Protocol for Net Tow Relative Abundance Determination

Materials

- Net Tow sample
- Pipette
- Microscope
- Petri dishes
- Microscope slides
- Cover slips
- Vaseline
- Tape
- Razor blade

Using a Dissecting Microscope

- Invert the cod end containing the net tow sample several times in order to mix sample
  - Using a pipette, collect 3-5mL of sample water from below the surface of the cod end
  - Place 3-5mL in three different petri dishes
    - The desired volume of sample should completely cover the bottom of the dish but not much higher
- View all three petri dishes under the dissecting microscope
  - Identify as many organisms as possible
  - For organisms difficult to identify
    - Can you place them into a category – diatom, dinoflagellate, zooplankton, etc.?  
    - Take notes on color, approximate size (i.e. – ½ Lingulodinium cell), pattern of movement or lack of movement, shape
    - Can you take a picture of the organism?
    - These notes will help to identify the organism at a later date
      - Email a picture and/or description to HABWATCHHELP@USC.EDU for assistance in identification
- After identifying as many items as possible, it is time to assign relative abundances
  - Start with organisms considered RARE (<1% of the organisms in the sample)
    - These are organisms only seen once or twice
  - Continue assigning a relative abundance code (R, P, C, A or D) to all the organisms identified
    - In the comment section, you can take notes on the approximate percentage you are assigning to each organism
      - Make sure these numbers do not add up to over 100% when you are finished
    - Things to remember:
      - Move the petri dishes around and try to view as much of the sample as possible
• Focus up and down through the sample to see all the different planes of view
  o Non-moving cells (such as diatoms) have a tendency to sink to the bottom
  o Moving cells (any flagellates) can be present anywhere from the surface of the sample dish down to the bottom as they swim around
• While there may be ~10 chains of the diatom *Chaetoceros* in your sample, each chain contains several cells – sometimes as many as 10 cells per chain
• Do not ignore cells that you cannot identify
  o Try to place them into a broad category (i.e. – Unknown dinoflagellate)

**Using a Compound Microscope**
• Preparation of microscope slides
• Invert the cod end containing the net tow sample several times in order to mix sample
  o Using a pipette, collect a small volume of sample water from below the surface of the cod end
  o Place 1-3 drops of sample onto three different microscope slides
    ▪ If you do not have a depression slide, slides can be made using the following techniques:
      • Vaseline
        o Place a small amount of Vaseline on the palm of your hand
        o Drag each edge of a microscope cover slip on the Vaseline in order to have each edge covered with a small amount of Vaseline
        o The cover slip can then be placed over the drops of sample on the slide
      • Tape
        o Place several small strips of tape on a microscope slide
        o Using a razor blade, cut a small square out of the center of the strips of tape
        o The now tape-free center can hold a few drops of water from the sample with a cover slip placed on top
• View all three slides under the compound microscope
  o Identify as many organisms as possible
  o For organisms difficult to identify
    ▪ Can you place them into a category – diatom, dinoflagellate, zooplankton, etc.?  
    ▪ Take notes on color, approximate size (i.e. – ½ *Lingulodinium* cell), pattern of movement or lack of movement, shape
- Can you take a picture of the organism?
- These notes will help to identify the organism at a later date
  - Email a picture and/or description to SCCOOSHABHELP@USC.EDU for assistance in identification
- After identifying as many items as possible, it is time to assign relative abundances
  - Start with organisms considered RARE (<1% of the organisms in the sample)
    - These are organisms only seen once or twice
  - Continue assigning a relative abundance code (R, P, C, A or D) to all the organisms identified
    - In the comment section, you can take notes on the approximate percentage you are assigning to each organism
      - Make sure these numbers do not add up to over 100% when you are finished
- Things to remember:
  - Move the slides around and try to view as much of the sample as possible
  - Focus up and down through the sample to see all the different planes of view
    - Non-moving cells (such as diatoms) have a tendency to sink to the bottom
    - Moving cells (any flagellates) can be present anywhere from the surface, down to the bottom of the slide as they swim around
  - While there may be ~10 chains of the diatom Chaetoceros in your sample, each chain contains several cells – sometimes as many as 10 cells per chain
  - Do not ignore cells that you cannot identify
    - Try to place them into a broad category (i.e. – Unknown dinoflagellate)