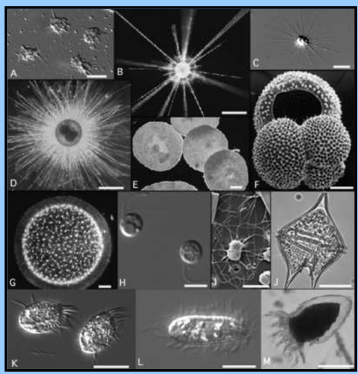
# The Ecology of Protists



Stefanie Moorthi

ICBM -Terramare, Planktology

## The Ecology of Protists

### Introduction

- distribution and nutritional modes
- => protists as primary producers
- => protists as consumers

### Concept of Microbial Loop

### Trophic Interactions

- competition
- consumption
- mixotrophy

### Seasonality in marine systems

### Harmful Algal Blooms

# Introduction

## High Abundance of Diverse Protists in Aquatic Habitats

Water Surface

Pelagial

Benthos

Interstitial

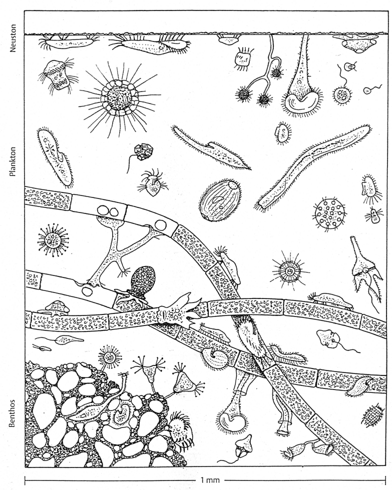



Fig. 301 Spatial distribution of unicellular organisms in aquatic habitats. Representatives of all groups of free-living protozoans live on different substrates (phase border air-water, benthos, detritus, aquatic organisms) or in the planctic community (after several authors).

Hausmann / Hülsmann 1996

## Factors influencing the distribution of protists

### Abiotic factors

chemical: concentrations of ions, pH, concentrations of dissolved gases (e.g. oxygen)

physical: temperature, light, water movement

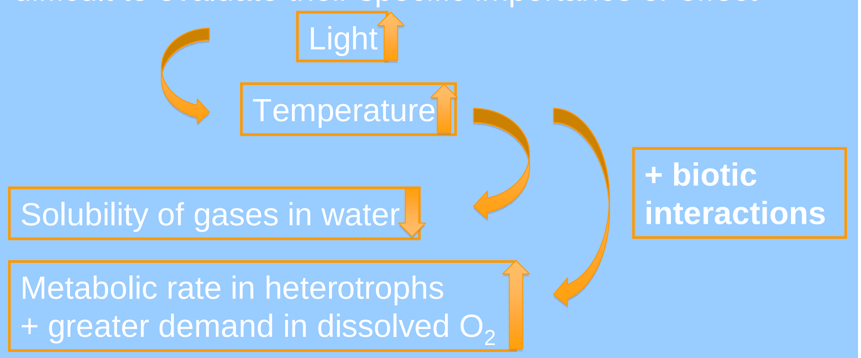
Protists are tolerant to wide range of physical and chemical environmental factors
⇒ found in a wide variety of biotopes and habitats

### Biotic factors

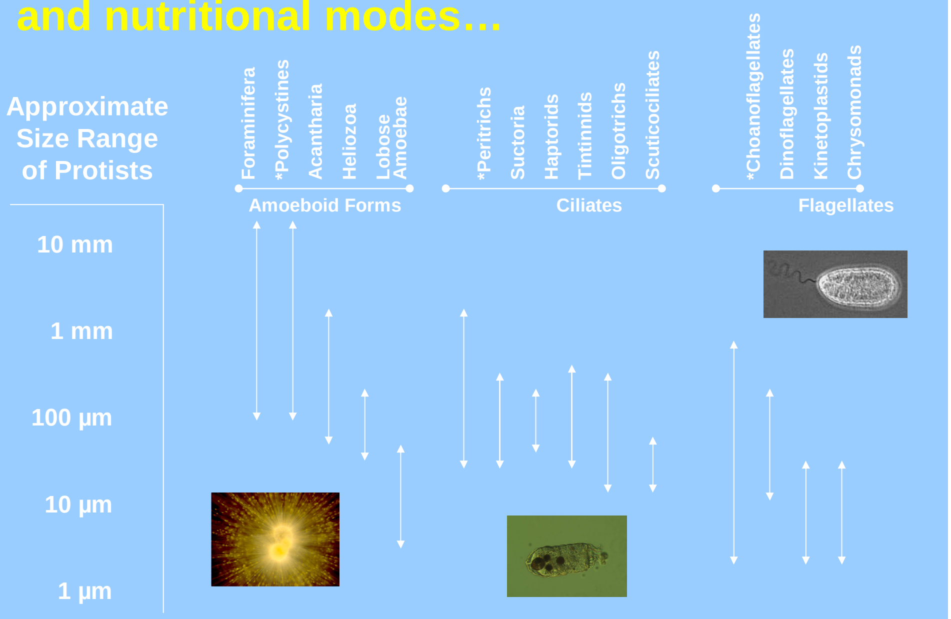
competition, predator-prey relationships

## Factors influencing the distribution of protists

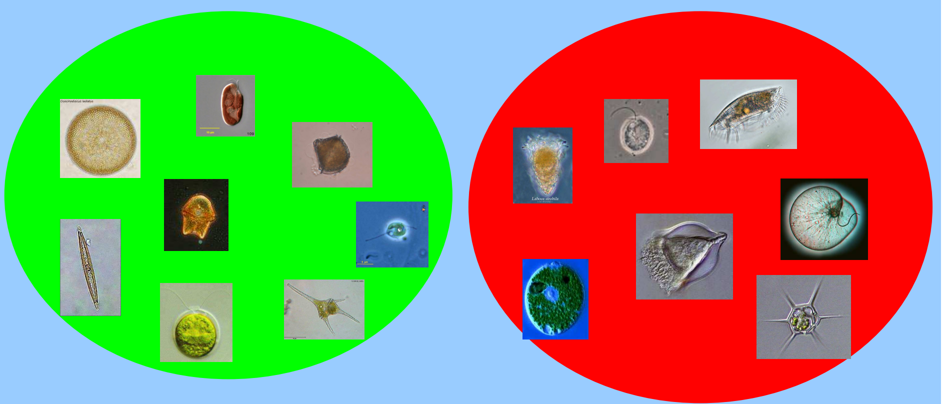
- not possible to isolate influences of individual factors from each other in nature
- act as a whole, abiotic factors often in conjunction with biotic factors
- difficult to evaluate their specific importance or effect



## Protists have many different sizes, shapes and nutritional modes…




## Nutritional modes

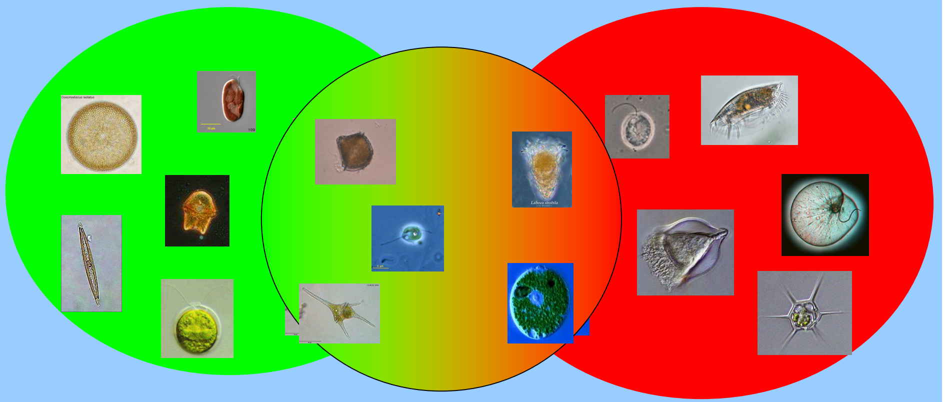


**phototrophic**

**heterotrophic:**
phagotrophic/osmotrophic

➡ Protists play a substantial role as primary producers and consumers in aquatic food webs

