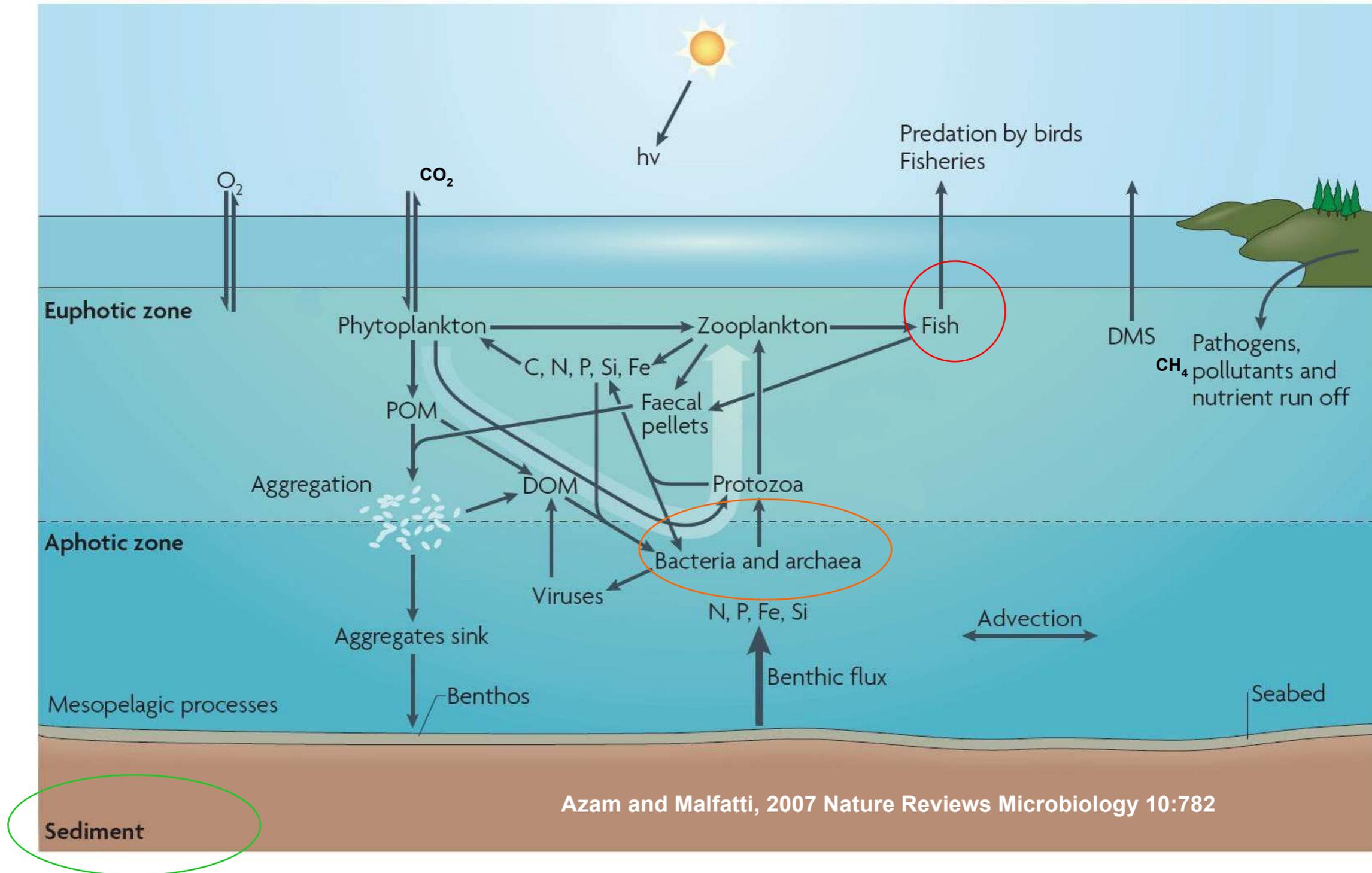
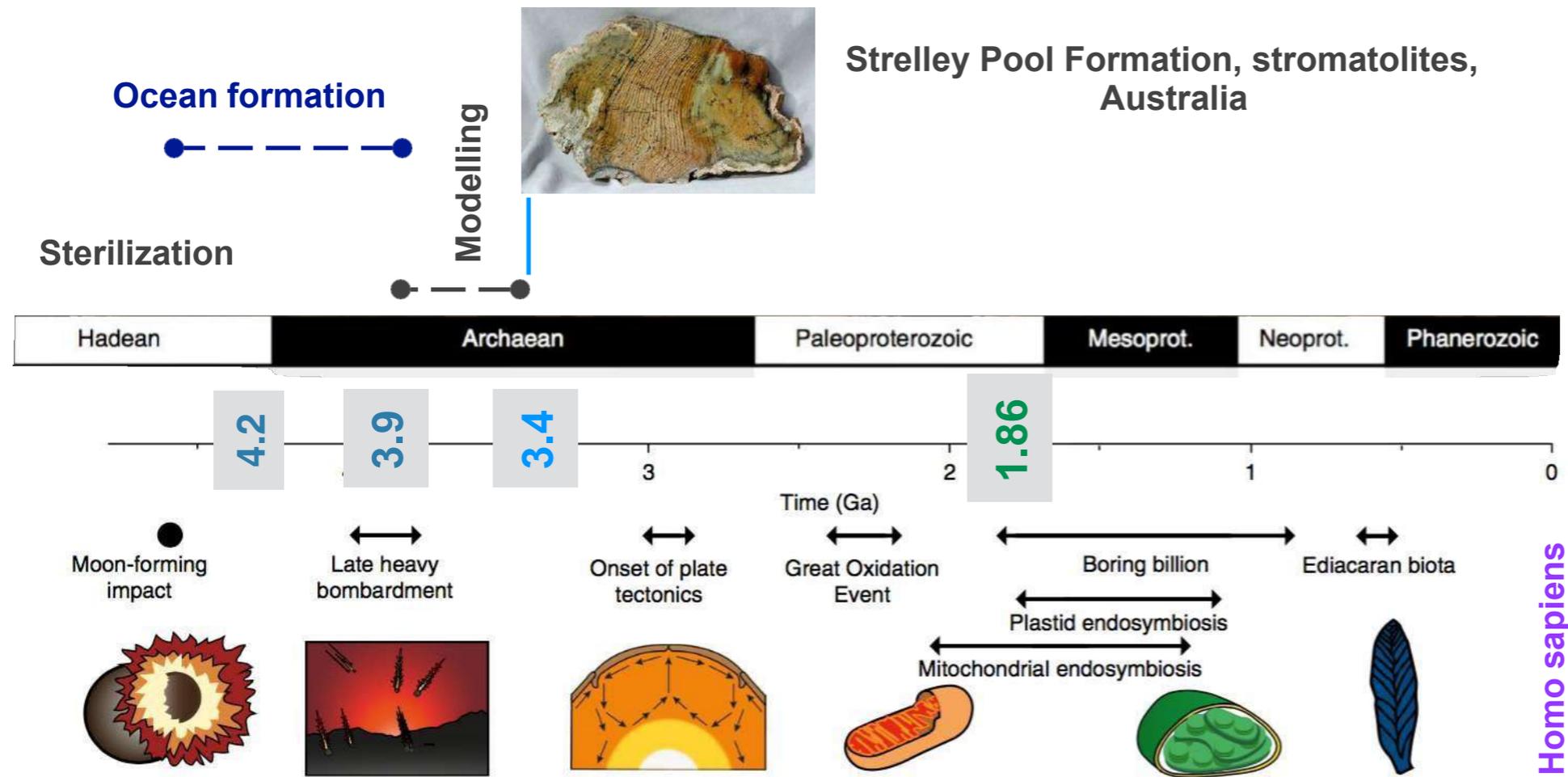


L02: Metabolic Diversity and Ecophysiology of Marine Microbes

Marine Microbial Carbon Biogeochemical cycle



Life on Earth and in the Ocean



Strelley Pool Formation, stromatolites, Australia

Betts et al., 2018
Moody et al., 2024

LUCA
LUCA: Last Universal Common Ancestor

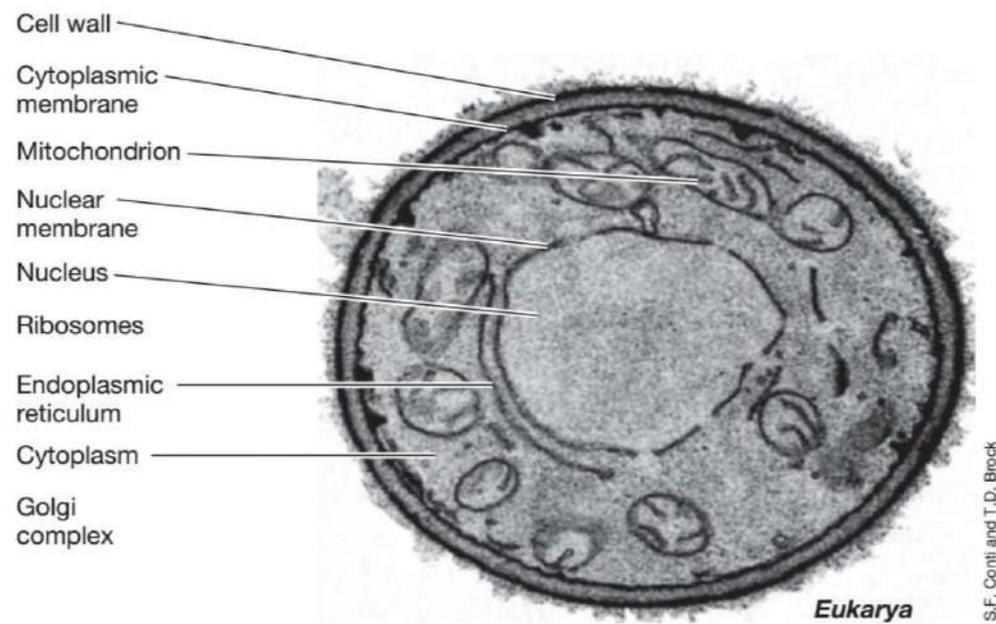
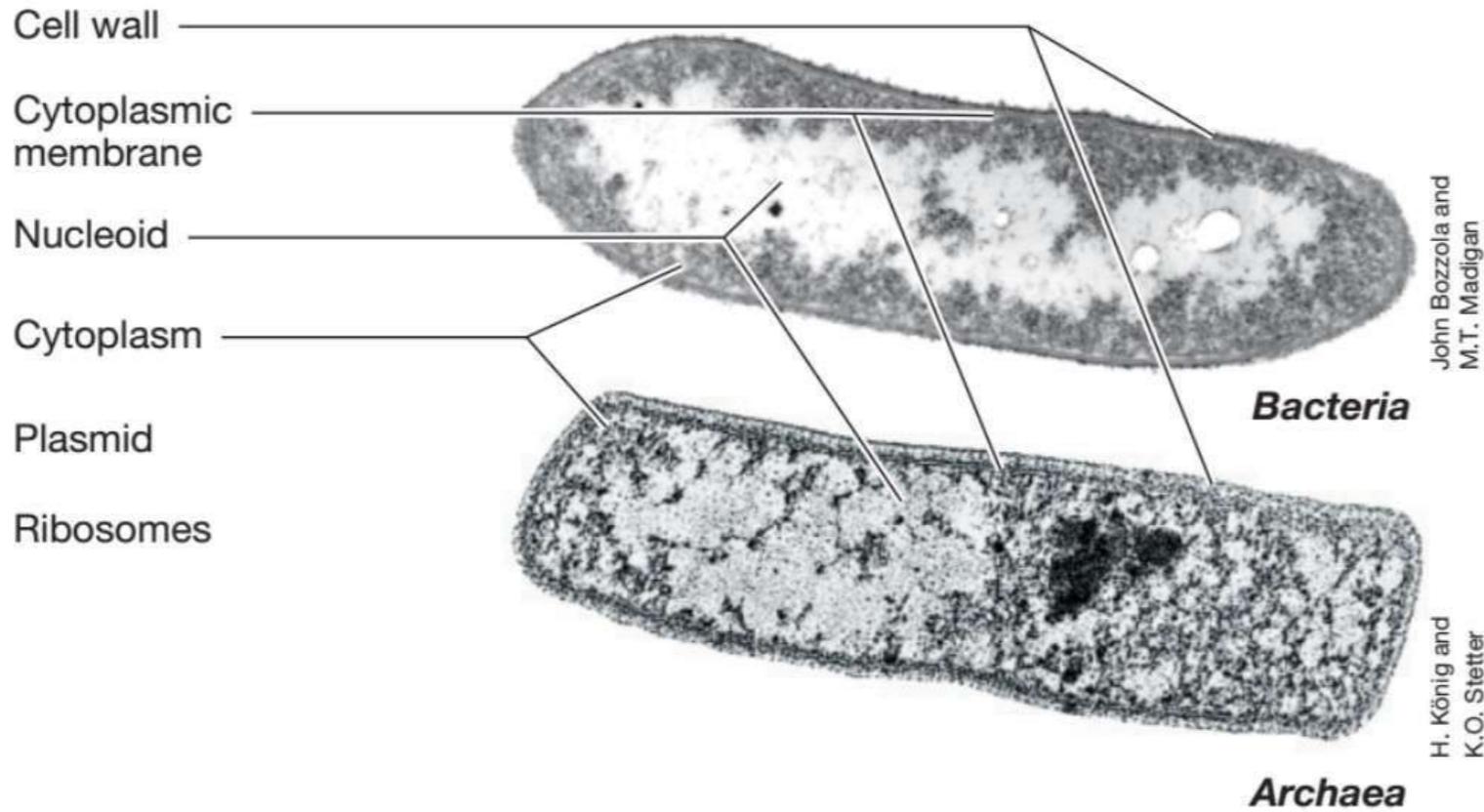
Bac & Arc

