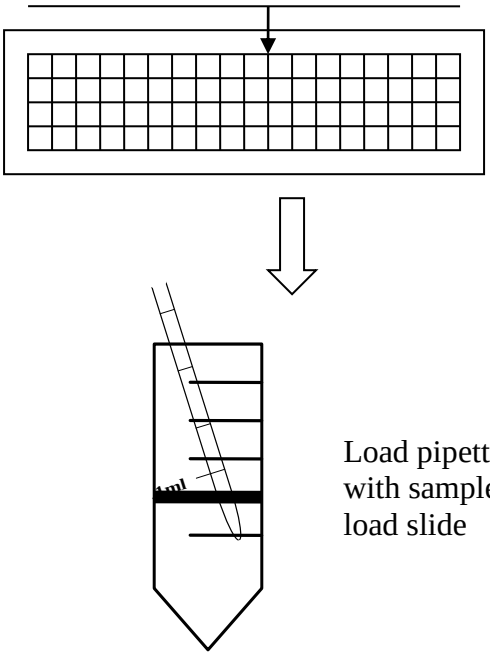
Sedgewick Rafter Counting Slide Protocol
Updated 030917

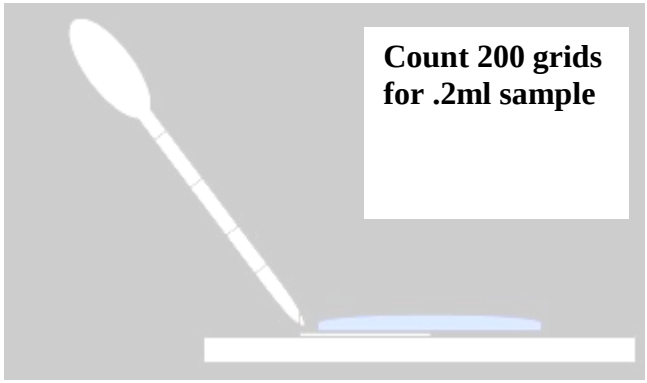
Samples should be examined within 24 hours.

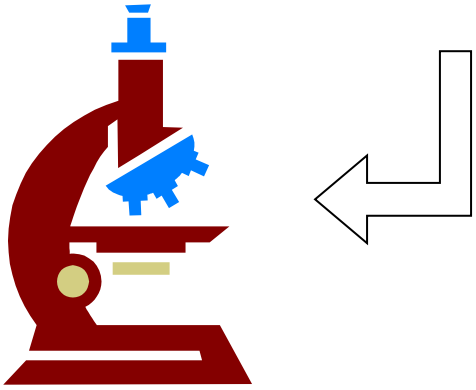
1. Invert tube three times to redistribute plankton in the sample.
2. Uncap and draw 1ml using a 1ml micropipette.
   - Cover the slide with a coverslip until only an opening no larger than the tip of the pipette can enter. This will prevent formation of air bubbles. Discharge 1ml from your pipette into the chamber.
3. Place the slide under your scope and scan under 4x to ID target species. Throughout process, take photos of target and biodiversity cells.
4. Begin count for target cells: *Alexandrium*, *Pseudo-nitzchia, Dinophysis spp., P. lima, and C. polykrikoides*. Analyze a total of **200 grids** (4 rows or 10 columns) using 10x magnification.
   - Be consistent when counting cells overlapping the grid lines, making sure they are counted only once.
   - Record number of grids analyzed if 500 or more cells of one target species are counted. Anything >500 becomes TNTC (too numerous to count). Analyze remainder of 200 grids for the other target species.
   - Count individual cells, not chains.
5. Biodiversity: Under 4x, identify the three most dominant species using the abundance index on the results spreadsheet.

1ml counting chamber



Load pipette with sample then load slide





Data Reporting and Target Cell Alerts

1) Complete all fields in Excel results spreadsheet. Calculate cells/liter using spreadsheet tab.
2) Target Cell Alerts should be issued to target cell distribution list for:
   a. AL ≥1ct
   b. PN ≥100ct
   c. DA/DN/DT ≥100ct

   Target Cell Alert Email should include:
   Station:
   Sample Date:
   Target Cell Species:
   Number of Grids:
   Cells/L:
   % Confidence in ID:
   Scotia Kit results (Positive/Negative/Invalid:
   *Picture of Scotia kit

3) If >2000 cells/L PN, complete Scotia test. Stop using Scotia tests after one positive test at site. If ≥15,000 cells/L PN, no Scotia test is necessary.
4) E-mail results spreadsheet to volunteer coordinator and volunteer network distribution list.