Cyanobacteria
Phytoplankton analysis & biomass determination

Jeff Janik, Ph.D.

California Department of Water Resources

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Overview

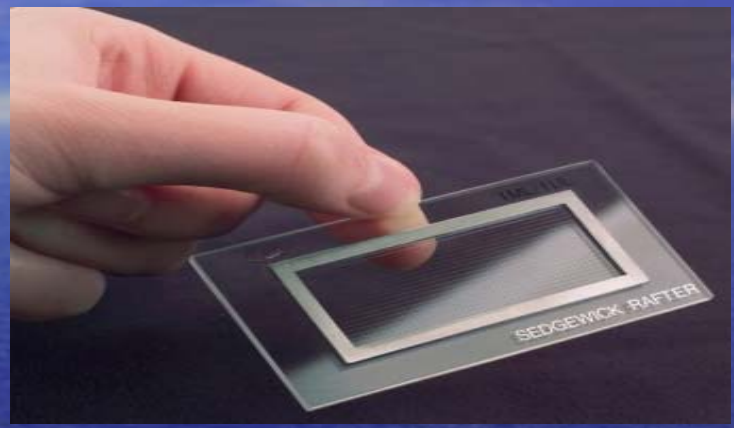
- Sampling
- Preservation
- Concentration
- Counting – Utermohl method
- Expressions of Populations
  - Cell abundance (cell/mL)
  - Why report Biomass
  - Examples

Algae Enumeration

- Time consuming
- Expensive
- Biomass often not reported
- Often done by untrained staff (picture keys)
- "Non" standard methods employed
- Nomenclature revisions
- Taxonomic literature often inadequate
  - Foreign language texts
  - European references often used

Counting Chambers

- Sedgewick-Rafter
  - Zooplankton
  - Limited to low mag.
  - Netplankton range



- Palmer Cells
  - Higher magnification
  - Low volume

Utermohl Method

- Inverted microscope method
- Introduced in 1930's
- Samples settled onto counting chamber
- Settling chamber = counting chamber
- High magnification (1400 x)
- Oligotrophic to eutrophic waters
- ** Widely accepted as most reliable

Utermohl (Inverted microscope) Method

Why measure biomass?

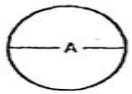
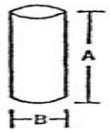
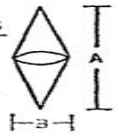
- Large range of cell size between spp.
  - Cell diameter may vary 1000 fold
  - Smallest (1 um) to 1000 um
- Cell volume 10^5 to 10^6 –fold
- Seasonal variation is cell size within spp.
- Potential toxin conc. related to biomass (dry matter)
- Conversion to dry weight, Chla, carbon
- Provides measure of standing crop

How is biovolume measured?

- Mean dimensions of cell determined
  - Measured with ocular micrometer
  - 25 measurements for common spp.
  - Cell shape is approximated to geometrical solids
  - Sphere, Cone, Cylinder (20+ formulae)

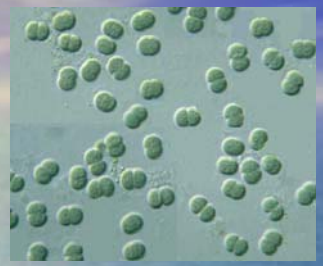
Cell Volume Formulae

APPENDIX D. BIOVOLUME FORMULAE

SHAPE	CODE	REPRESENTATIVE SPECIES	DIAGRAM	FORMULA	NO. MEAS
Sphere	Sp.	Sphaerocystis schroeteri		$\pi A^3/6$	1
Ellipsoid	EL	Scenedesmus bijuga Cryptomonas Euglena		$\pi AB^2/6$	2
Rod	Rd	Melosira granulata Cyclotella Asterionella		$\pi AB^2/4$	2
Two Cones	Cϕ	Ankistrodesmus falcatus		$\pi AB^2/12$	2
One Cone	CN	(horn of Ceratium)		$\pi AB^2/12$	2
Rectangular Box	RB			ABC	3