Euk

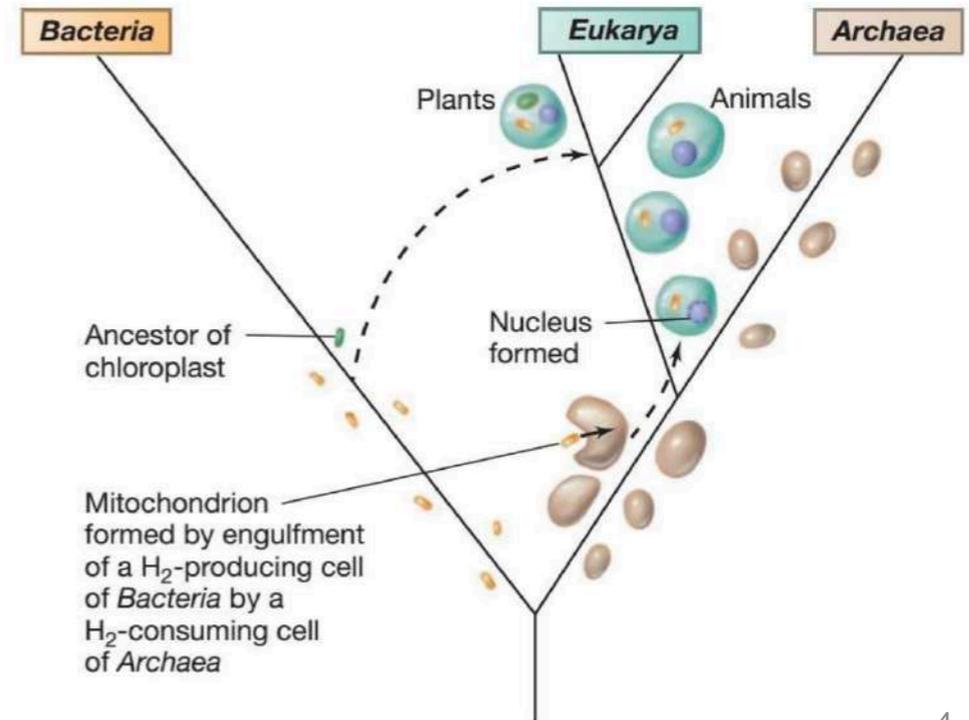
Homo sapiens

Earth and Ocean were very different than today

Bacteria, Archaea & Eukarya



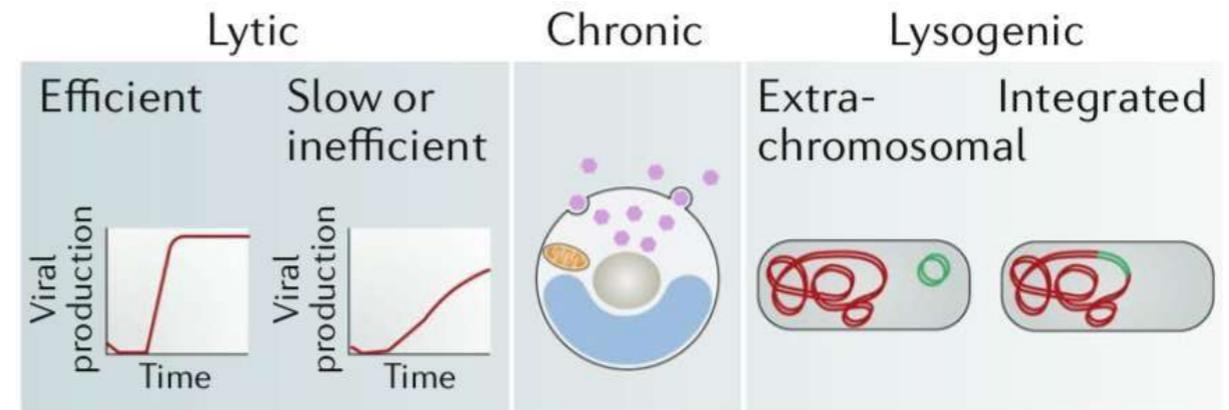
Madigan et al. 2018



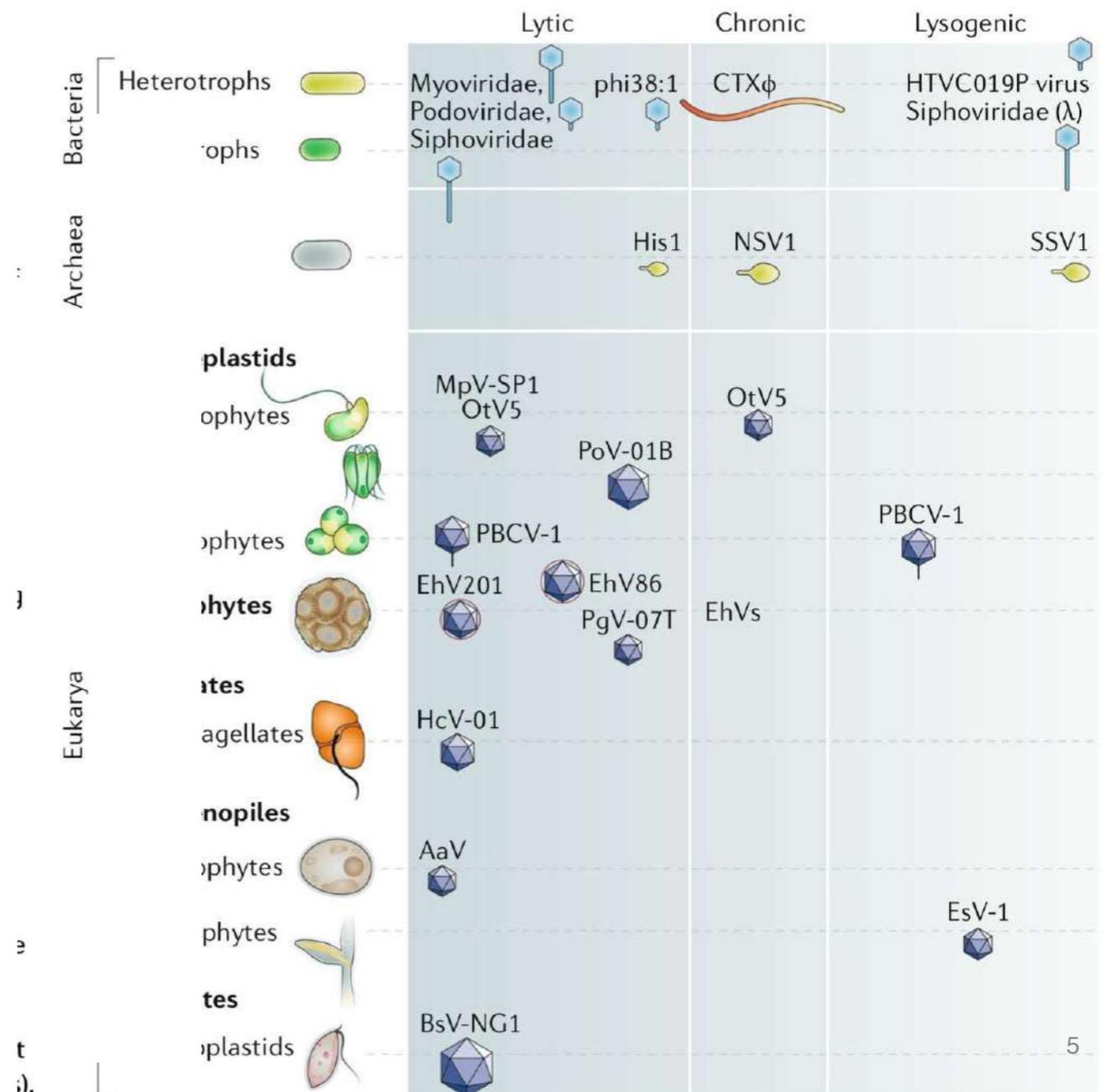
Viruses

- Moving genetic information
- Role in evolution
- RNA - DNA
- Diverse life strategies
- Interaction with every domain of life
- Interaction with other viruses
- Cells can become resistant

Spectrum of viral infection strategies

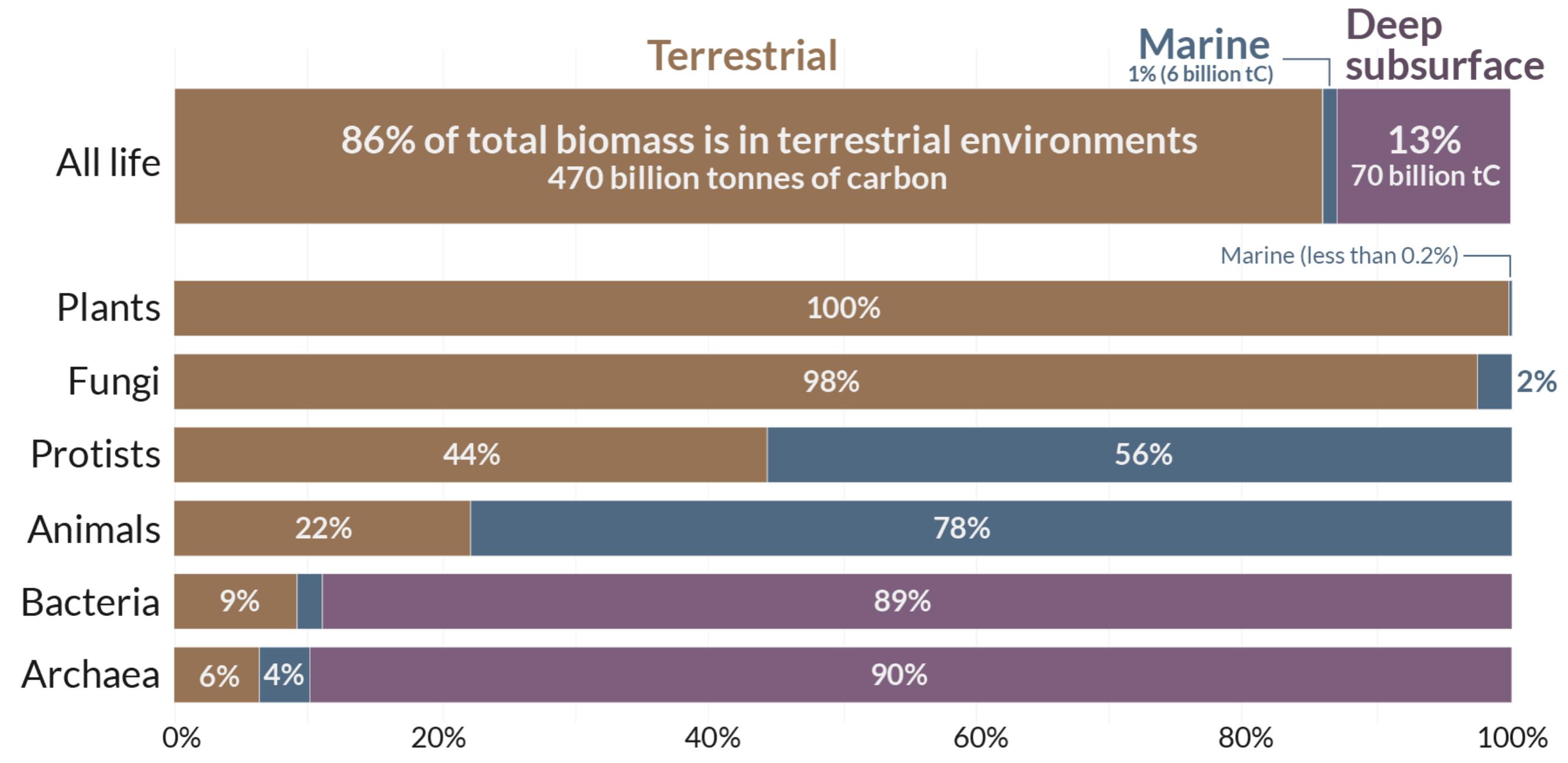


b Examples of observed infection strategies



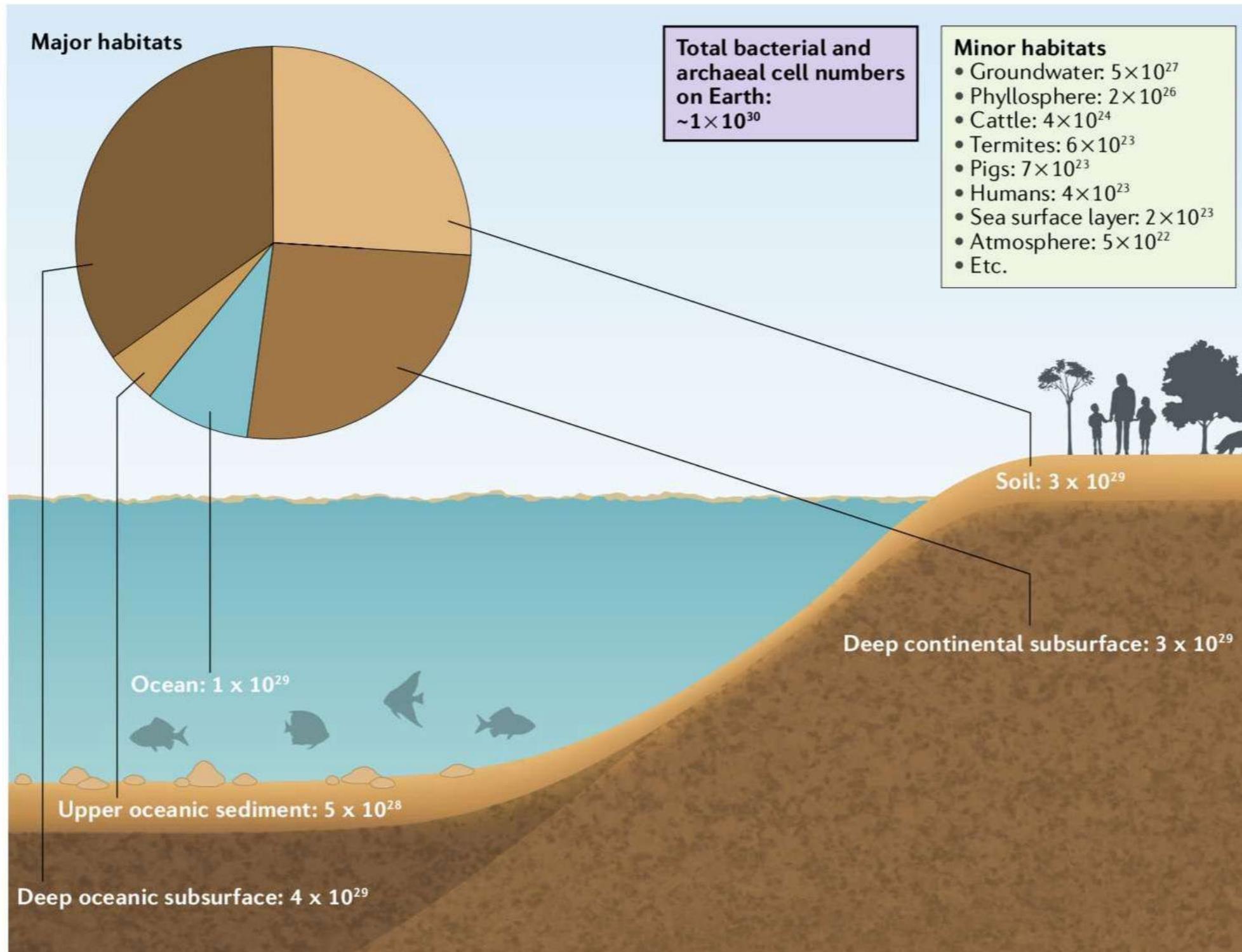
Where do we find life on Earth?

Distribution of global biomass across the world's environments. Biomass is measured in tonnes of carbon.

