cell lit.

DIRECT ENUMERATION ⟶ PROXIES

11/29/12

Stations Represented.

- SBC - Plumes ; Blooms
- CCE
- ~~BBO~~ / BATS
- SAN Pedro
- IP
- Autonomous DAP - Eurosite (R. Lampitt)
- BATS (M. Lomas)
- HOT (M. Church) picoplankton
- COPAS (F. Tapia) enso-oriented ; 10 yrs monthly obs.

Abandance phyto

Flow cytometry (direct counts) - small cells.
HPLC pigments (indirect proxy)
Fluorometry
Settling chambers + microscopy (larger taxa) - how is it counted?

Site specific differences in phyto will drive the method
HOT does not do settling chambers.
Fixed 1/2% Formaline (SPOT)

ICES - Chl (pigments) } consensus
microscopy (direct cell count) } of labs in N. Atlantic.

→ Counts reported in biovolume (units of C) and cell # / volume

PICOPLANKTON

1. FC
2. EF
3. ~~HPLC~~

site specific Coastal ts will have to use microscopy

Enumeration + size = C units

PHYTOPLANKTON

#3. Microscopy (inversion chamber) → taxonomy → C

#2. EF (some flow cytometers can do large cells, but its a ≠ setting)

#1. Fluorescence (Chl). It's not a proxy of cell abundance. Chlorophyll a could be intercomparable.

Have 2 methods + comparison?

Site - appropriate method (e.g. Chl + microscopy)

DIATOMS (diatoms and dinoflagellates) Chl + microscopy.

→ Calibration between Chl fluorescence (benchtop) and fluorometer from CTD

Sites that do multiple biomass meas. should report all of the related parameters

Combination of parameters will allow better understanding of syst.

* DNA extractions as a proxy of biomass. (future potential method)
* ATP - INDEX OF LIVE VS DEAD

STANDARDS: no real standards for biomass

FC: archiving and re-running samples as internal standard.

→ An exercise of sample exchange of archived samples is recommended (deep frozen samples last for up to a yr.)

→ Consensus standards are useful when there are no Reference material available

Nomenclature - Chl is conc.; cell abundance

ZOOPLANKTON

- net TOWS (mesh size)
- Optical Plankton Counter (OPC) - only gives you count; cannot ≠ phyto vs zoo. LOPC laser or otherwise;
- Bottles
- VPR (video plankton Recorder)

2. VPR / SIPPER

Recommended method (Mesozoopl.) — WP2 (type of net) recomm from UNESCO 1965

1. Net Tow (recommended size is 200 µm mesh size) → or DW.
3. Acoustic is ~~not~~ recommended for zoo (signal ≠ organism)
~~3.~~ Displacement volume (questionable) issue w/ Jellies
↳ non-destructive (fast too) but getting C is issue

DW is best ↓ Formaline x preservation. 8 week wait to get weight (on stabilized

Mesh size is very delicate - 200 µm ≠ 250 µm.
type of net is ship dependent (size)

Justifyable for larger organism (multifreq.) must be backed by net tows — Laboratory
→ counts
→ dry weight. (C)

Refrigerated best than formaline but must be done fast.

type of tow? Integrated tow (oblique) or vertical (vertical len material)

can cover large spatial scales

Assemblage of species you have - recommended to know a priori
Size of opening of net (e.g. 4x4 moc or larger) is dependent on site / objective / ship capability

to estimate flow through net w/ flowmeter. (electronic or mechanical GO flowmeters)

time / depth recorder integrated into the net so you can know the depths where the net was.

Day / night tows, or tows @ same time of day

~~Zoom~~ Cam? as proxy for taxonomy (cannot ID species but gives good information. ~~SIPPER~~? ← this is a lab thing, not for field.

Tolsam plankton splitter. MUST be used on land.

Emerging / perfecting tech - VPR, SIPPER (OPC is no good)

McLane ZPS does not work well for large zoo pl. Escape rate can be high?

BACTERIA

1. Flow cytometer
   microscopy (Epifluorescence?)
   DAPI staining ; cyber green staining , A orange , Hoechst
   ↓ stains very specific cells, good x enumerating
   ↓ by glow. (biovolume)
   ↓ DNA specific.

Recommended methods

1. EF – ~~bleck~~ 0.2 filter (polycarbonate) – pre staining is recommended
2. FC

Biovolume constrained w/ image analysis

Stain in open ocean do not matter too much
A. orange can be an issue in coastal ocean.

Flow cytometer 101 – Know what's there!
- There are several models
- centralized lab for flow cytometry is a recommendation for future

VIRUSES

FC (not yet tested)
EF microscopy

Subject to operator error (must be overlap between counts)

HETEROTROPHIC MICROPLANKTON / NANO

1. microscopy (autofluorescence) → proflavin (vital preservative 5%) → { Site specific filter
2. Flow cyt. separate by size class.