## Nutritional modes



**phototrophic**

**mixotrophic osmotrophic**

**heterotrophic:** phagotrophic/osmotrophic

➡ Protists play a substantial role as primary producers and consumers in aquatic food webs

## Resources

**<u>Resources</u>**

- Light
- C, O, H
- Macronutrients
  - N, P, Si, S, Na, Cl, K, Ca, Mg
- Micronutrients
  - Fe, Mn, Cu, Zn, B, Mo, V, Co

**<u>Resources</u>**

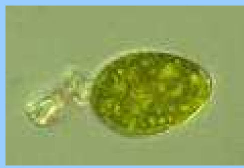
- organic compounds
- (prey items)
- oxygen

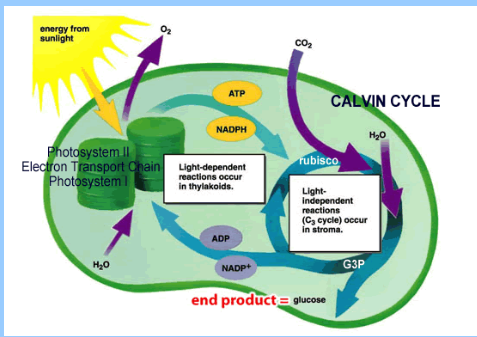
# Protists as phototrophic primary producers

Photosynthesis

$6\ CO_2 + 6\ H_2O + 2802kJ$

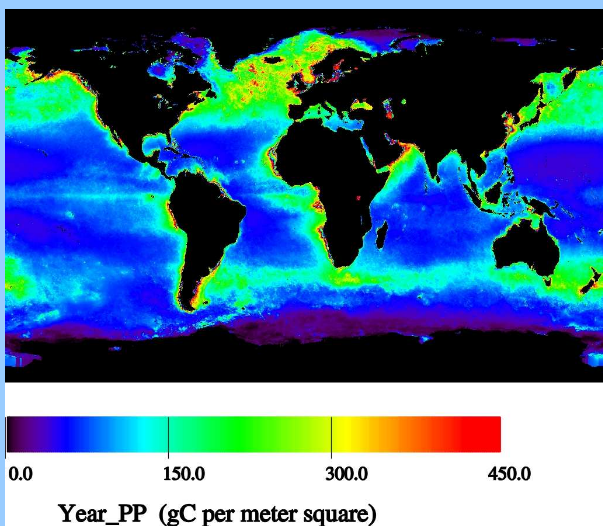
$\Rightarrow C_6H_{12}O_6 + 6\ O_2$






# Spatial distribution of ocean primary production

- high along the coast and in upwelling regions
- low in the Southern Ocean (Fe-limitation?) and in downwelling regions



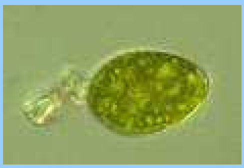
## Algal primary production

Like terrestrial plants algae use atmosphaeric $CO_2$ and light for growth and reproduction. By doing so they produce the oxygen that we breathe.

Photosynthesis

$6\ CO_2 + 6\ H_2O + 2802kJ$

$\Rightarrow C_6H_{12}O_6 + 6\ O_2$

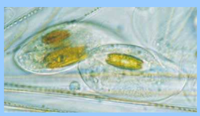


- total $CO^2$ uptake by plants: 104.9 giga tons per year
  - 1 Gt = 1.000.000.000 t
- 48.5 Gt/year of that by algae
  - ~ 47%
  - ~ every 2. oxygen molecule is produced by algae

## Protists as consumers can be…

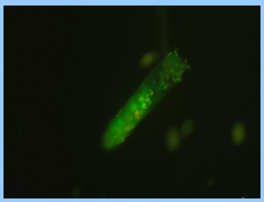
…bacterivorous

...herbivorous



...carnivorous

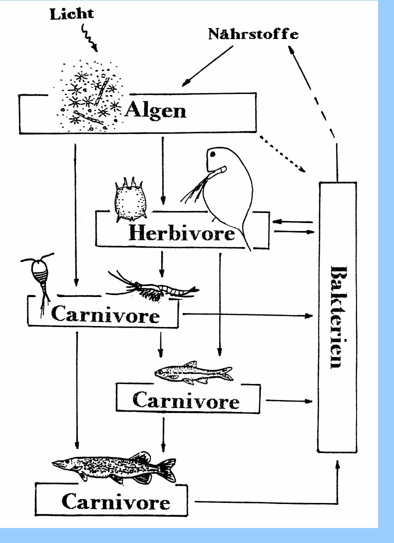
...omnivorous



## However, role of heterotrophic protists has been severely underestimated until the 1970ies…

**Classical Planktonic food web**

- Actual role of **bacteria?**
- Actual role of **primary production?**
- Role of heterotrophic **protists?**




## Concept of the Microbial Loop

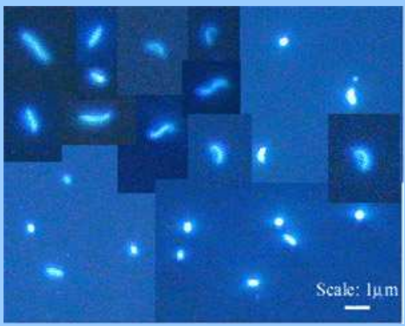
# Key findings leading to the concept of the microbial loop
## 1. Bacterial abundances

- Direct bacterial counts: Abundances are higher and more constant as assumed before
- instead of ca. $10^3$ - $10^4$ => $10^6$ $ml^{-1}$

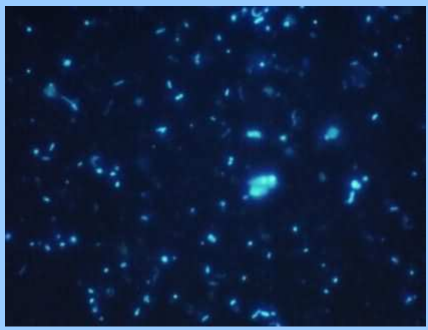
- Bacterial abundances are correlated with Chlorophyll concentrations
- Phytoplankton releases a major part of its photosynthesis products in form of dissolved exudates
- Bacteria take up 50 - 100% of this DOC (= conversion of DOC to POC)
- Nanoflagellates are very abundant and are able to effectively graze on bacteria
- Nanoflagellate-abundances are correlated with bacterial abundances

# New Method: Epifluorescense microscopy

Fluorescent stain and excitation with UV-filter:
Visualization of DNA & RNA,
and therewith of bacteria and eukaryotic nuclei




www.jochemnet



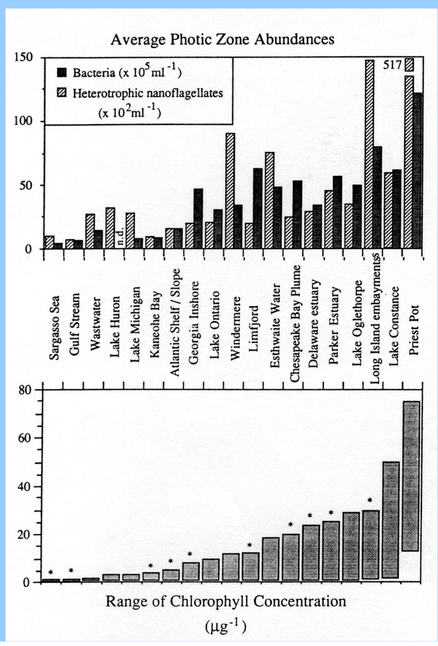
www.soest.hawaii.edu

# Key findings leading to the concept of the microbial loop
## 2. Bacterial nutrition (bottom-up)

- Direct bacterial counts: Abundances are higher and more constant as assumed before
- instead of ca. $10^3$ - $10^4$ => $10^6$ $ml^{-1}$
- ➢ Bacterial abundances are correlated with Chlorophyll concentrations
- ➢ Phytoplankton releases a major part of photosynthesis products in form of dissolved exudates
- ➢ Bacteria take up 50 - 100% of this DOC (= conversion of DOC to POC)
- Nanoflagellates are very abundant and are able to effectively graze on bacteria
- Nanoflagellate-abundances are correlated with bacterial abundances

