

Instructions for Cutting Paraffin Sections

MATERIALS NEEDED:

- ✓ **Treated microscope slides** (*Silane, Poly-L-lysine*) or positive charge (“Plus”) slides.
- ✓ **Slide box** to hold slides with sections
- ✓ **Artist paint brushes.** The lab has some available to use, but you may want to purchase your own.
- ✓ **Paraffin embedded samples.**

1. Set water bath to **44°C**. This can be set higher if tissue sections are thicker, or lower if sections break apart or the tissue is fragile.
2. Cut the block edges with a razor blade into a trapezoid to help sections cut more easily and help with section orientation.
3. Using the **coarse retraction button**, move arm on the microtome ALL the way back, **LOCK HANDLE** and insert block.
4. Use the left lever to loosen the blade holder and move blade close to, but not touching, the block. Tighten the left lever.
5. **The blade is VERY SHARP. Avoid touching it with your fingers.**
6. Set to “trim”, cut **30 µm** sections until you start cutting into the tissue. Adjust block angle as needed. **NOTE: Be sure to LOCK HANDLE to remove block, replace block or when doing any additional trimming with razor blade while on the microtome.**
7. Remove tissue block and put into the water bath for a few seconds to rehydrate tissue, pat dry, and then put in the **-20°C** for **~5 min** to harden paraffin.
8. Label charged (‘Plus’) or ‘silaned’ slides with a #2 pencil or histology marker while block is cooling.
9. Set microtome to cut (‘Feed’) **5 µm** sections (or desired thickness).
10. Cut a long ribbon (as long as things go well) with enough sections for your intended number of slides.
11. Use paint brush to move ribbon to room temperature **50%** ethanol (this helps spreading of tissue).
12. Use razor blade to cut ribbon into the desired number of sections for each slide.
13. Pick sections up with a large untreated slide, move to warm water bath (set to **44°C**), and let them float.
14. Pick up section by moving a labeled, treated slide underneath and lifting it up into the tissue at an angle (*this requires practice*). **NOTE: you cannot reposition sections once they adhere to the slides!**
15. Tap off excess water and dry bottom of slide, place slide in rack to drain and then move slides **to slide warmer** (set at **~44°C**).
16. Leave overnight on slide warmer to dry.