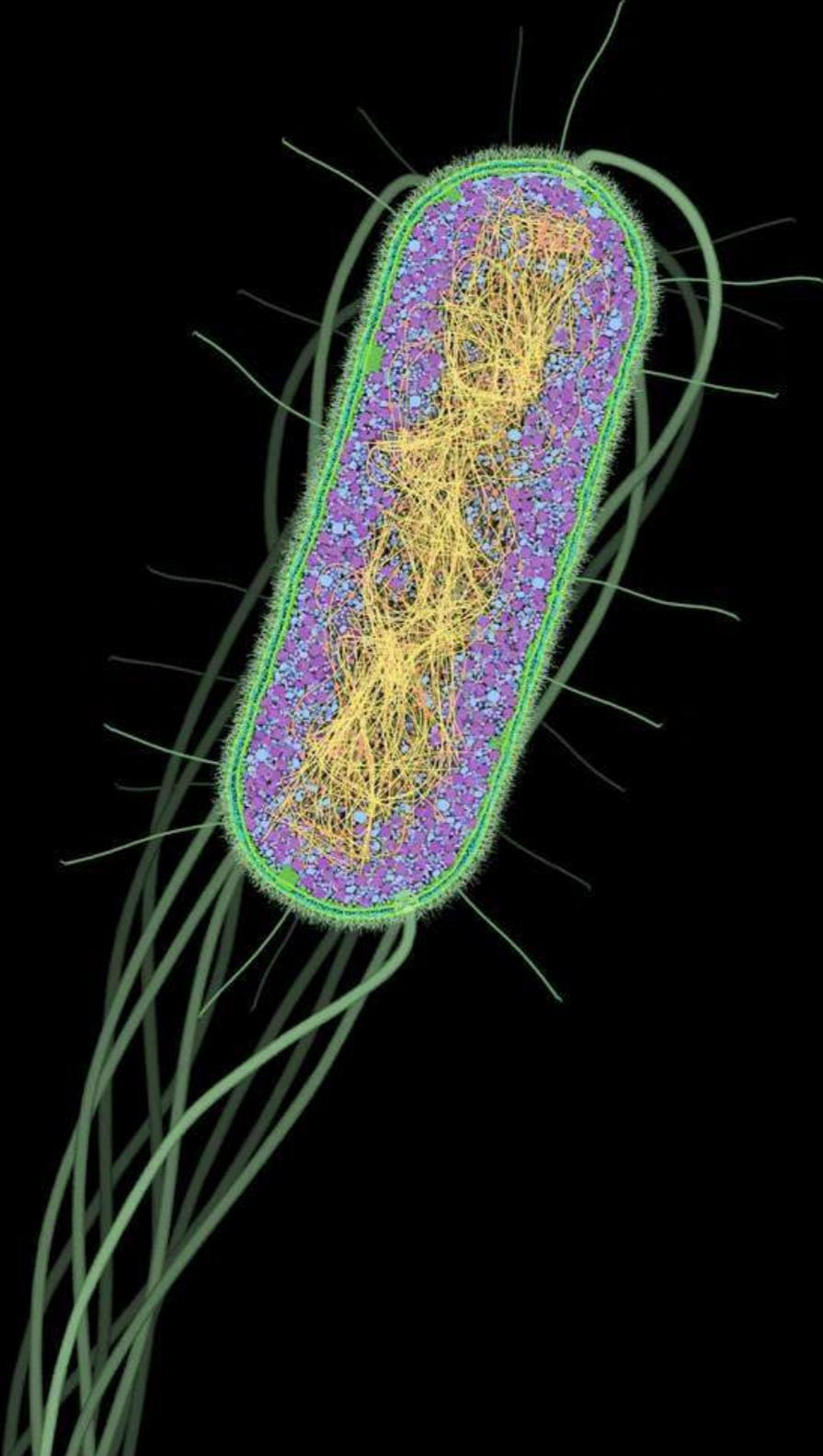


Data source: Bar-On, Y. M., Phillips, R., & Milo, R. (2018). The biomass distribution on Earth. *Proceedings of the National Academy of Sciences*.
 OurWorldinData.org – Research and data to make progress against the world's largest problems. Licensed under CC-BY by the authors Hannah Ritchie and Max Roser.

