

## **Lead Staining of EM Grids**

All solutions are contained in syringes with 0.2um filters. **\*\*Change filters on syringes before beginning to stain.\*\***

1. Prepare a parafilm template using an 8-12" piece of 4" parafilm and a Sharpie marker. **\*\*Do not touch the surface of the parafilm with your bare hands prior to staining.\*\***
2. Sato's Lead: 2.5 - 5 minutes
3. Use a petri dish (preferably glass) to draw a circle on the parafilm. Line the inner rim of the drawn circle with NaOH crystals. Place one drop of lead solution inside the NaOH ring for each grid you plan to stain. Place the grid section side down on the lead droplet, so that the grid is "floating."
4. Wash a total of 4 times for 1 minute each with distilled water
5. While the grids are floating on the lead droplets, lay down four columns of distilled water droplets (one drop for each grid). When the grids are done with their lead staining time, transfer them the droplets in the first column of water. After 1 minute, transfer them to the second column. Continue this process until the grids are in the fourth column.
6. Uranyl Acetate: 30 minutes
7. After the grids have gone through the distilled water washes, lay down one drop of uranyl acetate solution (saturated in 50% methanol; the solution will be yellow) for each grid you wish to stain. Transfer grids as above.
8. Wash a total of 4 times for 1 minute each with distilled water
9. See Step 3.
10. Sato's Lead: 15 minutes
11. See Step 2.
12. Wash a total of 4 times for 1 minute each with distilled water
13. See Step 3.
14. Dry grids and return to grid box

Dry grids by wicking them with a piece of filter paper.