# Correlation Chl. *a* - bacteria

oligotrophic




eutrophic

Sanders, Caron & Berninger 1992

# Key findings leading to the concept of the microbial loop
## 3. Fate of bacterial biomass (top-down)

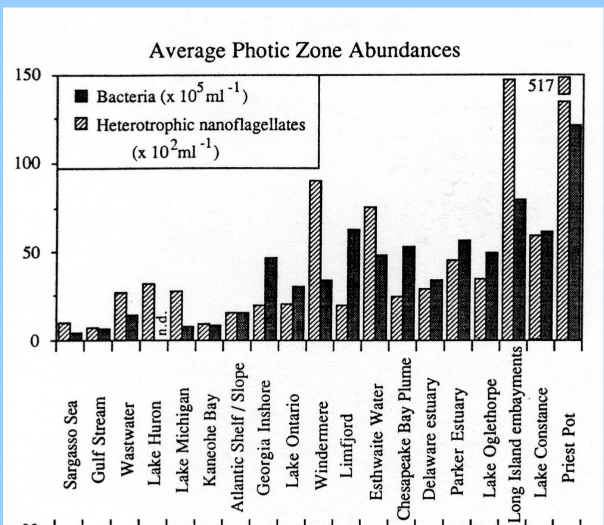
- Direct bacterial counts: Abundances are higher and more constant as assumed
- instead of ca. $10^3$ - $10^4$ => $10^6$ $ml^{-1}$
- Bacterial abundances are correlated with Chlorophyll concentrations
- Phytoplankton releases a major part of its photosynthesis products in form of dissolved exudates
- Bacteria take up 50 - 100% of this DOC (= conversion of DOC to POC)

➢ Nanoflagellates are very abundant and are able to effectively graze on bacteria

➢ Nanoflagellate-abundances are correlated with bacterial abundances

# Correlation bacteria/flagellates

oligotrophic




eutrophic

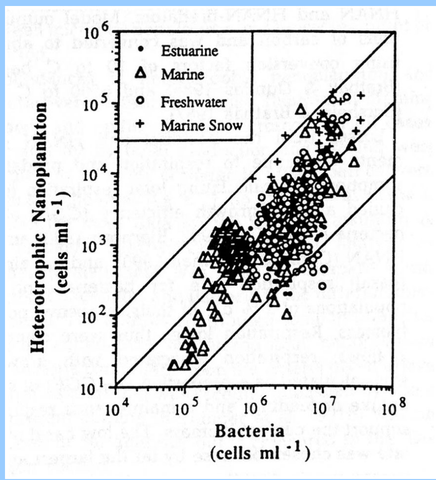
Sanders, Caron & Berninger 1992

## Bacteria / Flagellates

equally:

bacterivory

**1:1**




bacterial biomass production

Sanders et al. 1992

## The microbial food web

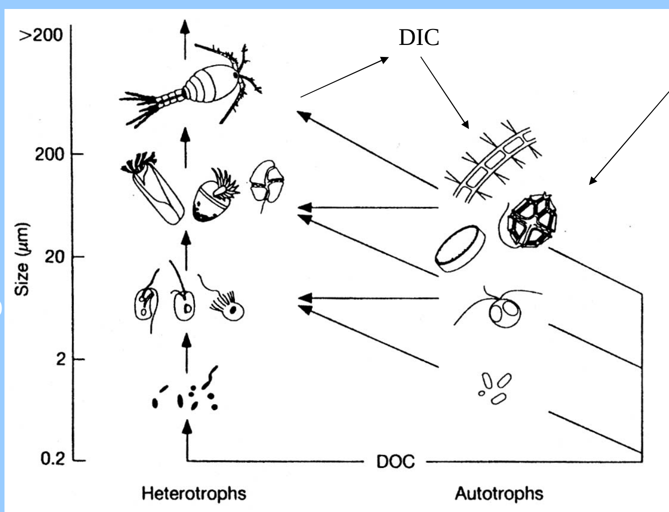
Meso

Micro

Nano

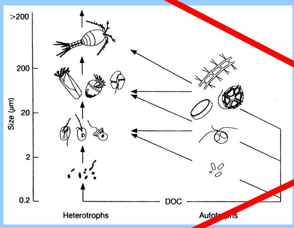
Pico

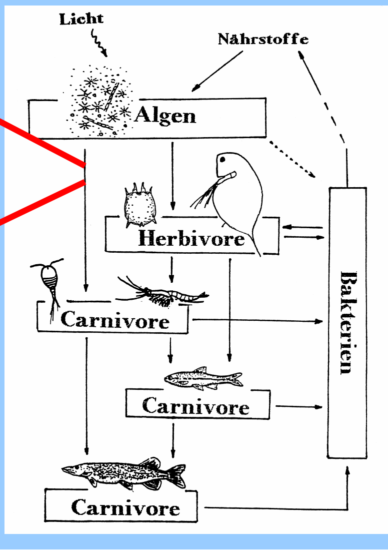
light



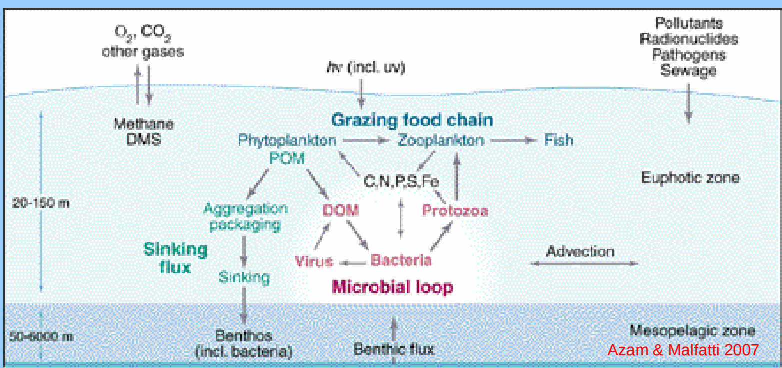

Fenchel 1982, Azam et al. 1983

## Pelagic food web:
## Linking the microbial with the classical food web




## Pelagic food web: Linking the microbial with the classical food web




Relative role of the microbial loop differs in time and space

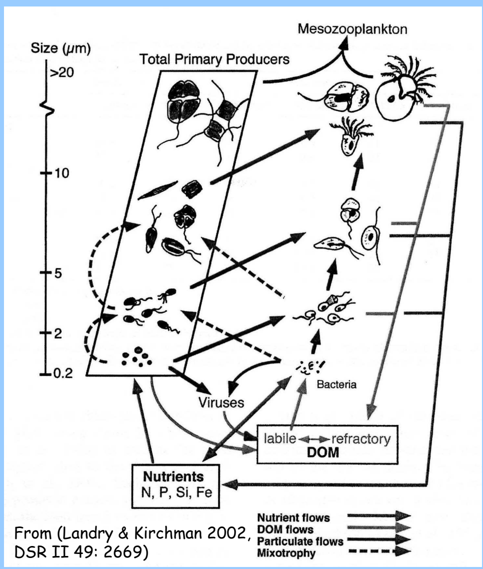
- microbial loop dominates oligotrophic waters whereas classical food chain predominates when mineral nutrients are not limiting (during spring bloom in temperate waters, in upwelling areas)
- competition for dissolved mineral nutrients favors small organisms, primary production is then mainly based on nutrients regenerated in the water column

## Summary: Microbial Loop

- Phytoplankton releases photosynthesis products as dissolved excudates
  - bacteria take up 50-100% of DOC (conversion to POC)
  - bacterial biomass is consumed and thus re-enters food web
- Microbial loop dominates in oligotrophic waters whereas the classical food chain predominates eutrophic systems

## Trophic interactions in microbial food webs

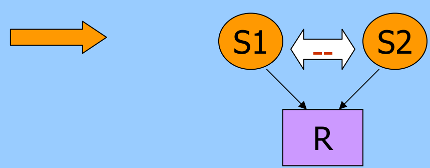
# Trophic interactions in microbial food webs