Abundance of bacteria and archaea in different habitats on Earth



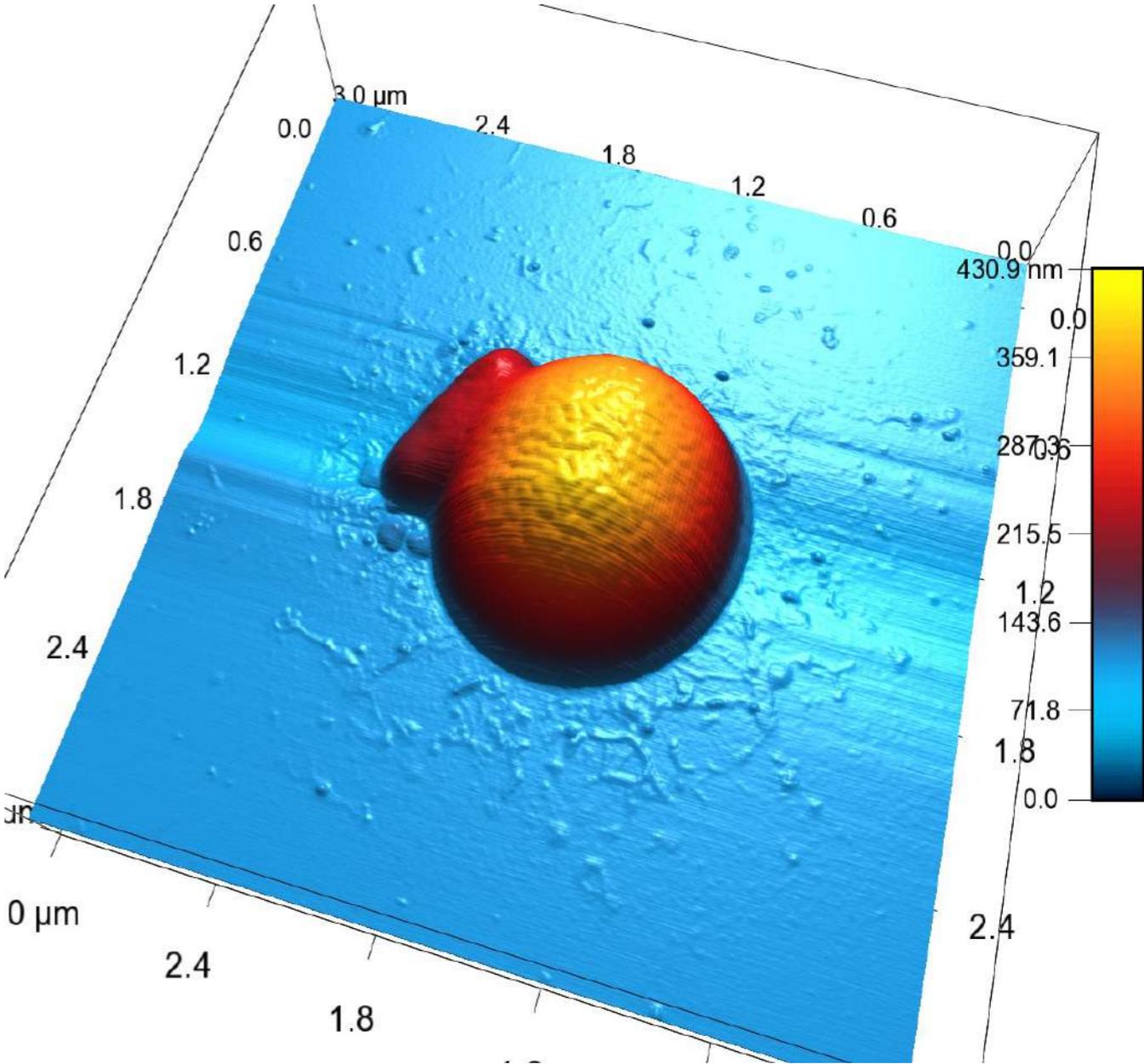
From micro to nanoscale



E. coli

David S. Goodsell

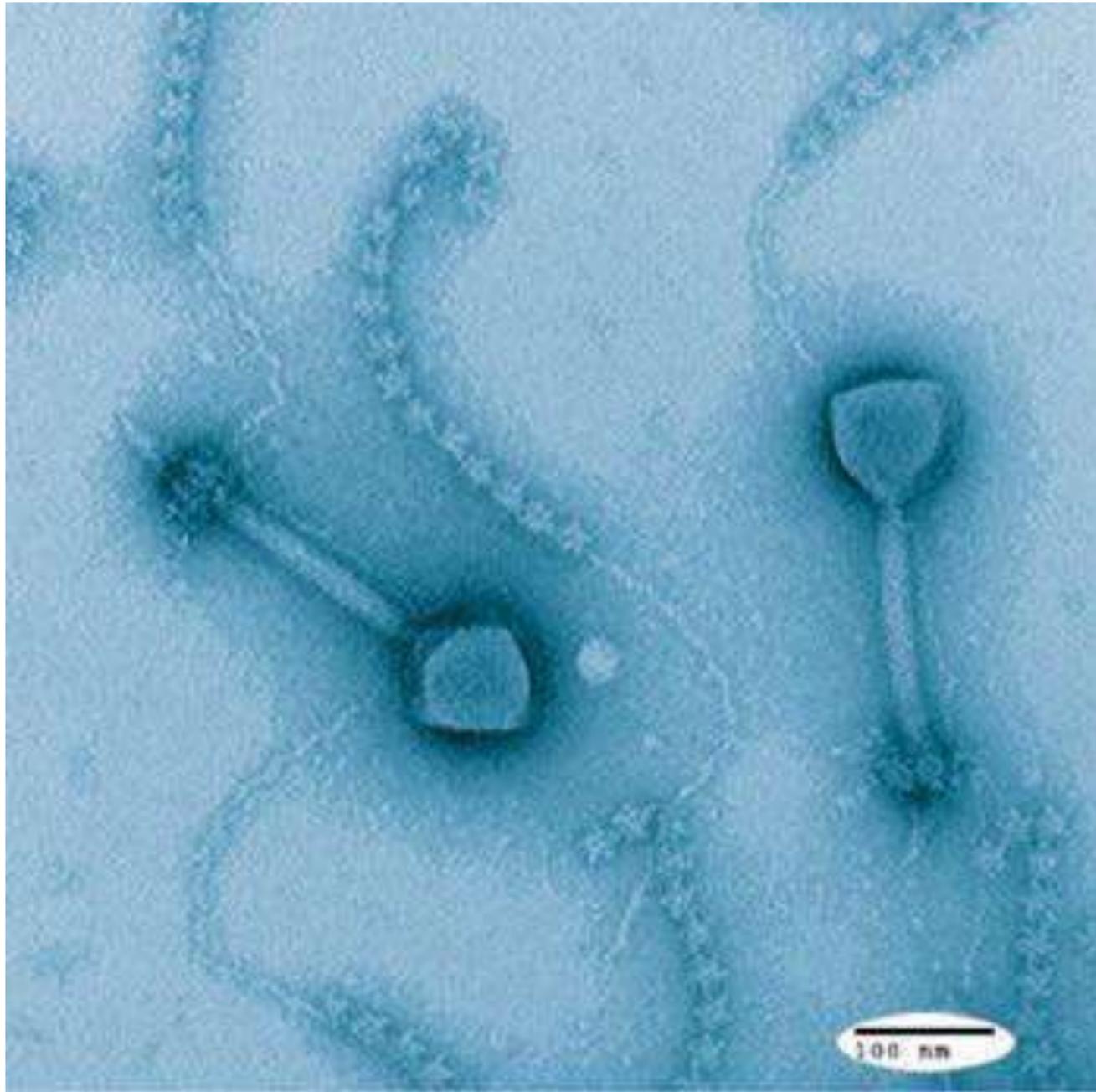
Synechococcus & heterotrophic bacterium



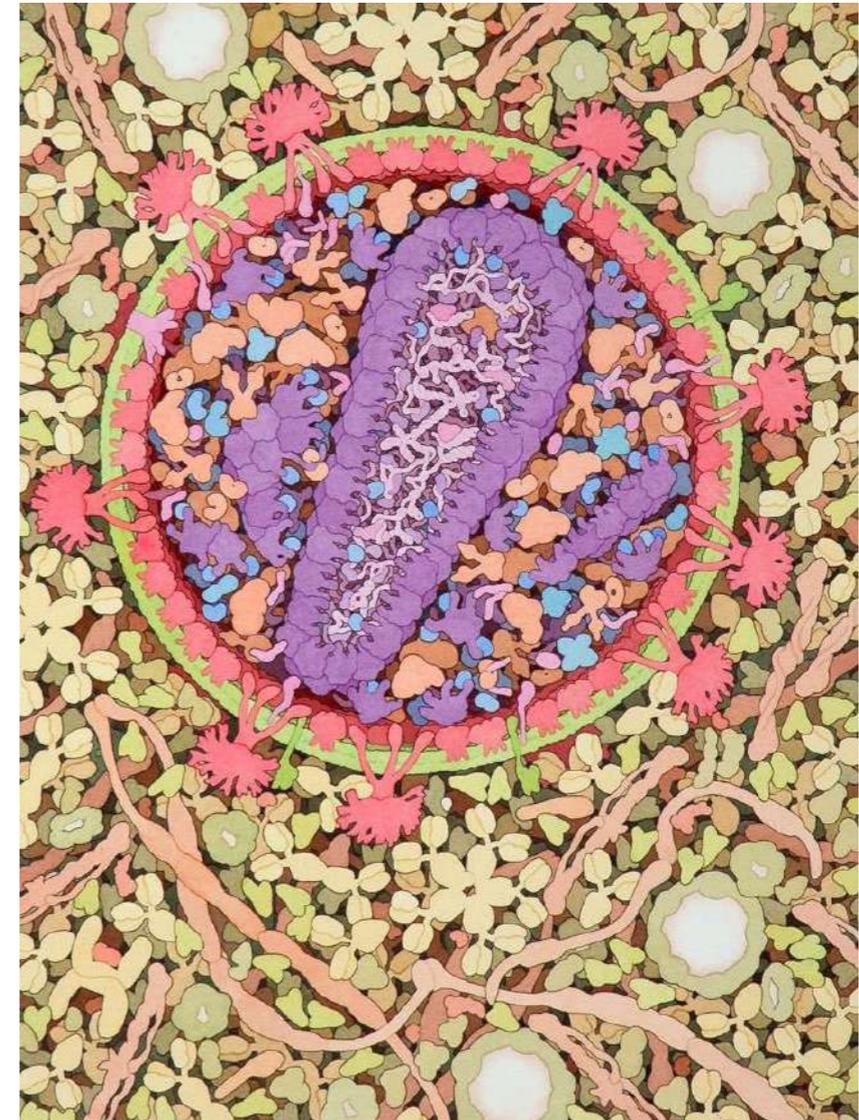
F. Malfatti

Viruses

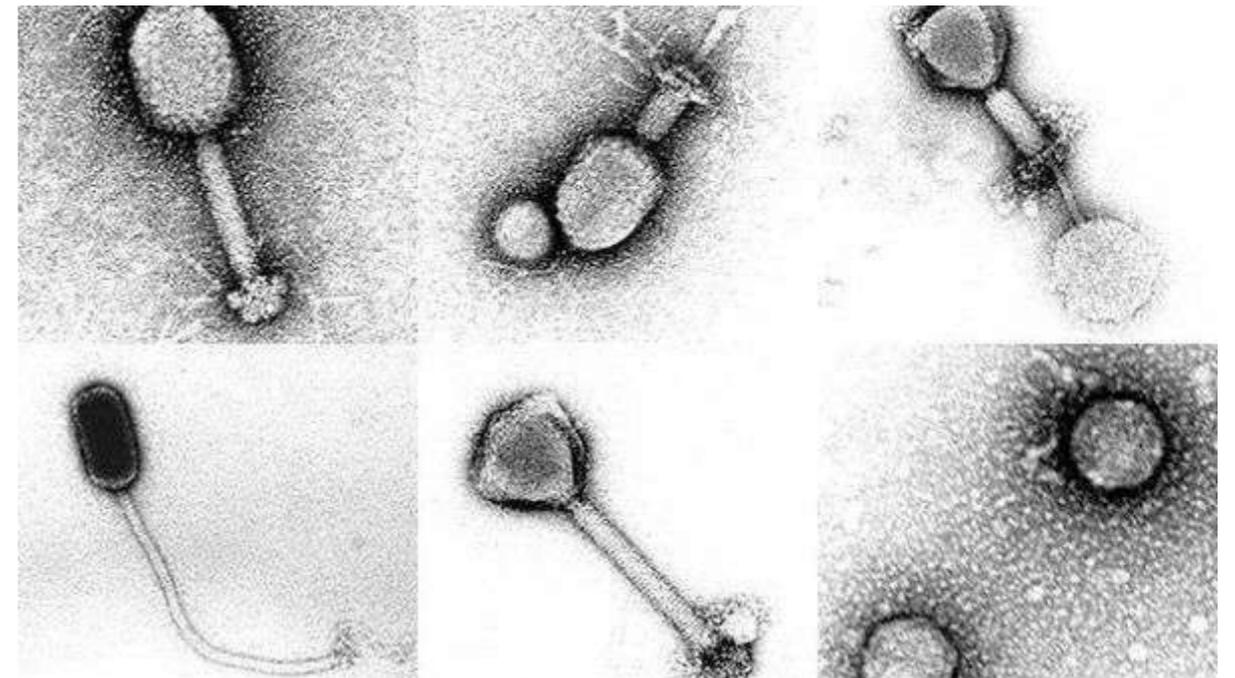
Nucleic Acid
Proteins
Some lipids



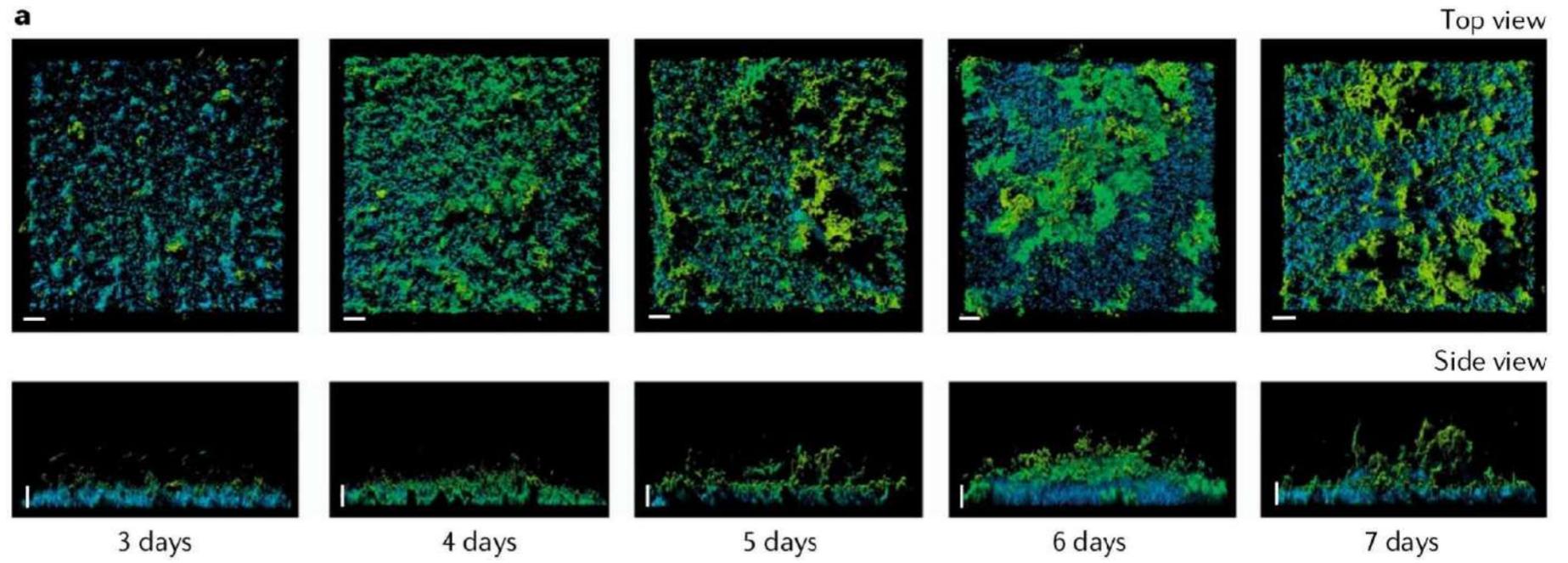
Jennifer Brum, Tucson Marine Phage Lab



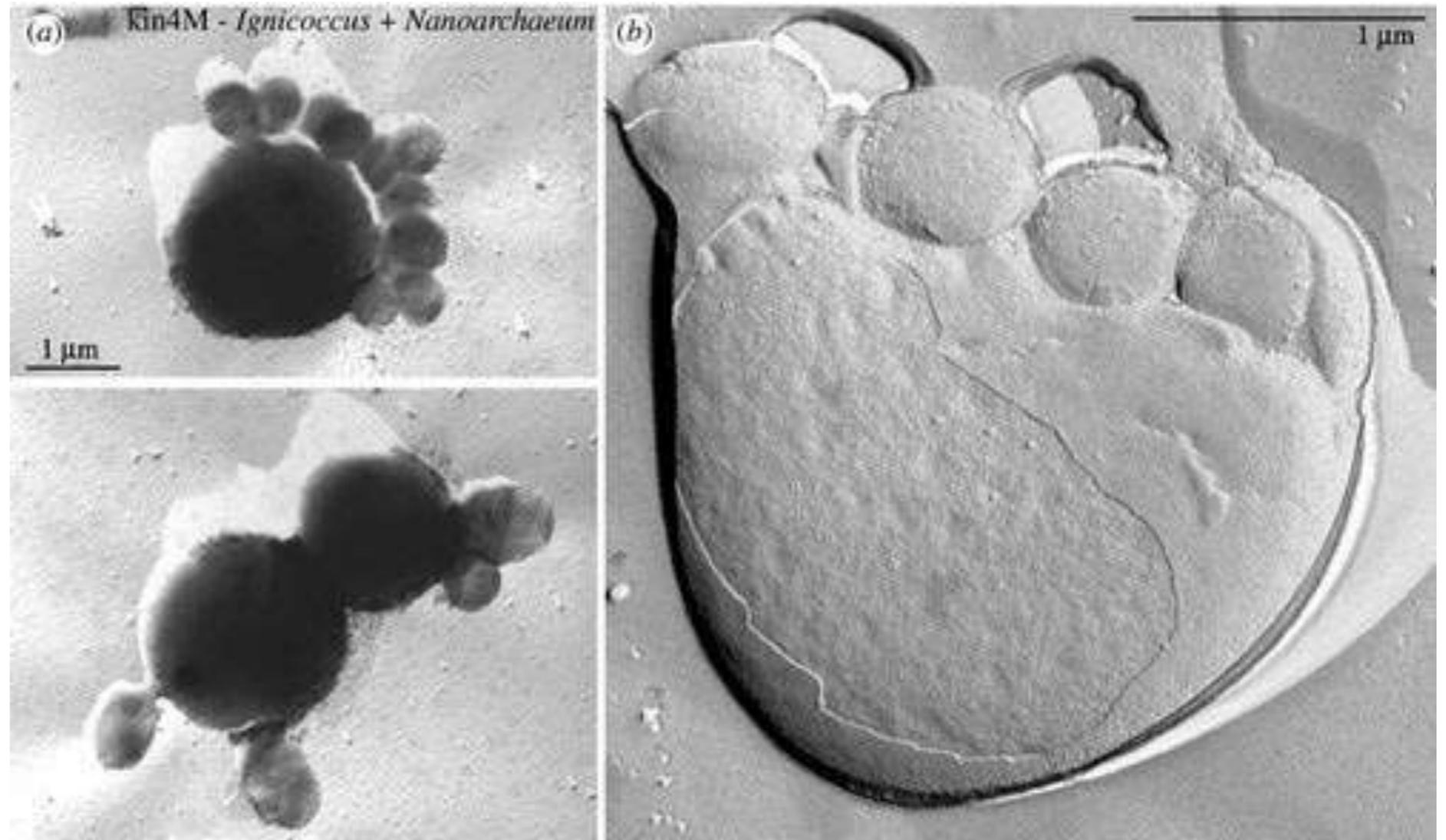
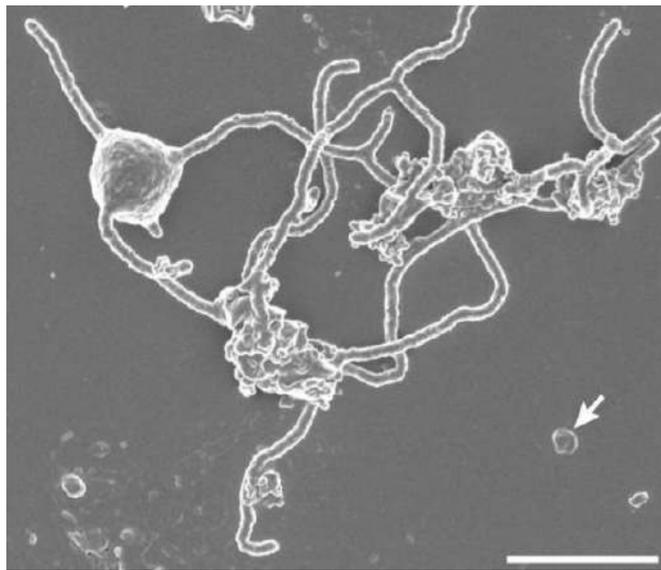
David S. Goodsell



Archaea



van Wolferen et al., 2018



Bacterial cities

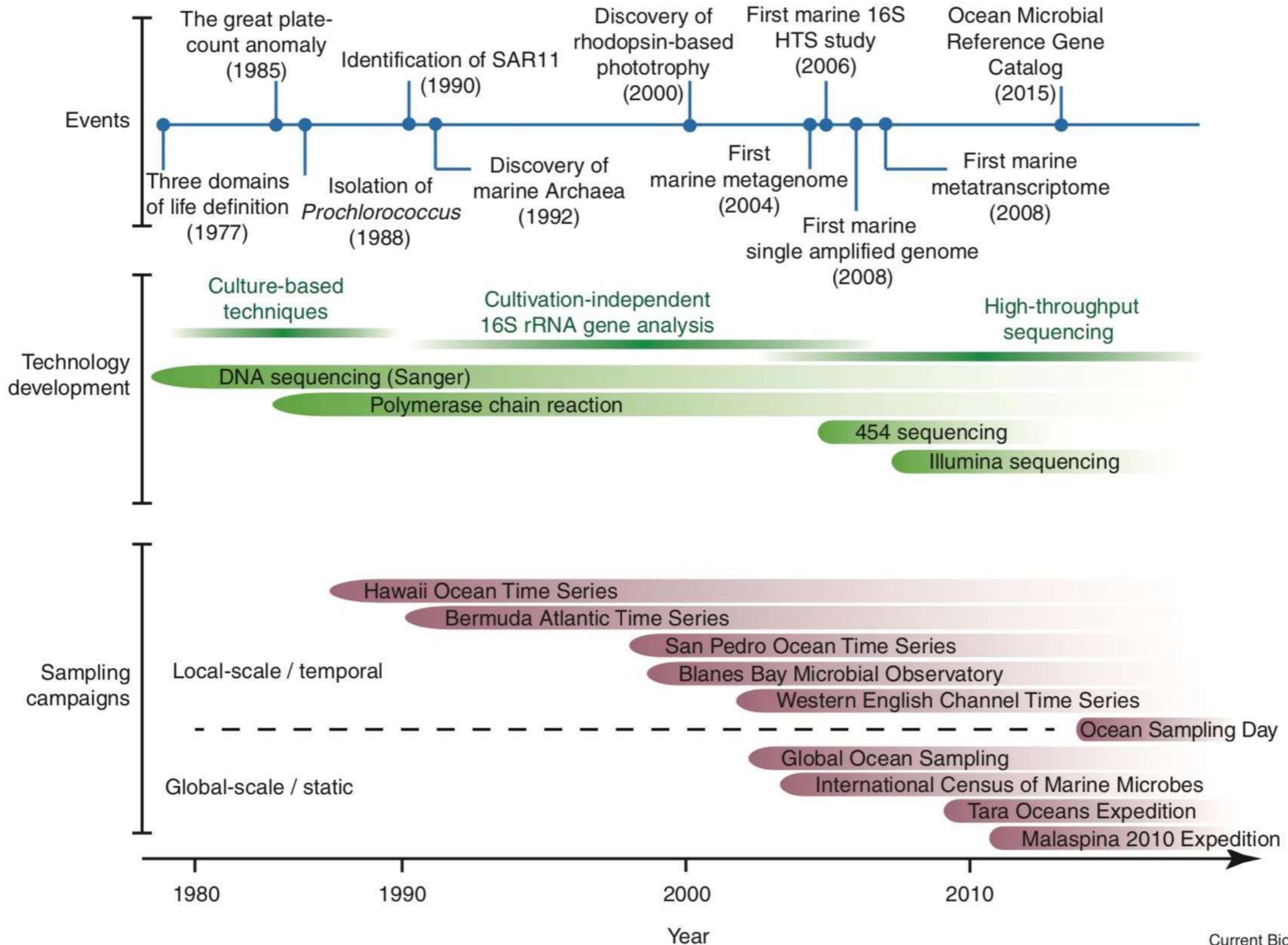
Marine bacteria colonies growing on an agar plate



Lophelia II 2012 Expedition, NOAA-OER/BOEM

- **The Ocean is a desert**
- **The Ocean is too cold**
- **The Ocean is too dark**
- **There are only 1000 bacteria per mL of SW...**
what can they do?

