Slide Subbing Recipe & Protocol

Subbing Solution	**500 mL**	**1 L**
– *Gelatin type A*	*2.5 g*	*5 g*
– *Chromium potassium sulfate dodecahydrate ($CrK(So_4)_2 \cdot 12H_20$)*	*0.25 g*	*0.5 g*
– *MilliQ water (H_2O)*	*500 mL*	*1 L*

Materials needed

1 L / 2 L beaker
500 mL / 1 L bottle
Absorbent bench liner
Filter paper
Funnel
Glass slide racks (for dipping slides)
Hot plate
Magnetic stir bar & retriever
Microscope slides
Plastic slide racks (for drying slides)
Square glass container(s)
Thermometer

Protocol

1. Add water, magnetic stir bar, and thermometer to a 1 L (or 2 L) beaker.
2. Using a hot plate, heat water to ~40°C with the heat element set low (1 or 2).
    a. Make sure temperature does not exceed 45°C.
3. Slowly add gelatin to water and stir for 5 minutes or until completely dissolved.
4. Add $CrK(So_4)_2 \cdot 12H_20$ and stir until completely dissolved.
    a. $CrK(So_4)_2 \cdot 12H_20$ will positively charge the slides, allowing them to attract negatively charged tissue sections.
5. Remove the stir bar and thermometer. Filter solution into a 500 mL (or 1 L) bottle.
    a. Wait for subbing solution to cool to room temperature before use. Solution may be placed in the fridge to speed up cooling process.
    b. If not using immediately, store in the fridge. When taking out the solution again, let it adjust to room temperature and then filter before use.
6. Place microscope slides into glass slide racks. A box of slides will fill around 8 glass slide racks.
7. Clean slides by submerging racks in soapy water and then transferring to tap water. Rinse off with MilliQ water.
8. Pour subbing solution into a square glass container to ~3/4 full. If working with another person, fill two containers in order to sub slides at the same time.
9. Dip racks containing clean slides 3 – 5 times (for ~5 seconds each) into the subbing solution.
10. Individually move subbed slides into plastic slide racks for drying. A box of slides will fill around 4 plastic slide racks.
    a. To remove bubbles, slides can be dipped individually into subbing solution. Blot excess solution from a slide by tapping its edge against absorbent liner.
11. Cover racks with paper towel to protect from dust and leave to dry for 48 hours.
12. Transfer dried slides to original slide containers and store at room temperature until use. For slides to be used for cryostat sections, store in the freezer at -20°C.