- competition
- consumption
  - bacterivory
  - herbivory
  - carnivory
  - parasitism
- mixotrophy

# Competition

- exploitative competition – indirect interaction
  - competition for resources (consumable environmental factors such as light, nutrients or prey)

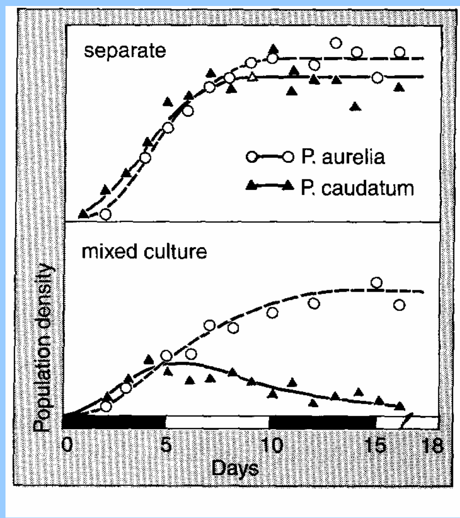



- Interference competition – direct interaction
  - Allelopathy (production of secondary metabolites affecting growth and development of other organisms)
  - Toxic algae

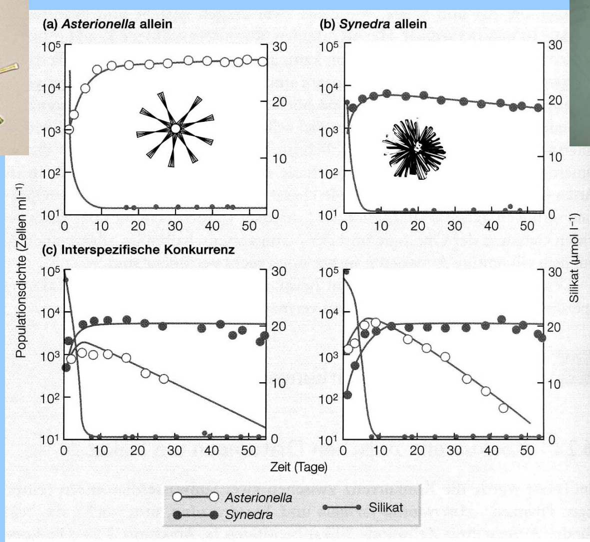
# Competition

- Two species competing for the same resource do not coexist at equilibrium
- Competitive exclusion principle

Gause 1934

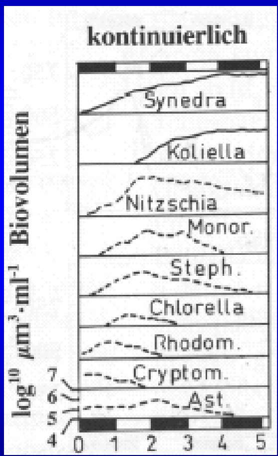


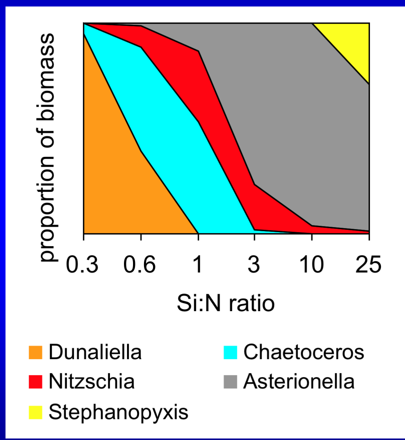

# Diatoms: limiting factor: silicate




Sommer 2005

# Competition for 2 resources




modified from Sommer '96

Sommer 1985: P and Si limiting
2 species survive

# Preventing competition

- Predictions from Tilman's model:

  In well-mixed communities at equilibrium, the number of coexisting species is equal or lower than the number of limiting resources

- The observed diversity is much higher, even in well-mixed communities with a small number of limiting resources

=> Why are there so many species?

=> **Paradox of the Plankton**

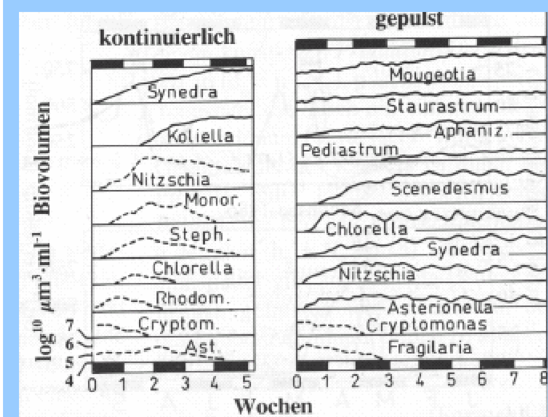
(Hutchinson 1961)

## Preventing competition

There have to processes preventing competitive exclusion

- temporal heterogeneity
- spatial heterogeneity
- disturbance

## Preventing competition




Stable

Oscillating

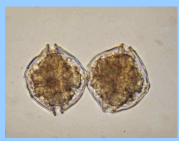
Decreasing

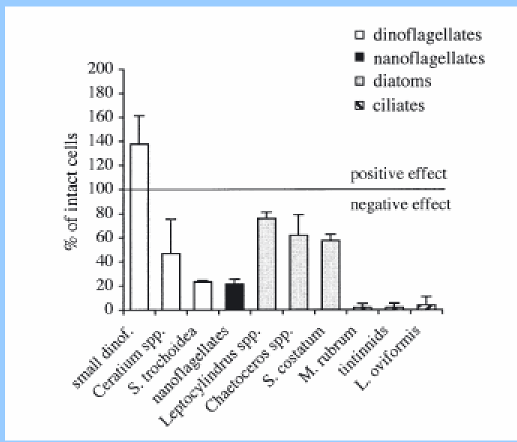
Pulsing resources increases the number of coexisting species

Temporal heterogeneity prevents competitive exclusion

## Interference competition

Allelopathy in *Alexandrium tamarense*






Fistarol et al. 2004

- *A. tamarense* affected whole plankton community by decreasing growth rates in most species and changing community structure
- different sensitivities of target species =>more resistant species may benefit from allelochemicals

## Consumption

- Consumption: Prey is consumed by consumer
  - bacterivores
  - herbivores
  - carnivores
  - omnivores

→ consumers influence the abundance and distribution of their prey and vice versa

- Consumers excrete or egest nutrients and therefore have positive effects on algal growth
- Consumers have comparably low plasticity in nutrient content and excrete nutrients which are not in short supply

## Bacterivory

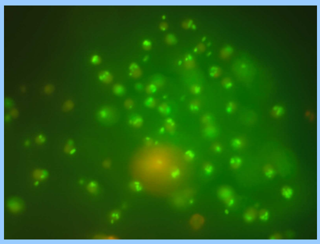
- **marine planktonic flagellate assemblage may graze 25 to >100% of daily production of bacterioplankton**
  - mismatch "less grazing than production" can be explained by...