Technology development boots marine microbial ecology



The Age of Discovery

- 1977 Bacteria 10^6 mL⁻¹ (10^3 x cfu: great plate anomaly)
- 79/80 High bacterial growth & C demand (dynamic populations)
- 84 Protozoa (10^3 mL⁻¹) major predators on bacteria
- 79-90 Viruses abundant (10^7 mL⁻¹) & major predators on bacteria
- 79 *Synechococcus* 10^3 - 10^5 mL⁻¹
- 88 *Prochlorococcus* 10^4 - 10^5 mL⁻¹
- 90 Widespread Archaea throughout the oceans (10^4 - 10^5 mL⁻¹)

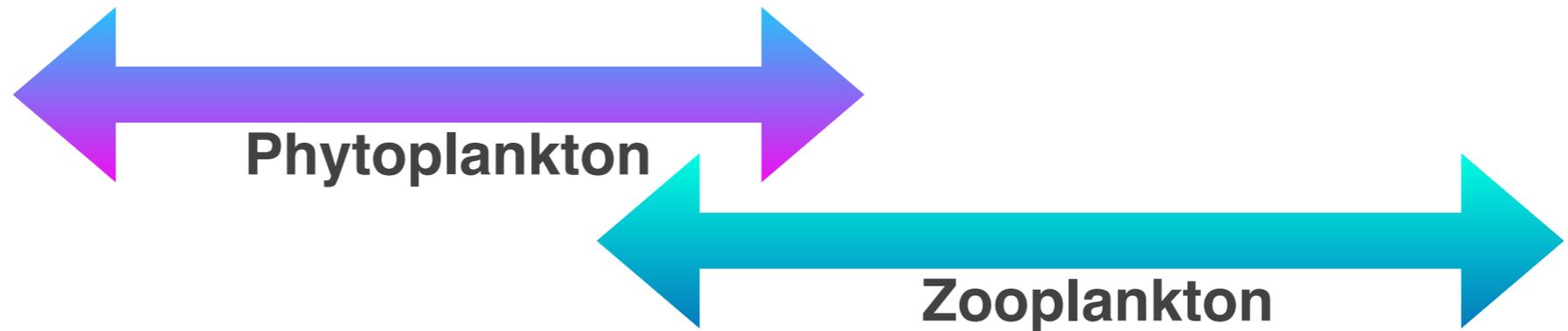
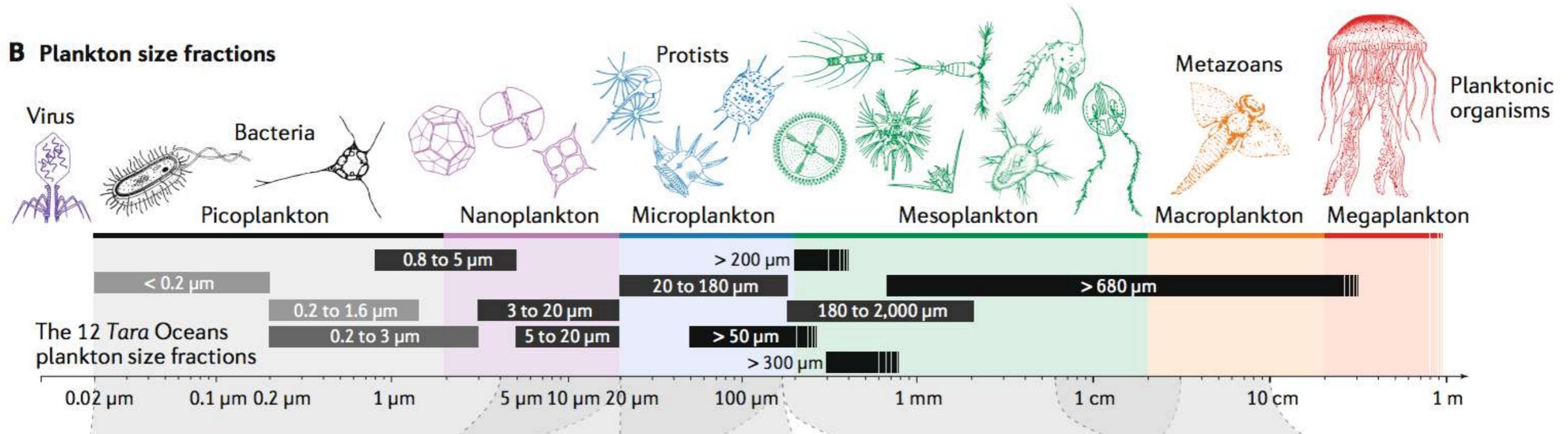
Form Farooq Azam

The Age of Discovery

Form Farooq Azam

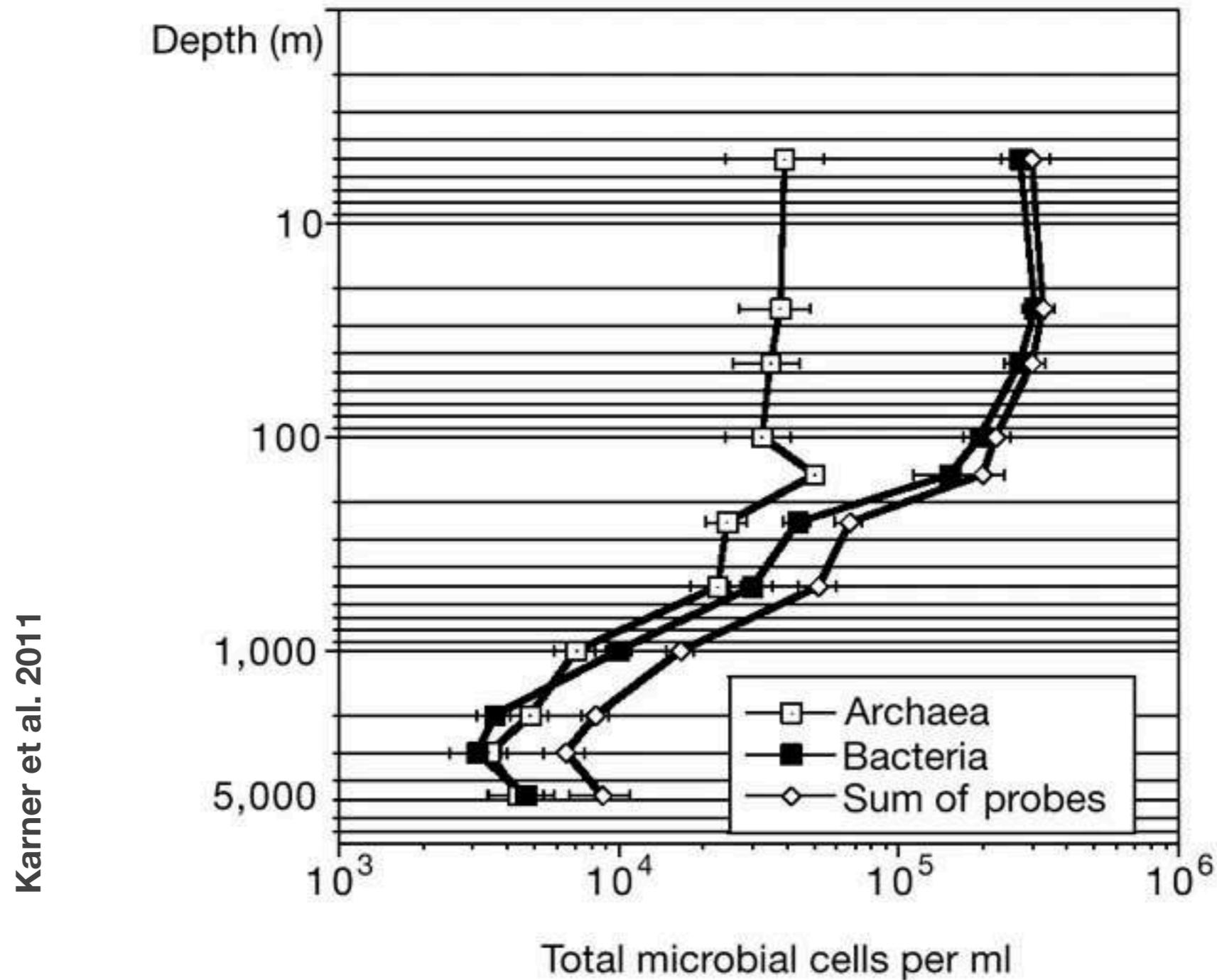
- 80-90-00 Microscale ecology
- Discovery of anoxygenic photosynthetic bacteria
- Rise of molecular ecology
- Massive marine genomics and metagenomics-- providing constrains for diversity and ecosystem function
- Culturing the “unculturable”
- Widespread picoeukaryotes
- Holistic approach: new chemicals, new way to design nano materials, detoxification
- OneHealth and Micorbes in Ocean-Climate models

Sizing the Microbial Ocean



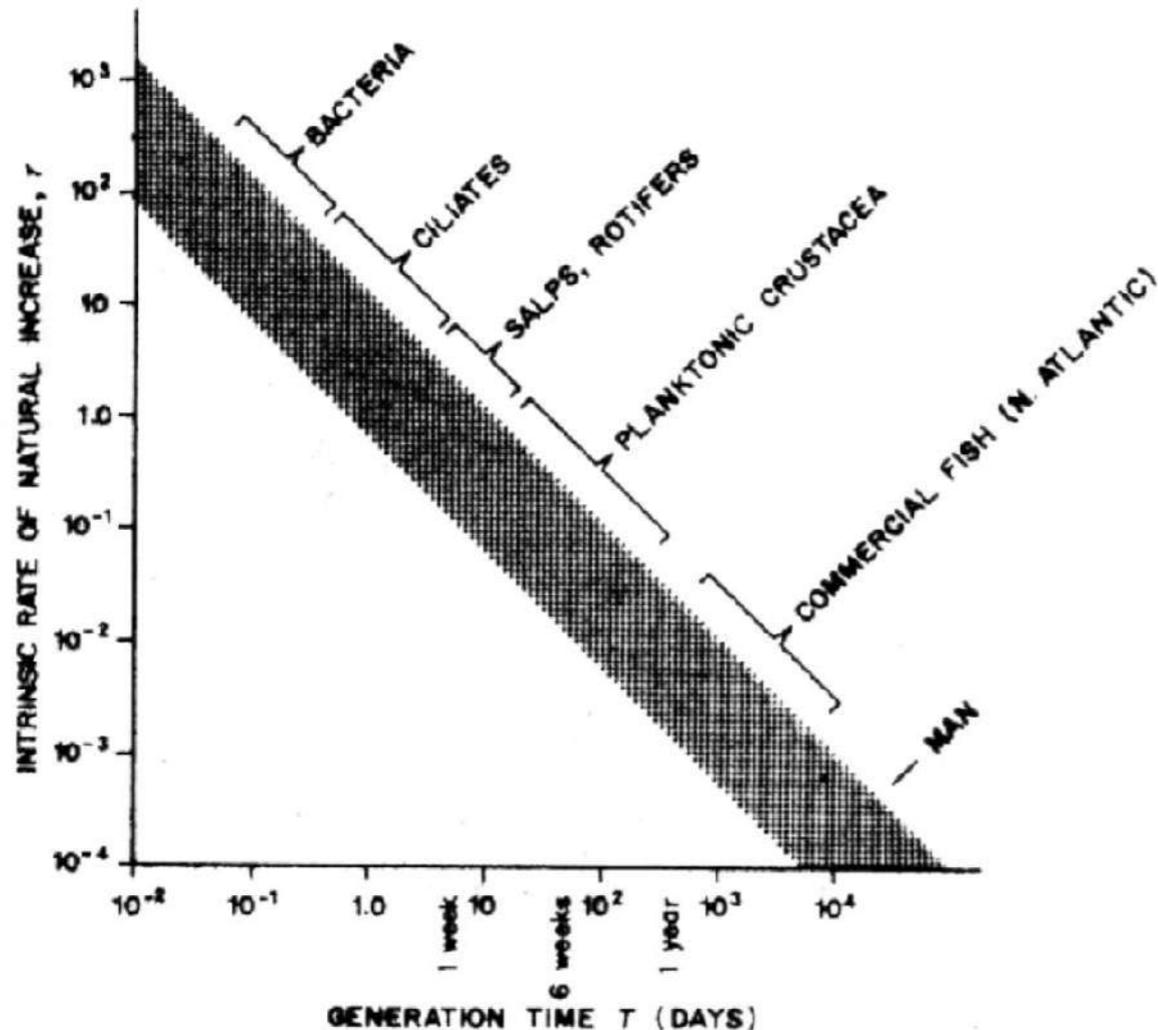
Sunagawa et al., 2020

SW Depth profiles of microbial domain cell abundance: Bacteria and Archaea



Data are averages of up to 14 roughly monthly samplings over a 1-yr period at the Hawai'i Ocean Time-series station, ALOHA

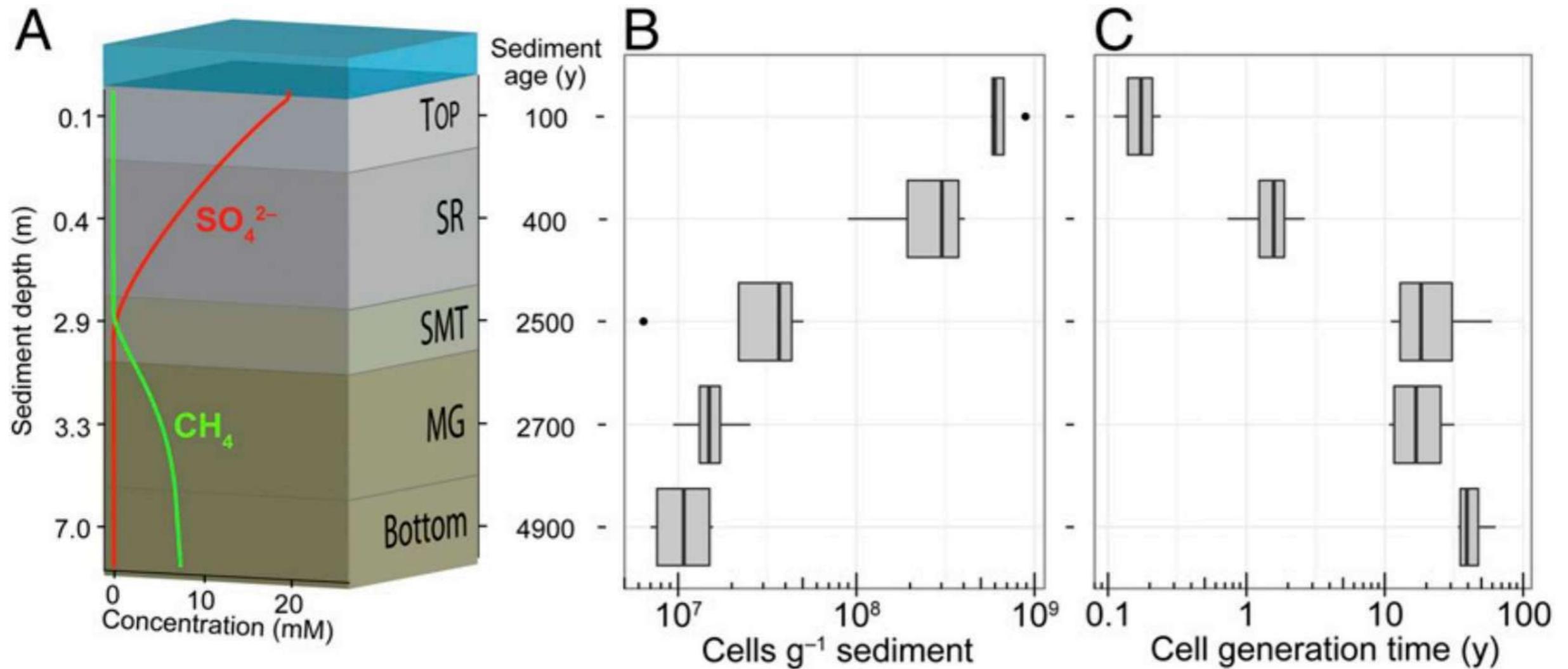
Bacteria and Archaea growth



- Fast generation time
- Slow generation time
- Diverse strategies to survive in the marine environments