Cell abundance

Each unit counted as 1



Synechocystis

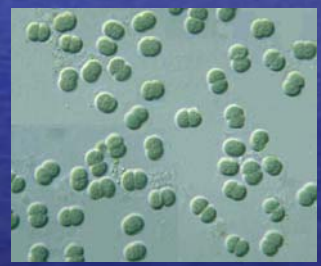
Microcystis

Planktothrix

Biovolume – example 1

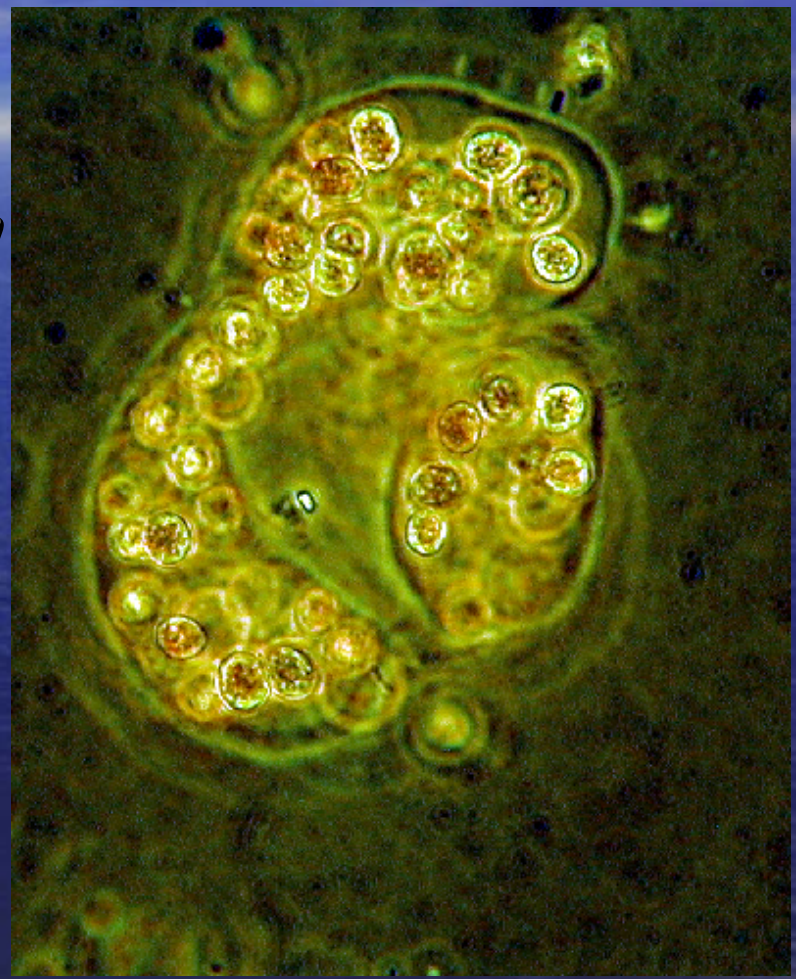
- *Synechococystis*
  - Cell shape: sphere
  - $V = (4r^3\pi)/3$
  - Unit = ind. cell
  - Cell diameter = 1 um

