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
    - 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 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
    - 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
    - Using a pipette, collect a small volume of sample water from below the surface of the cod end
    - 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
                - Place a small amount of Vaseline on the palm of your hand
                - Drag each edge of a microscope cover slip on the Vaseline in order to have each edge covered with a small amount of Vaseline
                - The cover slip can then be placed over the drops of sample on the slide
            - Tape
                - Place several small strips of tape on a microscope slide
                - Using a razor blade, cut a small square out of the center of the strips of tape
                - 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
    - 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 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)