Graphene oxide grids

GO!

Ingredients

Graphene Oxide stock - Sigma, 763705-25ml. Consider ordering this lot number – used previously (#MKCC0931).
Grids - we usually use Au300 R2/2 Quantifoil grids
Inverted anti-capillary-action tweezers
Whatman filter paper No. 1
Parafilm
$_{dd}H_2O$
Assorted tubes and pipettes

Protocol

Francesca Coscia leaves grids at least overnight before using, up to perhaps a week. However grids behave worse over time. Best to make however many you need for the freezing session.

Day 1 - Grid preparation:

- i) Glow discharge your grids
  - 40 mA; 1 min
- ii) Prep GO solution
  - Dilute stock 1 in 10 into MQ water in 1.5 ml (or 2 ml) tube. Pipette up and down to mix
    - Resuspend conc solution first by swirling
    - 450 ul MQ water in tube first, add 50 ul GO solution
    - Apparently can store for 1 month, but Francesca Coscia always dilutes fresh
  - Spin 600 x g until visible flakes pellet
    - We did for 45 secs (3 x 15 s)
    - If it's taking over 1.5 mins, try transferring to a 2ml tube (or 1.5 ml if you tried 2ml first, and try again)
    - Make sure there's no gradient of GO in the tube. This would mean you've centrifuged for too long
    - From this step, MOVE GO SOLUTION VERY SLOWLY.
  - Get another tube with approx. 2 ml MQ water in.
- iii) Grid preparation
  - Prepare parafilm with 3 x 20ul drops of MQ water per grid
  - Prepare Whatman filter paper like a tent
  - At 0s pipette 3ul GO onto grid (onto foil side)
    - Grid should be held in anti-capillary action tweezers
    - take GO from top
  - At 60s blot perpendicular onto paper
  - Take up 20ul drop of water from parafilm on GO side
  - Blot

Take up 20ul drop of water from parafilm on GO side
Blot
Take up 20ul drop of water from parafilm on reverse side
Blot as much as possible off
Leave grid for 10 mins to dry in tweezers.
After 10 mins, store in petri dish as normal.

Day 2 - Grid usage

DO NOT GLOW DISCHARGE

Be careful when pipetting on sample, you'll need to spread it around a bit by moving the tip while applying.