  - $V = 0.5\ um^3$ / cell



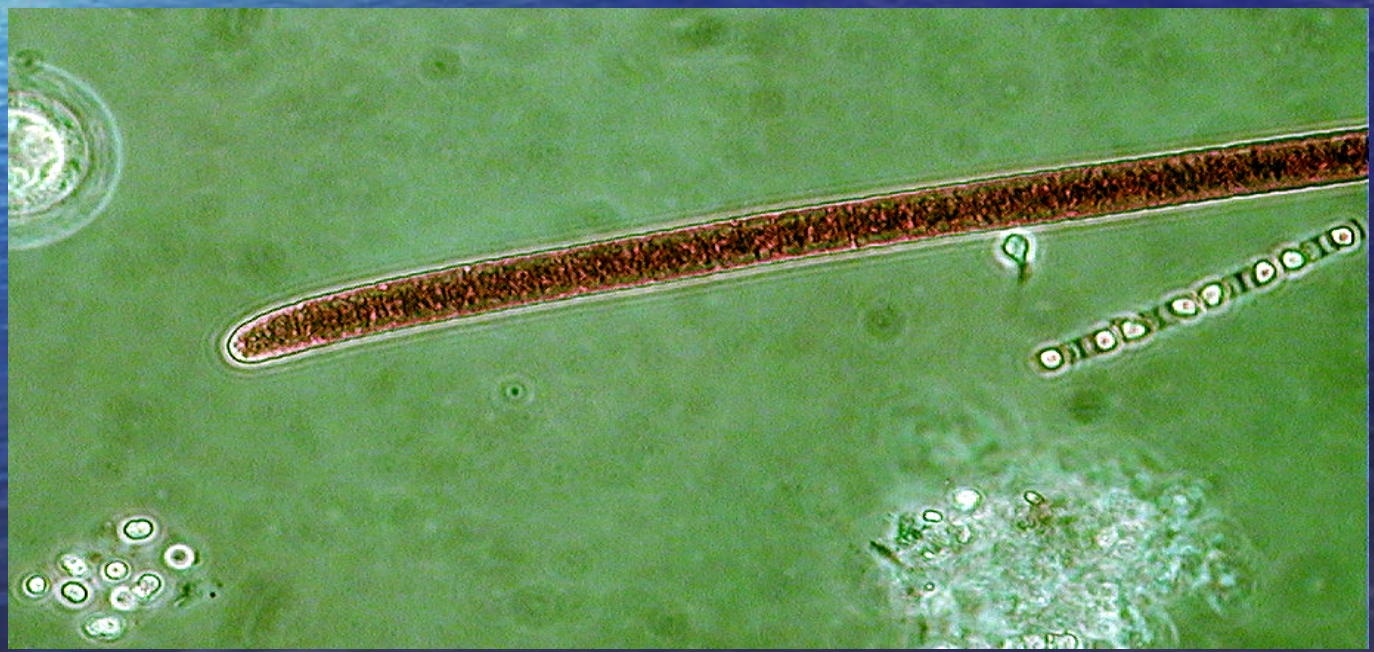
Biovolume – example 2

- *Microcystis aeruginosa*
  - Cell shape: sphere
  - $V = (4r^3\pi)/3$
  - Unit = ind. cell
  - Cell diameter = 5 um
  - $V = 65.5\ um^3$ / cell



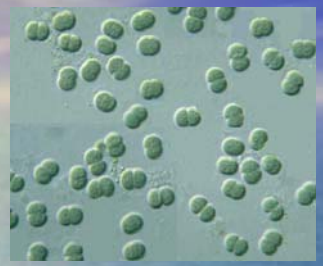
Biovolume – example 3

- *Planktothrix perornata*
  - Cell shape: cylinder ($r^2 \pi h$)
  - Cell diameter = 9 um; Length, h = 500 um
  - Unit = filament
  - V= 31,809 um^3



