

Imaging Cores – Electron

General Guidelines for Handling Grids

Technical Tips

1. Grids shiny side vs dull side

Adhesion of sections on grids during post staining and immunoelectron microscopy procedures is relevant in the electron microscopy laboratory. Grids are manufactured with a dull/rough side, and a shiny/smooth side. For grids with e.g., Formvar or carbon film, the smooth/shiny side should be used. In all coated grids the film is positioned on the shiny side of the grid. Epoxy sections exhibit a bumpy surface when viewed in the water boat. Scanning electron microscopy images of epoxy sections without embedded material also reveal an uneven surface.

2. Removing a charge from the surface of grids

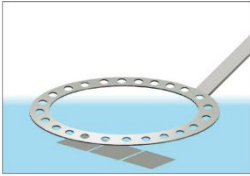
Sometimes when you are trying to pick up sections, they won't adhere to the grid surface. If you don't have time to glow discharge clean the grid surfaces, try this little trick. Dip the grids in distilled water for a moment and wick off the excess with filter paper. Let them dry while you are arranging your sections. Your sections should now adhere to the grid surface. Some labs soak the grids they will use for the day in distilled water until they are needed. If this procedure fails, reclean your grids with acetone or chloroform or glow discharge clean the grid surfaces. Reference: Jeanette Killius, NEOUCOM, Rootstown, OH.

3. On-Grid enhancement for post-embedding immunostaining

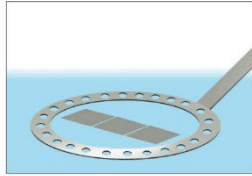
The use of nickel grids is recommended for on-grid enhancement, as nickel is relatively insensitive to silver enhancement. Gold or copper grids should not be used.

4. Section Pick-up with the Loop

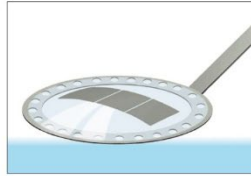
The use of a loop with the outside diameter same as the grid allows you to pick up sections consistently without causing any damage to the sections. Due to the fact that the loop and the grid are of the same diameter they are attracted to one another when in water and attach together through the surface tension of the water. Even if the section touches the inside of the grid during blotting the touching area is minor and, therefore, the section is not damaged. When the grid is removed from the loop the section remains in place without fail. The area equals the observation field (about 2mm diameter) of the electron microscope; thus, pieces can be fully observed.



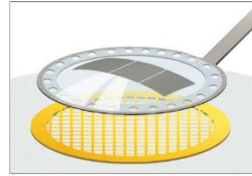
1. Centre the Loop above the sections.



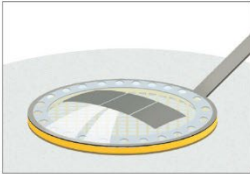
2. Slowly lower the Loop and touch the water.



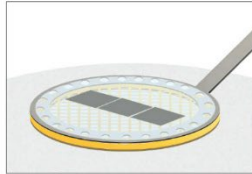
3. Gently lift up the Loop with the sections.



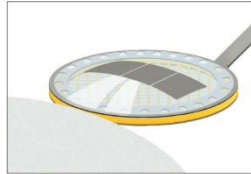
4. Bring the Loop above the grid on the filter paper.



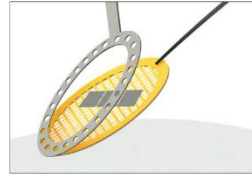
5. The grid sticks to the Loop by surface tension.



6. Lower the Loop to the filter paper to remove water.



7. For coated grids, touch with filter paper to remove water.



8. Separate the grid from the Loop with an eyelash.