    ...other types of grazers (mixotrophic phytoflagellates, ciliates)

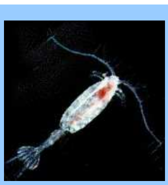
    ...bacterivores selecting larger, growing and dividing cells thus directly cropping bacterial production

    ...lack of methods to accurately measure protistan bacterivory

    ...bacterial mortality due to viral infection



## Herbivory



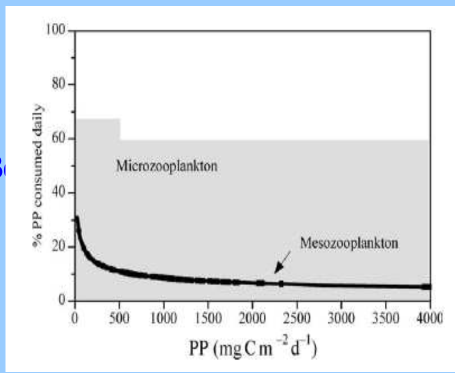
- phytoplankton can be effectively grazed by metazoans such as crustaceans, appendicularians and cnidaria
- **however**, much of carbon production in marine pelagial accomplished by cells <5µm (prasinophytes, prymnesionphytes, diatoms, coccoid cyanobacteria, prochlorophytes)

  => grazed by ciliates (tintinnids and aloricate choreotrichs), heterotrophic dinoflagellates and het. nanoflagellates (2 - 20µm, usually considered as typical bacterivores)

# Herbivores: protistan zooplankton

Calbet 2008: Schematic approximation to the global mean grazing impact on autotrophic production

- Microzooplankton (grazers <200µm) are key components of marine food webs
- Diverse groups play distinct roles in ecosystems
- Ciliates are important, but also other groups often ignored and poorly sampled => heterotr. and mixotr. small flagellates and dinoflagellates, radiolaria, foraminifera (+ metazoan microzooplankton such as rotifera, meroplanktonic larvae and copepod nauplii)




Percentage of phytoplankton primary production (PP, mg C m$^{-2}$ d$^{-1}$) consumed daily by microzooplankton (shaded area) and mesozooplankton (line) as a function of autotrophic production (mg C m$^{-2}$ d$^{-1}$). Data from Calbet (2001) and Calbet and Landry (2004).

# Herbivores: metazooplankton

# Herbivores: metazooplankton

Meroplankton

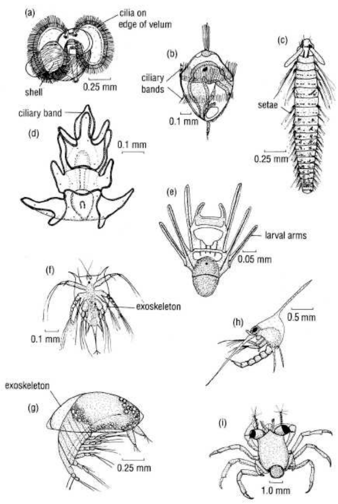



Figure 4.11 Meroplanktonic larvae of benthic invertebrates. (a) snail veliger; (b) polychaete trochophore; (c) late larva of a polychaete; (d) bipinnaria of a starfish; (e) echinopluteus of a sea urchin; (f) barnacle nauplius; (g) barnacle cypris; (h) crab zoea; (i) crab megalopa.

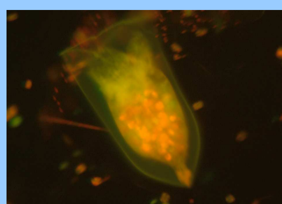
Lalli & Parsons 1995

# Herbivores: protistan zooplankton


Bo

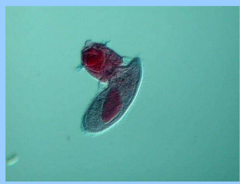
Moorthi


Sherr & Sherr



## Protists as consumers can be…

**…voracious predators**



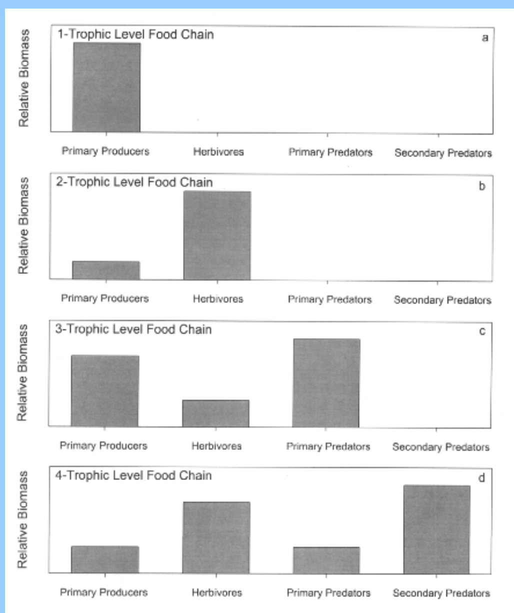
*Didinium nasutum*

*Didinium* is able to expand its cytostome (mouth) to such an extent that in can engulf an entire *Paramecium*

www.microscopy-uk.org.uk

## Trophic cascades

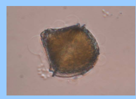
- alternating effects of regulating forces among trophic levels
- indirect interactions in natural communities are important
- predators can have positive or negative effects on primary producers, depending on food web configuration



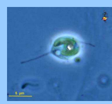

Holz et al., Association for Biology Laboratory Education (ABLE)
http://www.zoo.utoronto.ca/able

# Mixotrophic Protists

- Phototrophic and heterotrophic nutrition (most often phagotrophic)
- different types of mixotrophy:

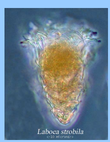


➢ phagotrophic algae that are primarily phototrophic



➢ photosynthetic protozoa that are primarily phagotrophic

➢ In many cases, like in ciliates, freshwater heliozoa or benthic marine foraminifera, photosynthetic protozoa are photosynthetic due to the presence of algal endosymbionts or due to sequestering and utilizing ingested chloroplasts (chloroplast retention)




# Mixotrophic Protists

**Mixotrophy**

photosynthesis → primary production

phagotrophy → consumption of particulate organic matter

↓

Support of growth in the dark

Supplementation of photosynthetic carbon fixation

Aquisition of nutrients such as N and P, of vitamins, essential fatty acids and iron

# Mixotrophic Protists

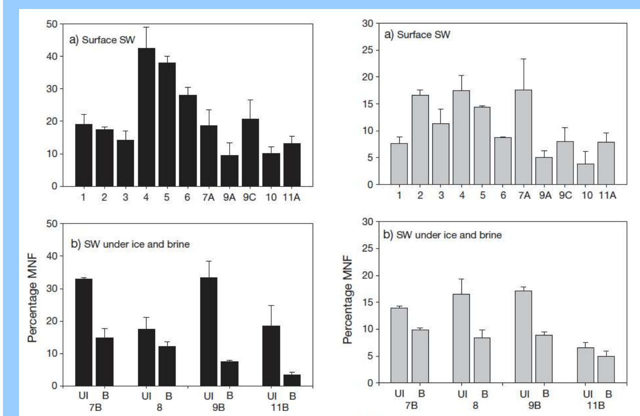
**Autotrophy** ⟷ **Heterotrophy**

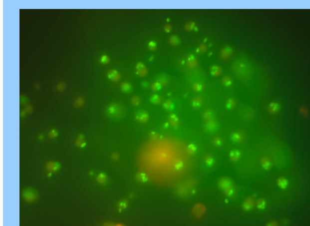
↑ ↑

light, organic and inorganic nutrients,
prey abundances

variabel on temporal and spatial scales, but can contribute major portions to the phototrophic and heterotrophic nanoplankton

# Mixotrophic flagellates in plankton and sea ice in the Ross Sea, Antarctica



Moorthi et al., AME 2009

Mixotrophs contributed up to 50% to the total bacterivorous nanoplankton in seawater and up to 20% in brine (sea-ice), while they contributed up to 20% to the phytoflagellates in seawater and up to 10% in brine (sea-ice)

=> mixotrophy seems to be an important nutrituinal strategy in this habitat

## High bacterivory by the smallest phytoplankton in the North Atlantic Ocean

Zubkov & Taran 2008, Nature 2008

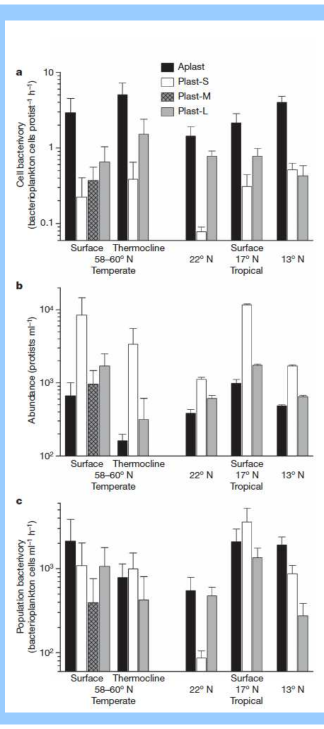
Planktonic algae >5µm are major fixers of inorganic carbon in the ocean

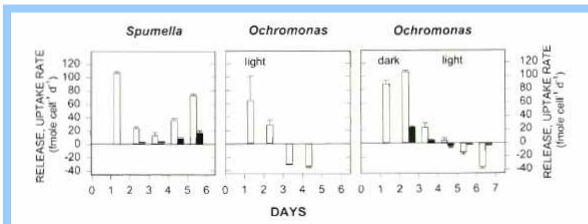
dominate phytoplankton biomass in post-bloom, stratified oceanic temperate waters

⇒ In this study these small algae carried out 40-95% of the bacterivory in the euphotic layer of the temperate North Atlantic Ocean in summer (37-70% in surface waters of the tropical North-East Atlantic Ocean

⇒ Global significance of mixotrophy

⇒ Smallest algae obtain ¼ of their biomass from bacterivory




## Nutrient regeneration

**Rothaupt 1997:**

Phagotroph released nutrients when feeding on bacteria and stimulated growth of algae

Mixotroph released nutrients in the dark or at high bac. densities in the light, when phagotrophic nutrition prevailed; but taking up nutrients when growing phototrophically

=> no phytoplankton stimulation

basic differences in the patterns of nutrient turnover by mixotrophs and phagotrophs!

