Notes:

* We should measure current. Some studies report that. We should also measure salinity and amount of suspended matter in the water
* Seems safe to assume no difference between leaf face

**1995 Dauby – Methods for removing epiphytes from seagrasses: SEM observations on treated leaves**

* A paper of all the things to avoid doing: shaking, scraping, sonicating, soaking in acid, waterflow, freezing , enzymatic clean
* SEM leaf prep methods –> old
* Removing epiphytes
  + Stirring and rinsing with DW is enough to remove a lot of the diatoms
  + pH is very important to consider in our sample prep. Acids bad for diatoms, bases bad for leaf. Should take a seawater sample from site and pH if we have to make buffers.

**2014 Majewska - Cocconeis Ehrenberg (Bacillariophyta), a genus dominating diatom communities associated with Posidonia oceanica Delile (monocotyledons) in the Mediterranean Sea**

* methods (for SEM)
  + spring sampling (2 years) to minimize “self-shading”
    - spring has “climax community” of diatoms. Not new leaf but no senescing
    - 20 shoots/year
  + SEM sample prep
    - 1-2cm2 leaf bits (basal, intermediate, and apical regions)
    - 1 h fix in 4% formaldehyde, 2.5% glutaraldehyde in filtered seawater
    - “Sandwich critical point drying method”
      * Rinse each sample with DW to remove extra fixative
      * Sandwish sample
      * Dehydrate in ethanol
      * Treat in critical point dryer (liquid CO2)
      * Mount and goal with gold-palladium
  + Did use TEM to analyze fine scale ultrastructures but ID the diatoms with SEM
  + Minimum (1 500 diatoms/sample)x2 replicates

**2009 Lebreton - Are epiphytes a significant component of intertidal *Zostera noltii* beds**

* Methods
  + Measured current and particulate matter in water (suspended)
  + Sample at two locations, 3 different times (spring, summer, fall)
  + Sample prep
    - In the field, fix in 4% fromaldehyde seawater
    - Rinse with DW
    - Dry
    - Image