

UNC CryoEM Core Convert QF to UltraAufoil

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Use protocol described by Christopher J. Russo and Loria A. Passmore with modification to procedure including (1) sputter coat TEM grids with Au using KJL PVD 75 and (2) remove carbon film by O₂/Ag plasma cleaning using TergeoEM. Protocol takes ~ 1-3 hours.

Use Quantofil Au 300 mesh R0.6/01, Lacey F/C Au 400 mesh or CF-1.2/1.3 Cu T-50.

Step 1. Sputter coat TEM grids with Au (1-2 hours)

1. The KJL PVD 75 Sputterer is located in CHANL in the clean room in Chapman Hall.
2. Instructions to operate the KJL PVD 75 Sputterer are online
3. <https://chanl.unc.edu/wp-content/uploads/sites/18385/2021/10/PVD-75-SOP.pdf>
4. Contact Bob Geil for training and questions regarding operations of the Sputterer.
5. Put TEM grids carbon side facing up inside PELCO Clamping TEM Grid Holder Block (16830-45).
6. Install Au target on position #1.
7. Mount the TEM Grid Holder Block on the Substrate holder using clamps and/or tape.
8. Follow instructions, on Run deposition select Au Sputter – set Final Thick to 0.46 kÅ (450 Å) check parameters before running. Run deposition when Vacuum reaches ~ 5E-5 Torr.
9. For deposition of carbon select Graphite Gun #3.
10. *If pumping the chamber takes longer than normal ~5 mins, first manually turn off/on the Roughing pump, and/or manually control the Roughing and TMV pump (Roughing → open PC Turbo Backing Valve → Turbo Pump PC).*

Step 2. Remove carbon film with O₂/Ag plasma (15-30 mins)

1. The TergeoEM plasma cleaner is located at the CryoEM Core in Glaxo 008. Follow instructions for using the plasma cleaner. For questions contact Joshua Strauss.
2. Put the TEM grids mesh side up on glass side or PELCO TEM Grid Holder Block (16820-25).
3. Put TEM grids inside chamber and check O-ring before pumping the chamber.
4. Select Recipe #4 Remove Carbon (Direct Plasma, Power 38 Watts, Duty Ratio 255, 1PT Gas2, Oxygen 9 PT Gas 3 Argon, 7 mins). This will remove the carbon film (tested using C-Flat TEM grid).
5. Vent and remove TEM grids.
6. Inspect TEM grids under LM before/after using.