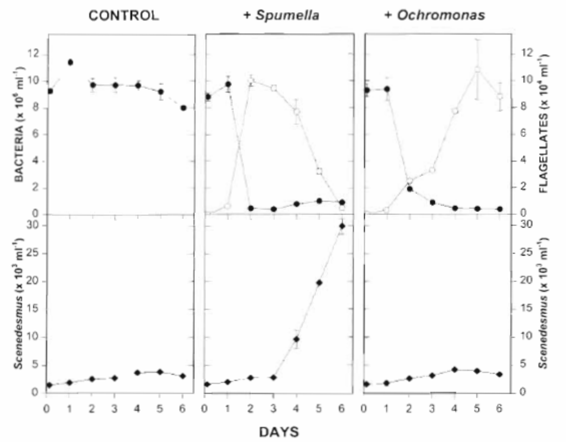



Fig. 3. Cell counts of bacteria (●), flagellates (○) and *Scenedesmus acutus* (◆) in the first phytoplankton stimulation experiment (batch culture, 16:8 h light:dark rhythm)

## Summary: Trophic interactions

<u>Competition:</u>

- exploitative + interference competition
- competitive exclusion principle
- Paradox of the plankton

Processes preventing competitive exclusion

- temporal heterogeneity
- spatial heterogeneity
- disturbance

## Summary: Trophic interactions

<u>Consumption:</u>

Bacterivory: heterotrophic + mixotrophic flagellates, ciliates

Herbivory: metazoan and protistan grazers => special role of microzooplankton (e.g. ciliates, heterotrophic and mixotrophic flagellates and dinoflagellates, radiolaria, foraminifera + metazoans)

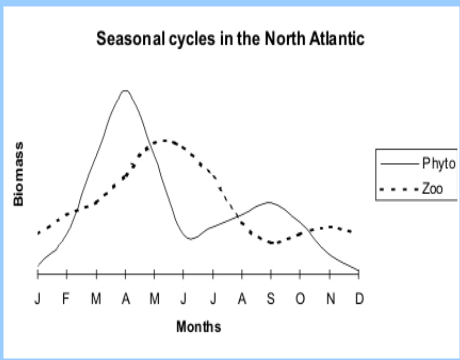
<u>Mixotrophic protists</u>: phagotrophy + phototrophy

- advantages for growth in dark and under low-nutrient conditions
- variable contributions on temporal and spacial scales
- can play a major role as bacterivores in polar, temperate and tropical marine ecosystems
- influenced by abiotic (e.g. light, nutrients) and biotic (prey abundances, presence of phototrophic or heterotrophic competitors) factors
- can have major impact on carbon fixation, nutrient dynamics and control of prey (bacteria, algae, heterotrophs)

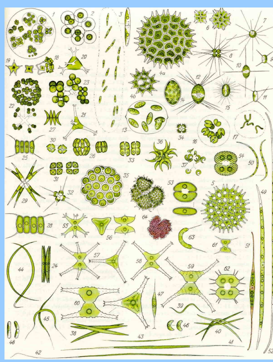
# Seasonality in marine plankton

## Herbivore-prey oscillations

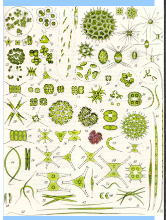
- Seasonality of phytoplankton

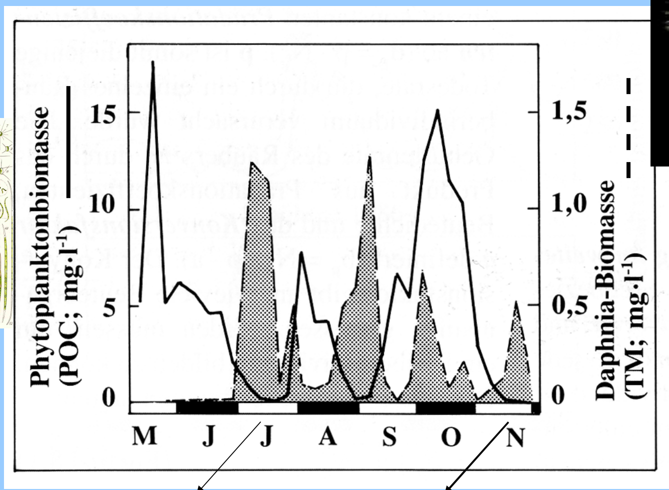



} -plankton



## Herbivore-prey oscillations
## Phytoplankton - *Daphnia*






clear water state (grazing)

Physical. causes (sinking, light limitation)

Sommer 1994

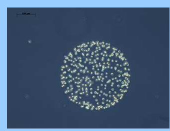
But sometimes consumers are not able to control phytoplankton blooms...

## Selective feeding of herbivorous zooplankton

Inedible or actively avoided species experience lower mortality than well edible species.

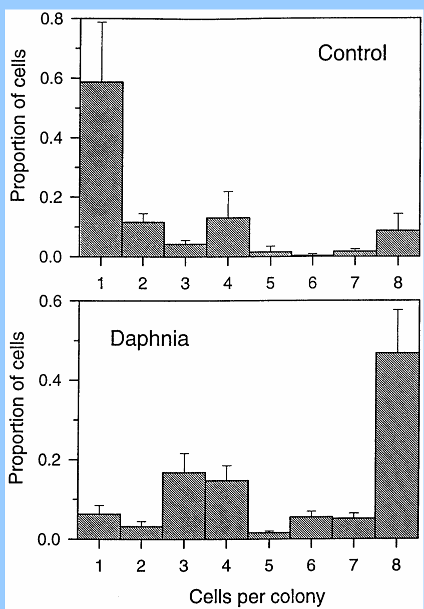
How to get feeding resistant:

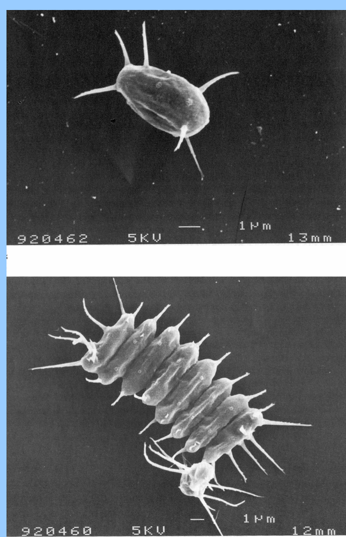
- size
  - big single cells
  - colony formation
- forming of appendages
- mucus production
- indigestibility
- chem. intolerance / toxicity



➡ Selectivity depends on species, size and feeding mode of consumer

## Morphological Defense Mechanisms of Algae




Hessen DO, Van Donk E (1993) Arch Hydrobiol 127:129-140

## Morphological Defense Mechanisms of Algae

*Phaeocystis* sp.: Beach on Spiekeroog after storm event (17.05.07)

Some algae such as dinoflagellates form "red tides" (massive algal blooms) under certain environmental conditions

