reach as close to the OG as possible.



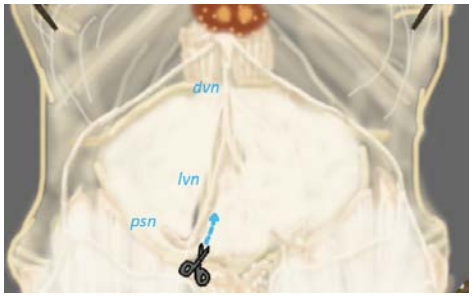
Cut off the "omelettes", those fluffy, pale-yellow blobs over the CoGs.



Peel up the yellow, wispy tissue toward the bottom of the prep. Get rid of as much of it as possible, especially the stuff right next to the STG. The *mvn*s are usually weakly attached to this stuff so use the blade of your scissors to clear it off of the nerves.



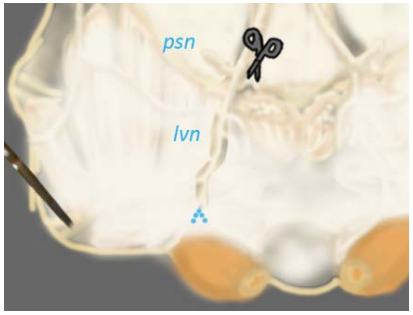
Locate the *psn*, which is usually sticking out over the crescent-shaped ossicle. Follow it into the delicate, white tissue where it meets the *lvn*. Cut through that tissue until you find the *dvn* where the *lvn*s branch from.



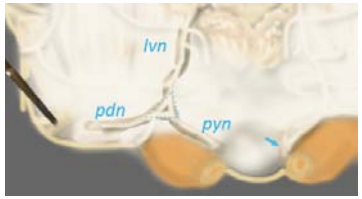
Cut through the delicate, white tissue to uncover both *lvn*s and the *dgn* if possible. The *dgn* branches many times under the *lvn*s and *dvn*. The *dgn* innervates the long muscles that lie beneath the nerve.

These may be all of the nerves that you need, in which case you can skip ahead to cutting the STNS loose. For experiments where additional nerves are needed, many of them are found in the bottom part of the prep near the pylorus.

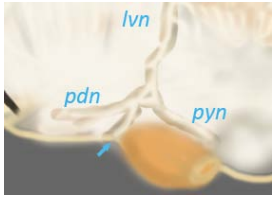
The *psn* branches from the junction between the *lvn* and the *dlvn*. Cut down through the delicate white tissue to continue uncovering the *lvn* until you reach the branch point right above the pyloric ampulla.



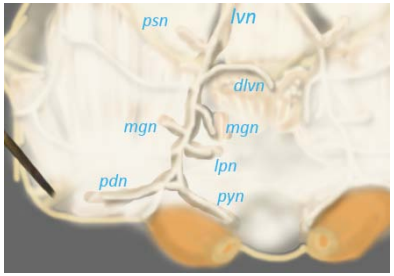
The *pyn* and *pdn* branch off of a little triangle. The *pyn* wraps around the top of the ampulla, the *pdn* is lateral to the *pyn* and lies over the muscles it innervates. Sometimes the end of the *pyn* sticks out making it easier to start there and work up to the triangle.



The nerve wedged between the lateral side of the ampulla and the muscle can be mistaken for *pdn* but often contains a PY signal.



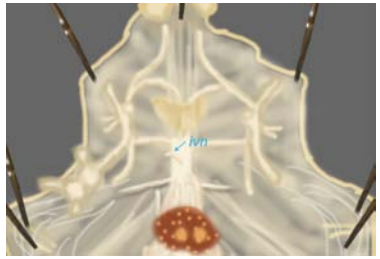
Uncover the *lpn*, *mgn*, and *dlvn*. An MG signal can be found on either a lateral or medial nerve. The *dlvn* wraps over the gm3 muscles.



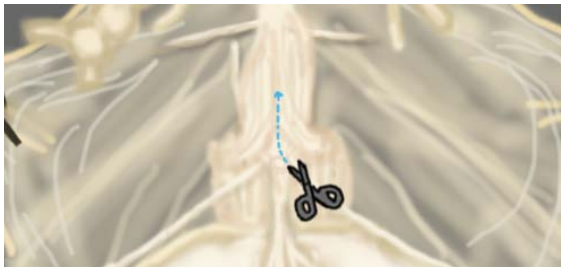
Uncover the length of the *lgn* till it joins the *mvn*.

Even though the *mvn* has VD and IC signals on it, for either of those cells individually follow the *mvn* toward the bottom. Where the *mvn* branches and enters the cv1 muscle all the way at the edge is where you can get a nerve with only VD on it. The branch right next to that goes into cv2 where you can get IC alone.

Sever the brain from the rest of the STNS by cutting the *ivn* and cut one CoG from the brain.

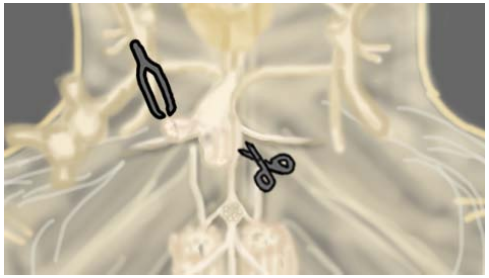


Now cut the spotted hypodermis patch off, creating an opening in the artery that surrounds the STG. Cut through the opening in the artery to separate the two muscles that flank the STG.

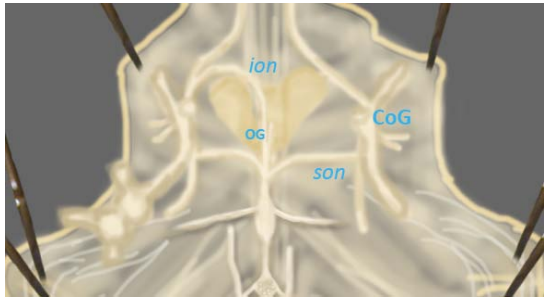


Lift the end of one of the flanking muscles and gently cut it away from the rest of the artery. The *aln*s are right underneath. Cut as far up as the cartilage that joins the flanking muscles. The flanking muscles can be cut off at this point.

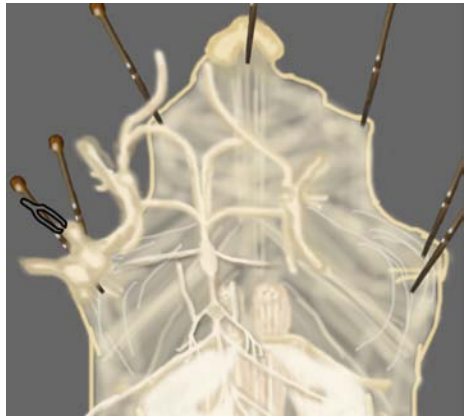
The *stn* is right underneath the strip of cartilage. Cut right under the cartilage to separate it from the *stn* up to where the cartilage becomes softer tissue. At that point the cartilage stub can be cut off.



Locate the *stn* and cut through the thin tissue right above it. Keep cutting until you reach the OG where the *ion*s branch off. The *son*s branch off just slightly below that. Uncover each *ion* and *son* by cutting through the remaining tissue on top of them.



Starting with the CoGs, cut the end of each nerve from the rest of the stomach tissue until the entire STNS is separated from the stomach. The brain can grabbed with the forceps to move the STNS out of the dish.



Condition a Sylgard-coated petri dish by rubbing the remaining stomach tissue over the surface. Place the STNS into the dish, add cold saline, and pin the STNS down taut. The STNS is now ready for desheating and extracellular recording.