Cell volume

um³ / cell or filament



0.5 um3

65 um³

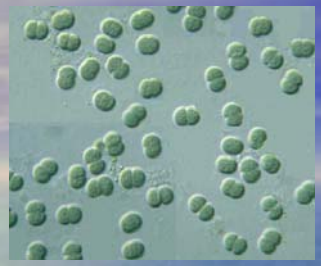
31,809 um³

From biovolume to biomass

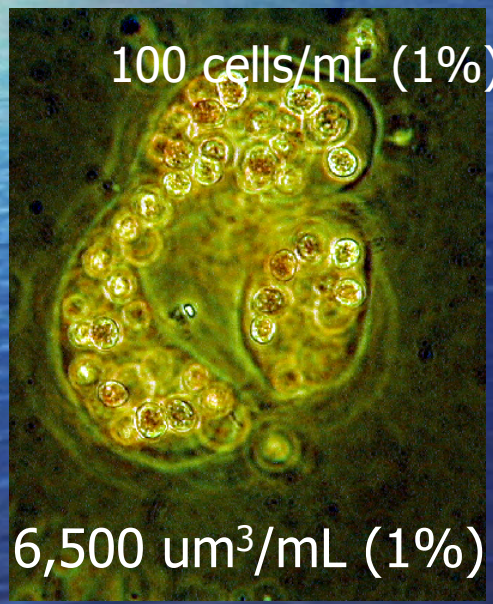
- Mean cell volume for each species multiplied by cell abundance = total volume for each species
- Specific weight of 1 mg /mm^3
- Biovolume units (mm^3/ml, um^3/mL) = biomass (mg/ml, ug/ml, mg/m^3)

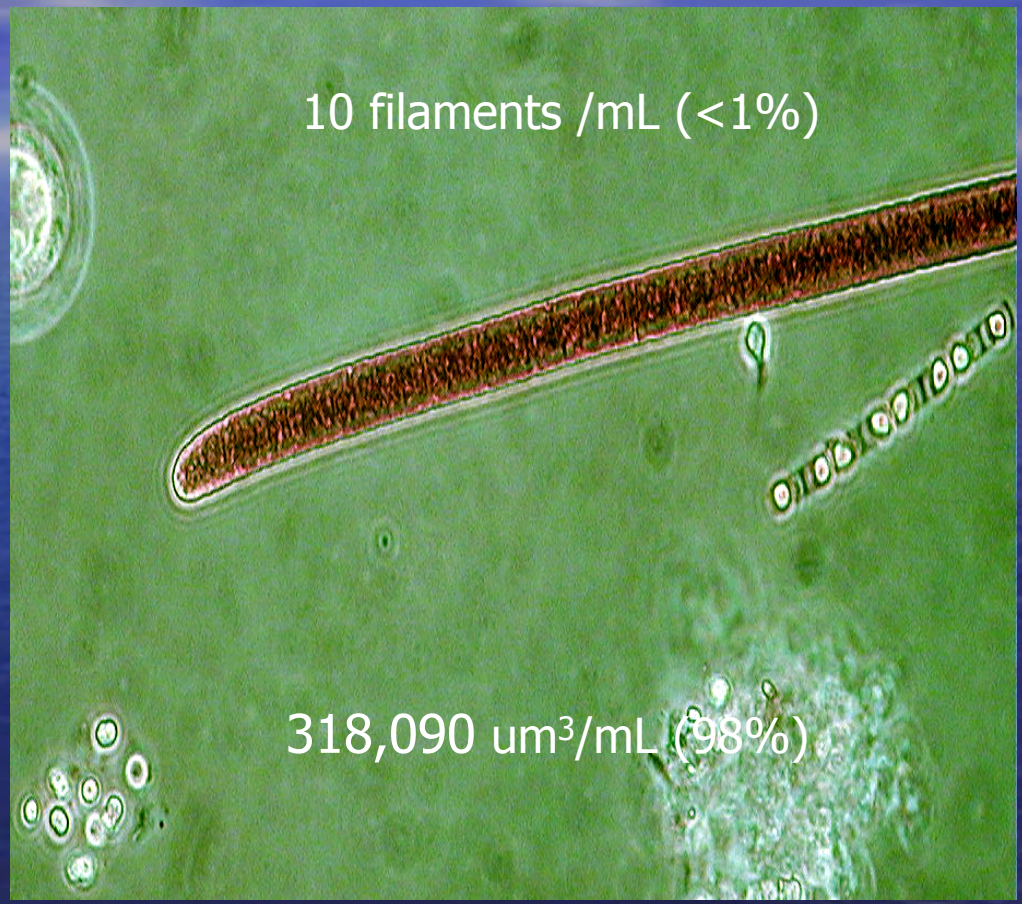
Biovolume

10,000 cells/mL (99%)



5,000 um^3/ml (1%)





Algal Enumeration Data Sheet

Phytoplankton data - xxxx --

Jeffrey Janik, Ph.D.