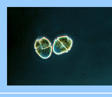
=> some of these blooms can be toxic

Red Tide off the coast of La Jolla, California

# Algal Toxins

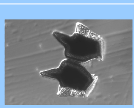
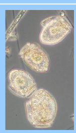
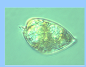
**Amnesic Shellfish Poisoning (ASP)**:
domoic acid, diatom *Pseudonitzschia*





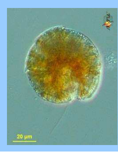
**Paralytic Shellfish Poisoning (PSP)**:
Saxitoxin, dinoflagellates such as
*Alexandrium, Gymnodinium and Pyrodinium*

**Diarrhetic Shellfish Poisoing (DSP)**: Okadeic acid,
dinoflagellates such as *Prorocentrum and Dinophysis*

**Neurotoxic Shellfish Poisoning (NSP)**:
Brevetoxin, dinoflagellate *Karenia brevis*

**Ciguatera Fish Poisoning (CFP)**: after eating
poisoned fish, Cigatoxin, Gambiertoxin,
dinoflagellate *Gambierdiscus toxicus*



# Why?

## Allelopathy

a way to outcompete other algal species. Nutrient ratios affect toxin concentrations

## Grazer deterrence

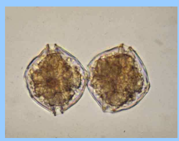
avoid being eaten

**BUT** for most substances not fully understood yet!!

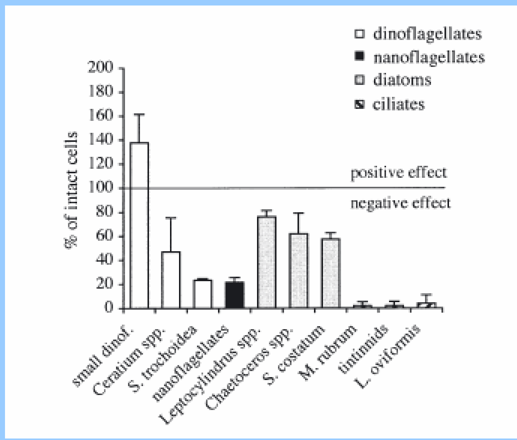
metabolic products stored in the cells for other reasons, toxicity not directed at competitors or consumers

# Interference competition

Allelopathy in *Alexandrium tamarense*



- *A. tamarense* affected whole plankton community by decreasing growth rates in most species and changing community structure
- different sensitivities of target species =>more resistant species may benefit from allelochemicals



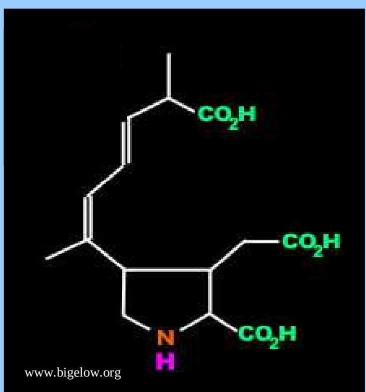

Fistarol et al. 2004

# *Pseudo-nitzschia* spp

**Domoic Acid**

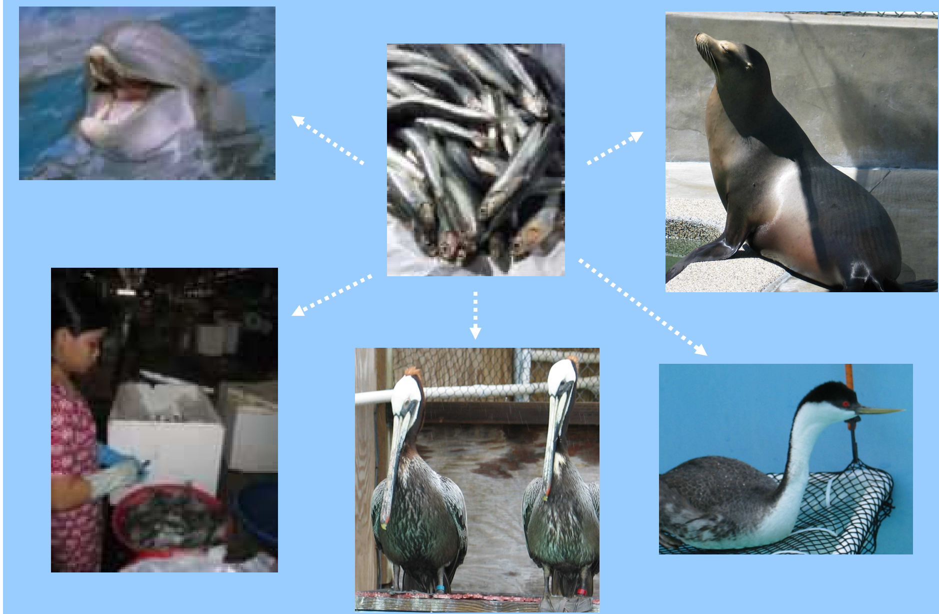
Amnesic Shellfish Poisoning (ASP)

> 1500 mammal strandings in 2003

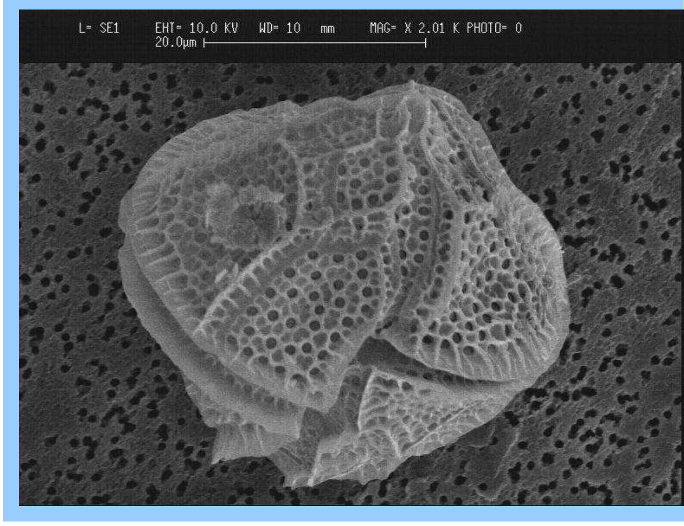


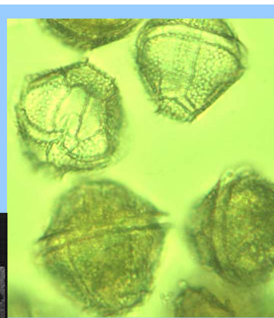

**Symptoms**:
Nausea, vomiting, abdominal cramps, headache, dizziness, confusion, disorientation, short term memory loss, motor weakness, seizures, cardiac arrhythmia, coma, possibly death

## Domoic Acid Poisoning



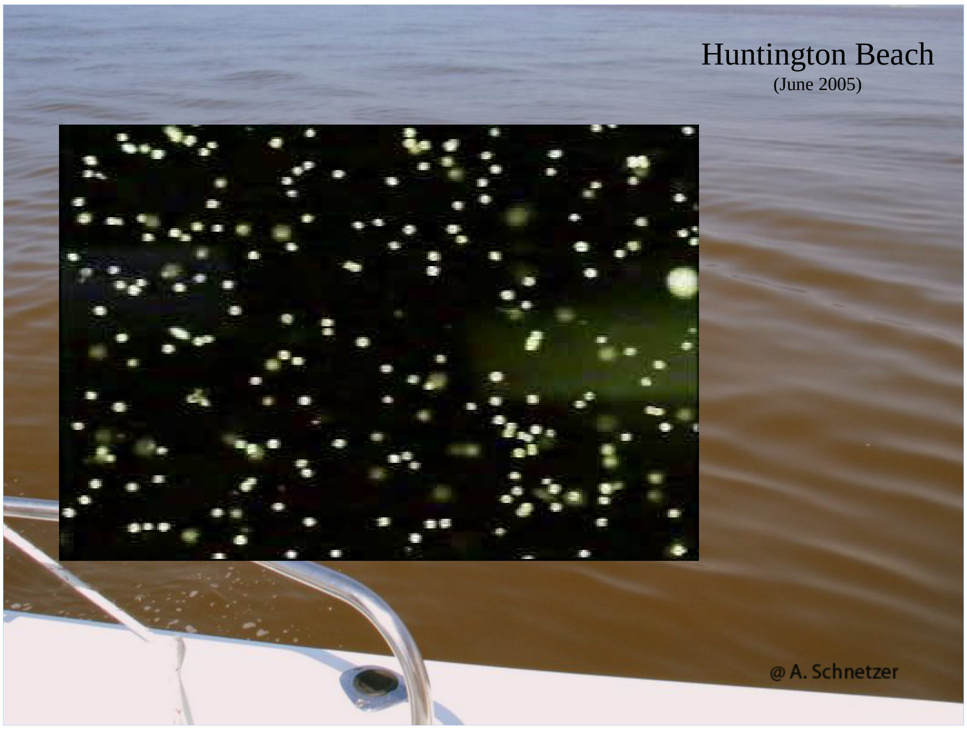
## *Lingulodinium polyedrum*



A ‘red tide’ without toxic consequences…
…so far.
(potential producer of yessotoxin)