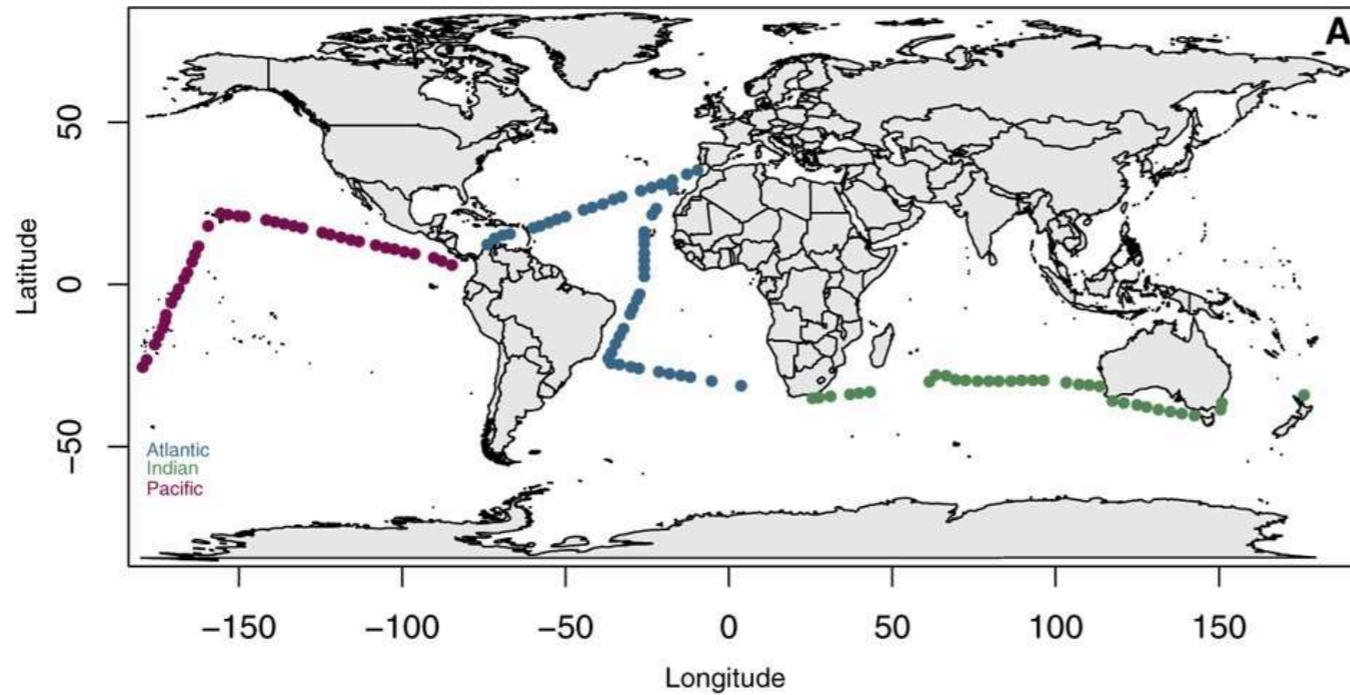
Figure 12B The intrinsic rate of population increase r , per day as a function of generation time. (Redrawn after Heron, 1972; in Anderson, 1981)

Sediment depth profiles of microbial cell abundance

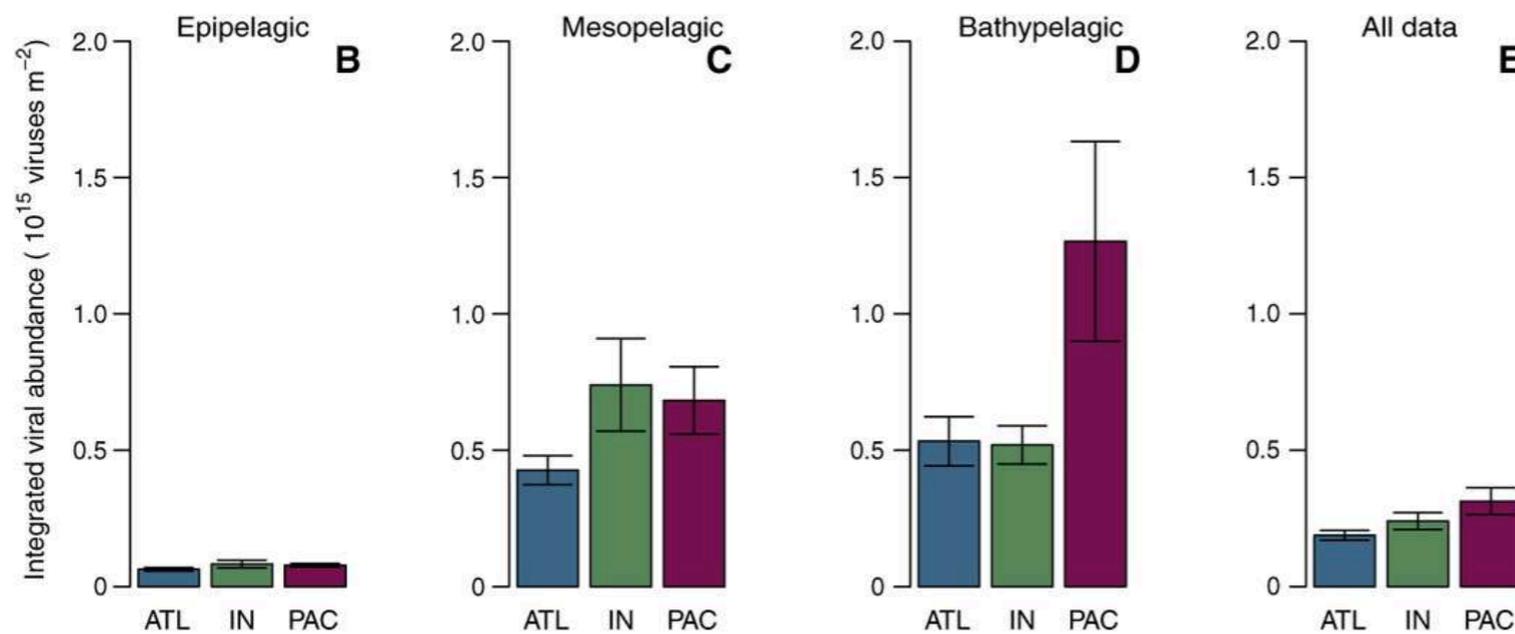


Starnawski et al., 2017

Virus depth profiles in the ocean

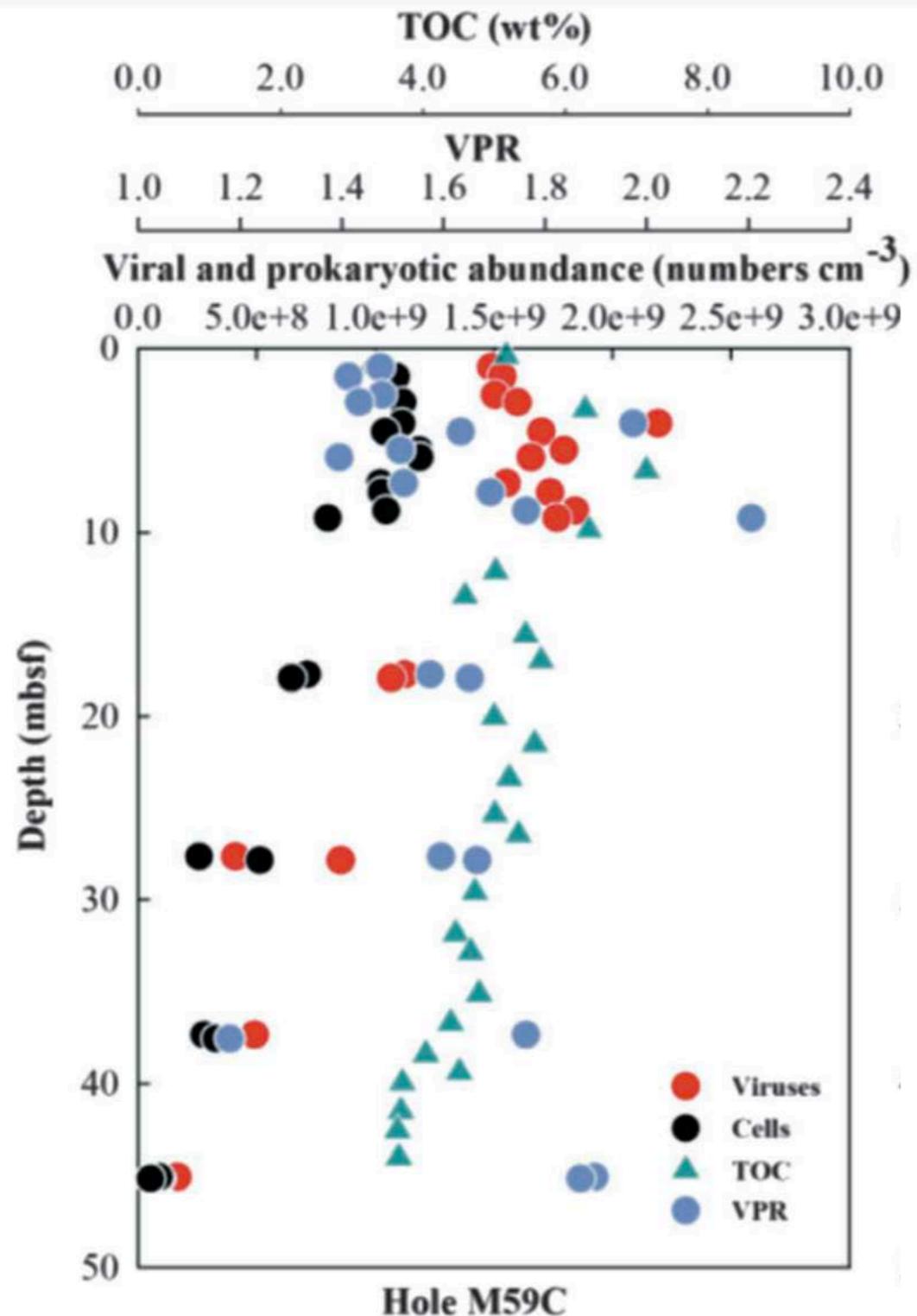


- Malaspina
- 10X change from 7.76×10^6 viruses mL^{-1} in the epipelagic layer (0 to 200 m) to 0.62×10^6 viruses mL^{-1} in the bathypelagic layer (1000 to 4000 m)



(B) epipelagic (0 to 200 m), (C) mesopelagic (200 to 1000 m), and (D) bathypelagic (1000 to 4000 m) layers

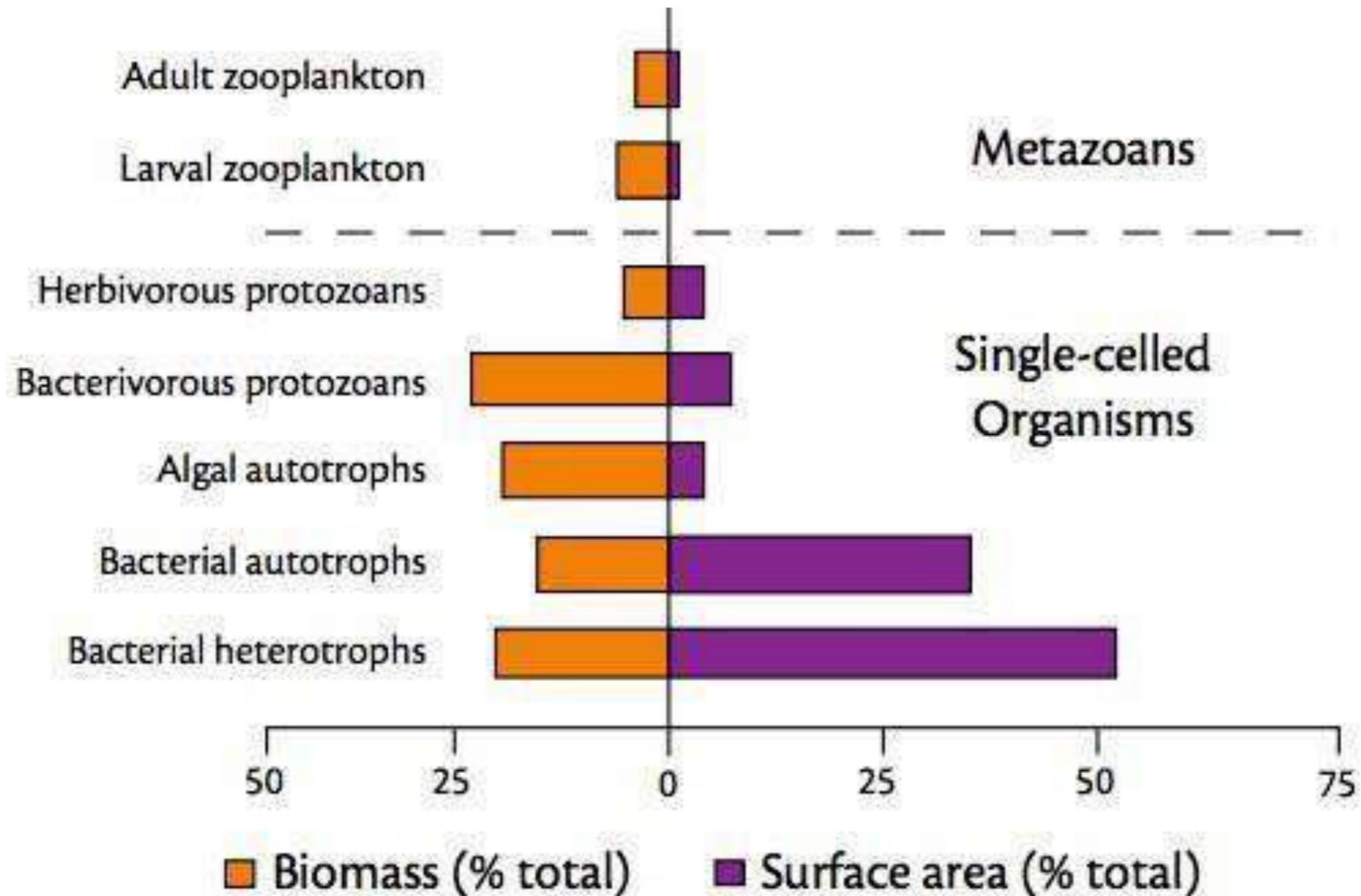
Virus depth profiles in the sediment



- Baltic Sea sub-seafloor biosphere harbors highly abundant viruses with densities up to 1.8×10^{10} viruses cm^{-3}
- High potential viral production down to 37 meters below seafloor in ca. 6000-years-old sediments