	ABUNDANCE		BIOMASS	
	Cells/mL	(%)	um^3/mL	(%)
Station: xxxxxxx-3				
Sample Date: 12/06/04				
Dictyosphaerium pulchellum	4500	0.1	45900	0.8
Monoraphidium contortum	500	0.0	1900	0.0
Monoraphidium komarkovae	2550	0.1	38250	0.7
Tetrastrum staurogeniaforme	2000	0.1	48800	0.8
Total CHLOROPHYTA	**9550**	**0.3**	**134850**	**2.3**
Amphiprora alata	147	0.0	146843	2.5
Nitzschia spp.	2000	0.1	114200	2.0
Total BACILLARIOPHYCEAE	**2147**	**0.1**	**261043**	**4.5**
Cylindrospermopsis raciborskii -st	1000	0.0	130000	2.2
Cylindrospermopsis raciborskii - st (h)	2550	0.1	184620	3.2
Aphanocapsa delicatissima	**2805000**	**81.7**	**1122000**	**19.4**
Aphanothece clathrata	255000	7.4	136000	2.4
Planktolyngbya subtilis	48450	1.4	1069130	18.5
Planktolyngbya contorta	7650	0.2	175185	3.0
Microcystis incerta	255000	7.4	51000	0.9
Planktothrix agardhii	220	0.0	314664	5.4
Pseudanabaena limnetica	**33150**	**1.0**	**1851980**	**32.0**
Pseudanabaena sp.	2550	0.1	287640	5.0
Total CYANOBACTERIA	**3410570**	**99.4**	**5322219**	**92.1**
Microflagellates 3-5 um	10200	0.3	63240	1.1
Total MICROFLAGELLATES	10200	0.3	63240	1.1
TOTAL	**3432467**	**100.0**	**5781352**	**100.0**

Example – Cells counts vs biomass

Comparison of two methods

- Spring, summer, fall sample
- Split sample analyzed by two labs
  - Lab A – Cell counts only
  - Lab B – Cell counts and biomass
- One algal species present– *Microcystis*

Algae Analysis Report – Lab A
cell abundance only

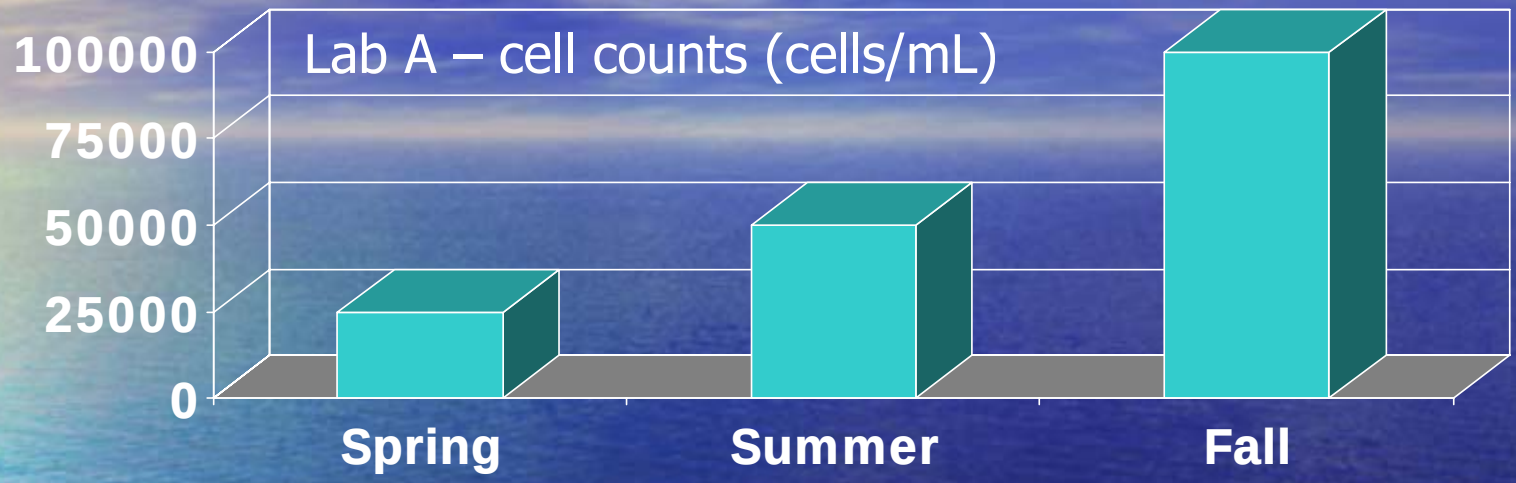
- Spring – 25,000 cells / ml
- Summer – 50,000 cells / ml
- Fall – 100,000 cells / ml