Huntington Beach
(June 2005)
@ A. Schnetzer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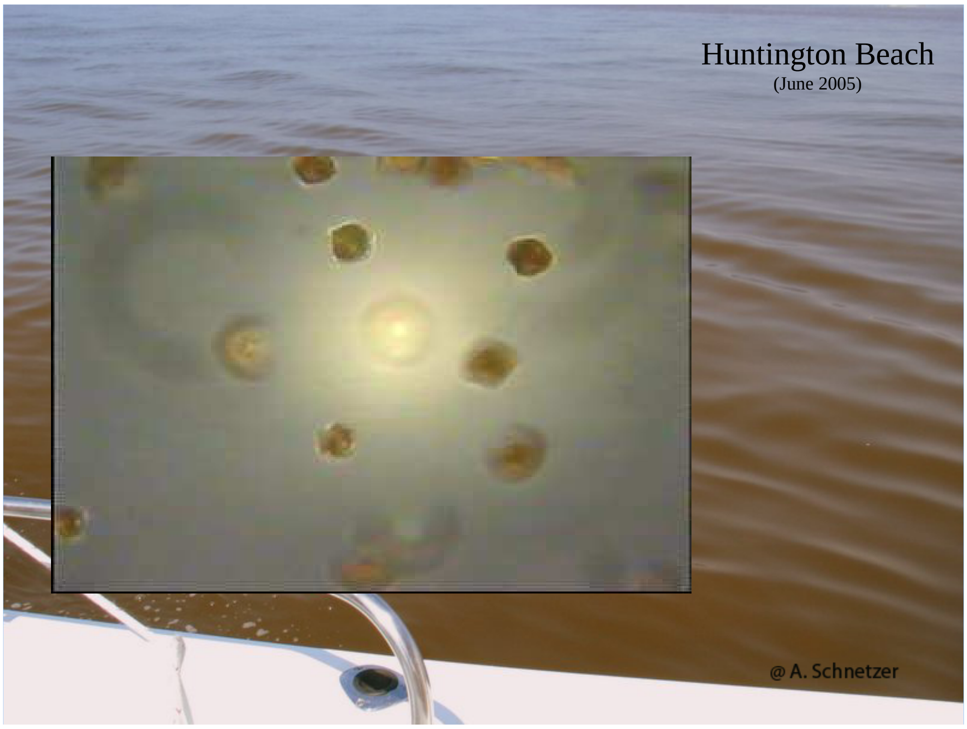
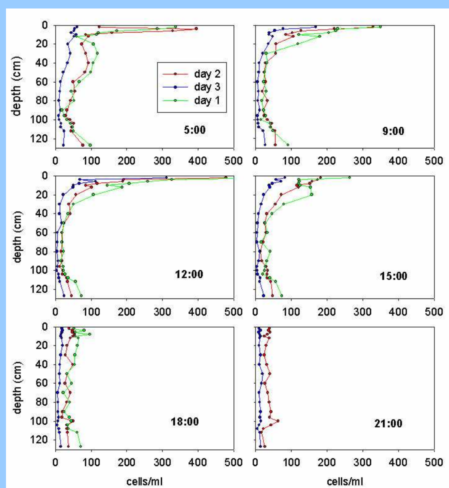



# Vertical migration behavior of *L. polyedrum* in a laboratory observation

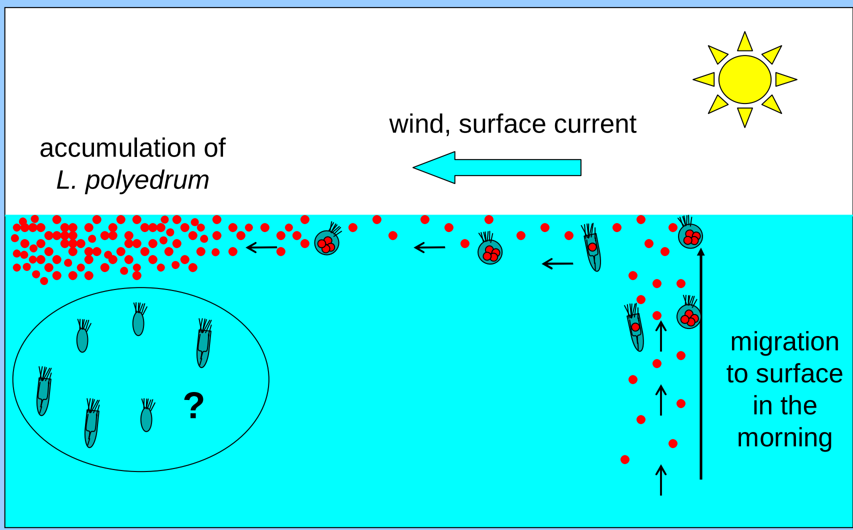
lights on: 6am

lights off: 5pm




Moorthi et al. Microb Ecol 2006

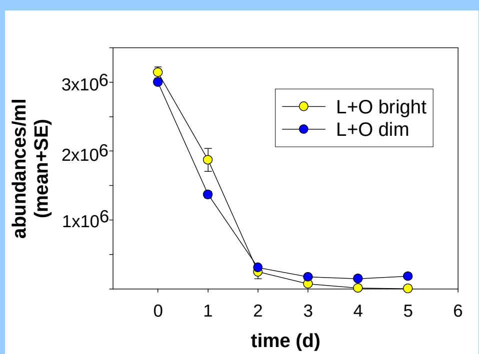
## Hypothesized bloom formation of *L. polyedrum*: an interplay of behavior and physical forces?

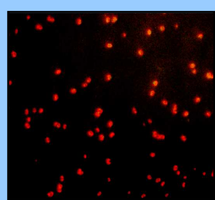


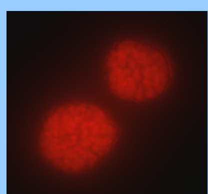

## Grazing of *L. polyedrum* on *Ostreococcus* sp.

*Lingulodinium*: 1000 cells / ml

*Ostreococcus* abundances:





Max. Ingestion Rates:

100 cells / Lingulodinium x hour$^{-1}$

Moorthi et al., in prep.

## Summary Seasonality and HAB

- herbivore and prey dynamics oscillate (clear waterstate when grazed down)
- Selective feeding => grazing resistance
- ind. size (cell size, colonies)
- indigestibility (chemical intolerance/toxicitiy)
- forming of appendages
- mucus production

**Harmful Algal Blooms (HAB) and red tides:**
allelopathy, grazer deterrence, secondary metabolites produced for other reasons (not directed at consumers or competitors)

many red tide organisms are mixotrophic and do not only have a major impact as phototrophs, but also as grazers

## Literature


- Sommer, Biologische Meereskunde (2nd ed., Springer)
- Valiela, Marine ecological processes, Springer
- Begon, Harper, Townsend: Ecology (4th ed, 2005, Blackwell)
- Hausmann & Hülsmann, Protozoology (2nd edition, Thieme)