Quick & Dirty Methylene blue procedure

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Demo before students start their dissections.

Methylene blue stock should be about 0.01% solution, made with saline. Depending on need—dilute this to by another 10:1 (saline: stock blue) or even 50:1.

For the slow flexor preparation

Do dissection as shown in video, for the physiology prep

 Expose the superficial muscle layer

 Expose the 3rd (or any) ganglion and central nerve cord (connective) between ganglion 3 & 4.

Drop the saline level just below the level of your dissection—the idea is not to flood the ganglion & muscle with saline.

Cut a strip of kimwipe or tissue to just cover the ganglion & connective and another strip of tissue to cover the muscle layer. Apply a few drops of methylene blue solution (dissolved in saline) to the tissue paper so that it is good and moist.

Put the prep in the fridge and check it every 15 minutes or so. The prep should be well-stained within a half hour, maybe sooner depending on stain concent ration. You will be looking for the 3rd root—showing where it exits the connective and how it projects to the flexor muscles—if you are lucky, you will see individual nerve terminals as they innervate each muscle fibers (the likely-hood of getting a prep like is about one in three).

This is pretty fool proof. Just make sure that before you pop the prep into the fridge for “stain development & uptake into the cells that the cord & muscle are kept most with the methylene blue-saline solution—don’t let it dry out—but don’t flood the to-be-stained prep with the bath saline, which would severely dilute the stain in the kimwipe strip.

This is very useful to show the students “what they’re looking for” before they begin the dissection. This is also shown in the Crawdad video but it’s useful to have a “in living color, 3-D” prep that they can see for themselves, unmediated by a computer monitor screen.