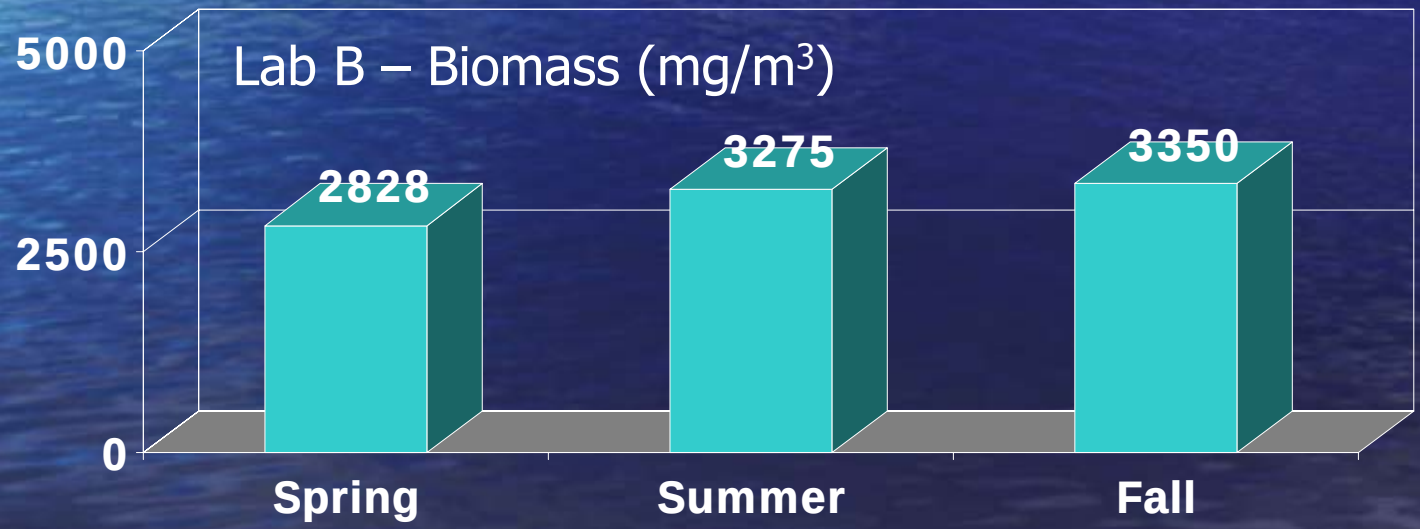
Algae Analysis Report – Lab B

Microcystis : cell diameter 4 – 6 um

- Spring – 25,000 cells / ml
  - *Cell diameter = 6 um, V = 113 um^3/cell*
- Summer – 50,000 cells / ml
  - *Cell diameter = 5 um, V = 65 um^3/cell*
- Fall – 100,000 cells / ml
  - *Cell diameter = 4 um, V = 33 um^3/cell*

Results


Lab A – cell counts (cells/mL)
100000
75000
50000
25000
0
Spring
Summer
Fall


Lab B – Biomass (mg/m3)
5000
2500
0
2828
3275
3350
Spring
Summer
Fall

Summary

- Important to use standard method
- Conducted by experienced phycologist
- Cell counts first stage
- Cell counts may over-emphasize small spp.
- Biomass provides invaluable information
- Small changes in mean cell size = large change in population biomass

Anabaena flos-aquae bloom, spring 2005