Chlorophyll extraction protocol

\*note the long incubation and plan accordingly\*

1. Add 10 ml of 90% acetone to 15 ml tubes using squirt bottle.
2. Label the tubes to match your samples. Transfer all info from foil packet label to tube label.
3. Keep the foil packet labels somewhere safe for reference.
4. **Working in a dark room**, transfer filters from foil packages to labeled tubes.
5. Invert tubes three times, then check that the filters are still submerged in acetone.
6. Wrap tubes in foil.
7. Incubate for 6 to 24 hours in refrigerator (2-6˚C). Invert tubes midway through incubation, checking that filters are still submerged in acetone.
8. Remove tubes from refrigerator and warm to room temperature on the counter.
9. Centrifuge tubes on centrifuge in PELL lab in IEC Clinical Centrifuge on setting ‘5’ for five minutes.
10. Read on Turner fluorometer:
    1. Put ‘chl-a NA’ module in fluorometer
    2. Turn on fluorometer using switch on back
    3. Select ‘chl-NA’ from the opening screen
    4. Calibrate: select ‘calibrate’, then ‘use stored calibration’, then select latest chlorophyll calibration (currently ‘Chl2Nov21’), then ‘select’
    5. Fill round glass cuvette ~about half full with sample
    6. Wipe cuvette with kimwipe, then place in fluorometer
    7. Select ‘measure fluorescence’
    8. Enter the volume filtered (usually 40 ml), then click ok
    9. Enter the volume of acetone used (usually 10 ml), then click ok
    10. Record value in notebook
    11. Repeat
11. When finished, all acetone should go in labeled waste acetone jug
12. Glass cuvettes get washed by triple rinsing in DI, then drying in drying oven