Bacteria & Archaea biomass

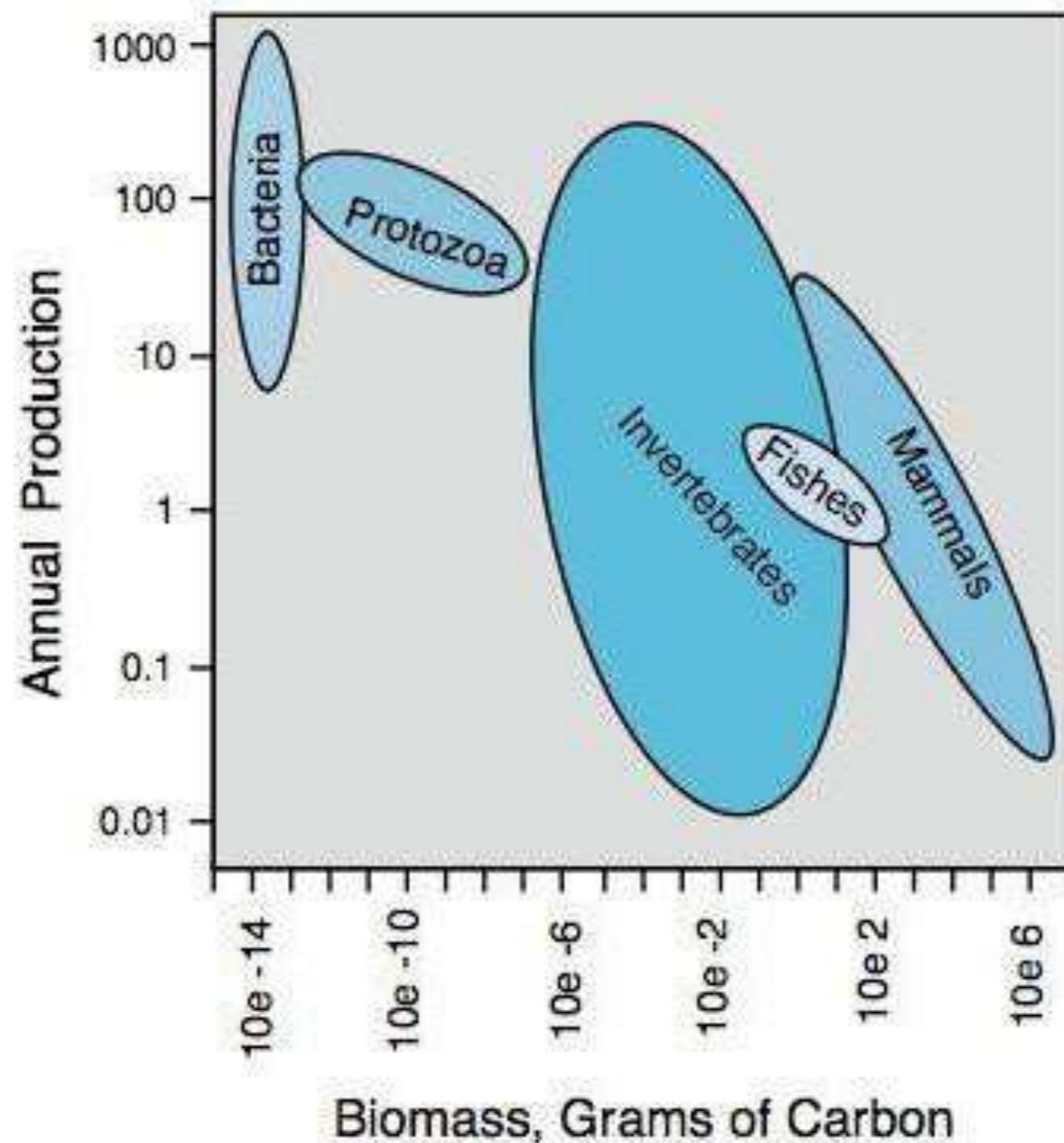
Pomeroy et al. 2015



•Microbes represent 90% of the total biomass in the ocean

Bacteria & Archaea production

Pomeroy et al. 2015



- Microbes are the most productive organisms in the ocean

Bacteria & Archaea respiration

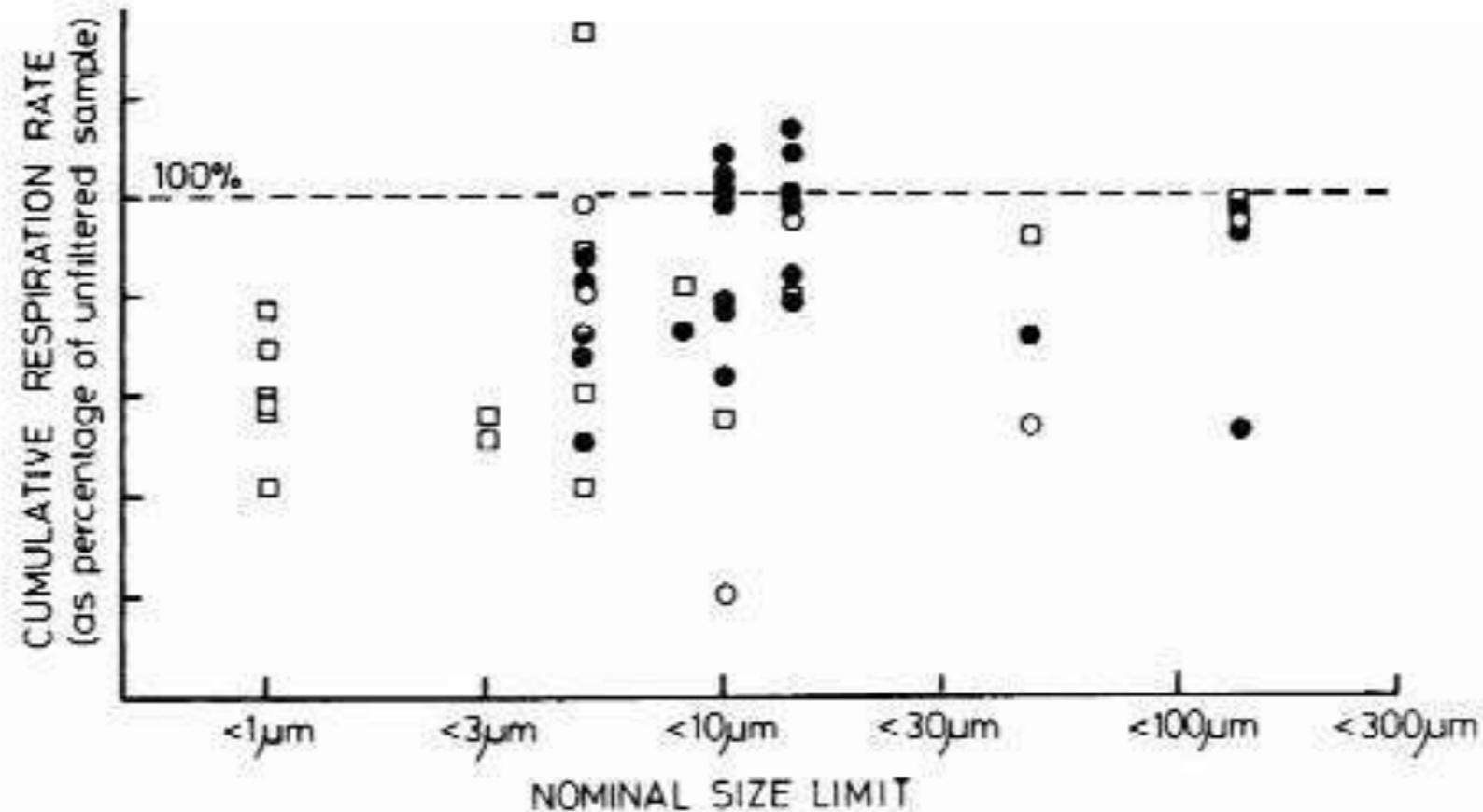
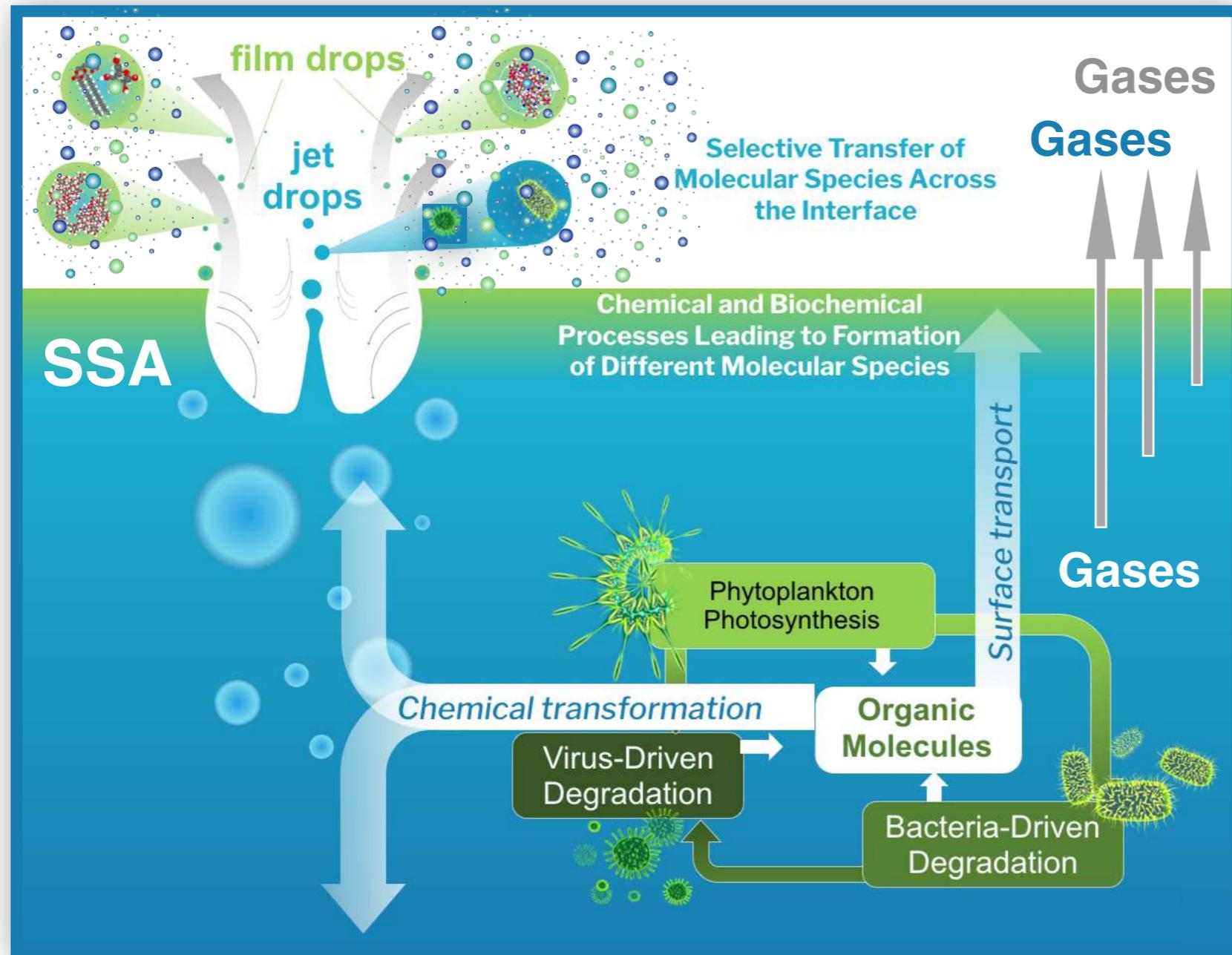


Figure 6. Distribution of respiratory activity with size. (□) CEPEX, samples from bag; (○) Loch Ewe, samples from bag; (●) Loch Ewe samples from outside bag. Data are expressed as cumulative respiration up to various size limits, normalized against the rate in the unfiltered sample. All the data points are for a single size horizon and are not replicates.

Williams (1984)

- Microbes contribute up to 90% to biota respiration

Microbial atmospheric structuring



- ~2000-10,000 Tg/yr of SSA is ejected from ocean globally
- **SSA has effect on climate and atmosphere**
- Ocean-atmosphere exchange ~ millions of microbes per m² every day



Microbial activities, biotransformation, influence ocean and atmosphere chemistry

Microbial abundance across the atmosphere-ocean-sea sediment:

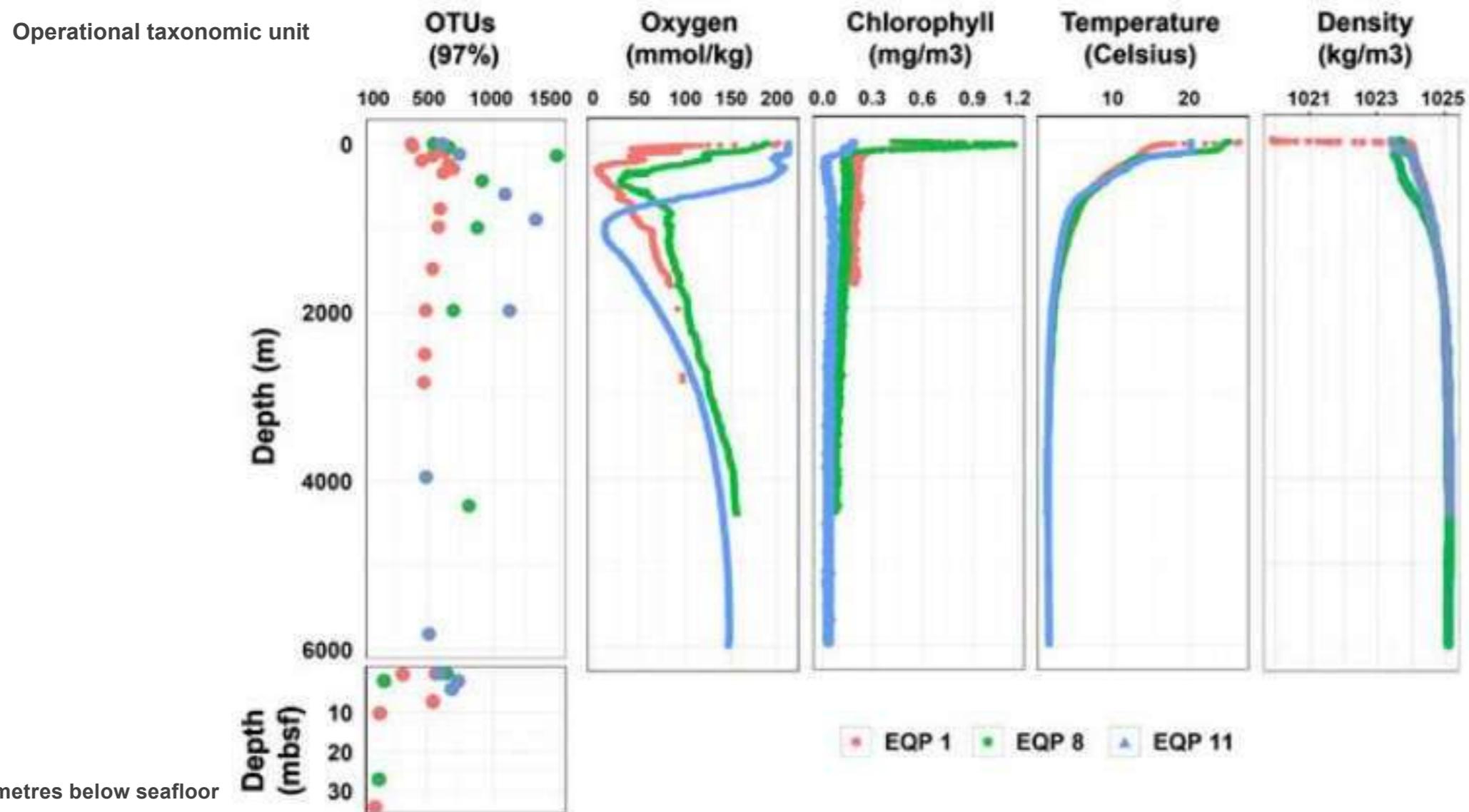
Sea-spray aerosol: 10^2 - 10^5 cells L^{-1} of air

Sea Surface Microlayer: 0.5 - 3×10^9 cells L^{-1}

Water column: < 0.01 - 1×10^9 cells L^{-1} ($> 5 \times 10^9$ cells L^{-1}); L: 20000 “species”

Surface sediment: ~ 0.001 - 1×10^{10} cells cm^{-3} ; g: 5000-19000 “species”

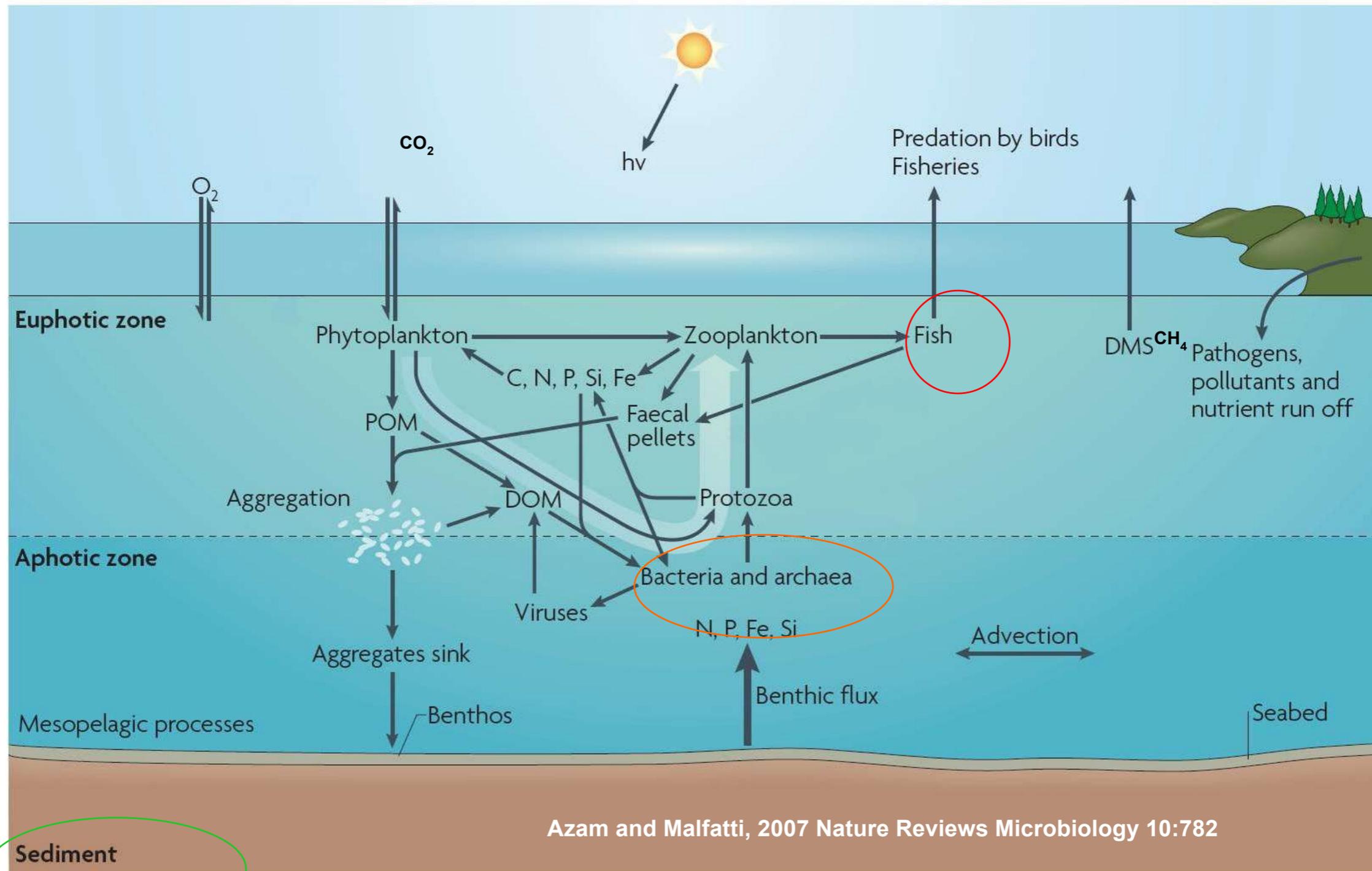
Deep subsurface sediment: ~ 0.001 - 1×10^{10} cells cm^{-3}



Walsh et al., 2016

Healthy Ocean \longleftrightarrow Functioning Ocean

Functioning Ocean \longleftrightarrow Functioning C biogeochemical cycle



Azam and Malfatti, 2007 Nature Reviews Microbiology 10:782

Global CARBON Reservoirs, Fluxes, and Turnover Times

Pools in Gt C, Fluxes in Gt C y⁻¹, Gt = 10¹⁵ g;
 * = living pools; (turnover times)

From Farooq. Adam

Terrestrial

NPP = 50 y⁻¹ ↓
 Deforestation 1.4 y⁻¹ ↑
 Combustion (80's) 5.4 yr⁻¹ ↑

Atmosphere

750 (3-5 y)
 Ann. increment = 3.2 y⁻¹
 (~ +1.5 ppmv CO₂ y⁻¹)

Marine

NPP = 50 y⁻¹ ↓
 New production = 10 y⁻¹

Plants*

550-680 (50 y)

Rivers

DOC: 0.2 y⁻¹
 POC: 0.2 y⁻¹

Coastal Ocean

20% of NPP

Ocean CO₂ Exchange
 90 y⁻¹ ↑ 92 y⁻¹ ↓

Open Ocean

80% of NPP

Soils (~1m) 1580
 peat 360 (>1000 y)
 mineral 1220

microbial* 15-30 (<10 y)
 POC 250-500 (<100 y)
 remainder 600-800 (10²-10³ y)

Surface Sediments (~1m)

150 (0.1-1000 y)
 80% coastal
 20% deep sea

Surface
 100 m

DOC 40 (? y)
 POC 5, Living 2* (0.1-1 y)

Deep
 3.8 km

POC
 7 y⁻¹ ↓
 DOC 700 (5000 y)
 POC 20-30 (10-100 y)
 DIC 38000 (~2000 y)

References:

Hedges, 1992; Eswaran *et al.*, 1993;
 Siegenthaler & Sarmiento, 1993;
 Schimel *et al.*, 1994

Respiration ≅ NPP

Sediments

kerogen 15x10⁶ (>>1 my)
 methane clathrates 11x10³
 limestone 60x10⁶

Sedimentation

(long-term burial)
 0.1 y⁻¹ ↓

WSR 1997

Microbial growth I

1. Energy source to generate ATP

2. Carbon source of assembling cellular building blocks

3. For maintenance of existing cells/ for growth of new cells

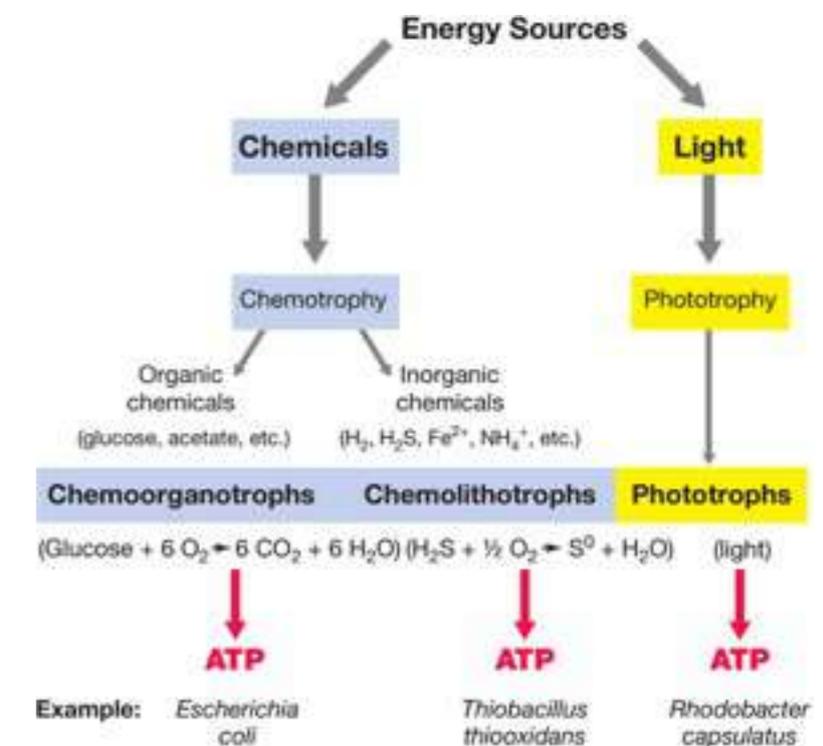
- > Successful exploitation well-defined energy and carbon source
- > Physical, biological and chemical properties vary in space and time

Carbon source	Energy source		
	Chemical, organic	Chemical, inorganic	Light
Fixed organic	Chemosynthetic organoheterotroph (Example: humans, fungi, <i>Pseudomonas</i>)	Chemosynthetic lithoheterotroph (Example: <i>Beggiatoa</i> sp.)	Photosynthetic heterotroph (Example: purple and green bacteria; <i>Rhodospirillum</i>)
Gaseous CO ₂		Chemosynthetic lithoautotroph (Example: ammonia-, hydrogen-, and sulfur-oxidizing bacteria; <i>Nitrosomonas</i> , <i>Aquifex</i>)	Photosynthetic autotroph (Example: plants, algae, <i>Prochlorococcus</i>)

Terminology:

- Autotroph: carbon from CO₂ fixation
- Heterotroph: carbon assimilated from (fixed) organic compounds
- Photosynthetic: energy from light
- Chemosynthetic: energy from oxidizing reduced chemicals
- Chemolitho: energy from oxidizing inorganic reduced chemicals
- Chemoorgano: energy from oxidizing organic reduced chemicals.

Madsen, 2016



Metabolism & Growth in a limited environment

Habitat characteristics and nutrient limitations faced by three physiological classes of microorganisms			
Habitat type	Photoautotroph	Chemolithotroph	Chemoorganoheterotroph
Ocean water	Daily light cycle, light penetration depth; scarce iron	Flux of reduced inorganic compounds, especially NH_3 , H_2S , H_2 , or CH_4 from nutrient turnover and hydrothermal vents	Carbon flux from phototrophs, dead biomass, and influent waters
Lake water	Daily light cycle, light penetration depth; scarce phosphorus	Flux of reduced inorganic materials, especially NH_3 , H_2 , and CH_4 from nutrient turnover	Carbon flux from phototrophs, dead biomass and influent waters
Sediment (freshwater and oceanic)	Daily light cycle, light penetration depth	Flux of reduced inorganic materials, especially NH_3 and H_2 from nutrient turnover or H_2 , H_2S , or CH_4 from hydrothermal vents	Flux of organic carbon from phototrophs and dead biomass; flux of final electron acceptors to carbon-rich anaerobic strata
Soil	Daily light cycle, light penetration depth	Flux of reduced gaseous substrates, especially methane from nutrient turnover by anaerobes	Slow turnover of soil humus, dead biomass, plant root exudates; leaf fall from vegetation
Subsurface sediment	No light	Flux of reduced inorganic materials, especially H_2 and CH_4 from geothermal origin	Carbon flux from nutrient turnover

Microbial Growth II

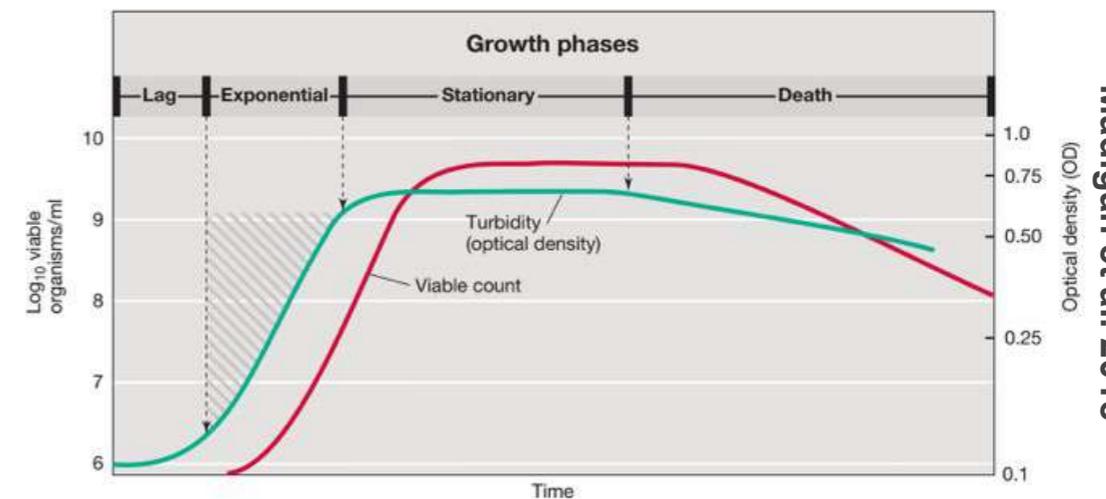
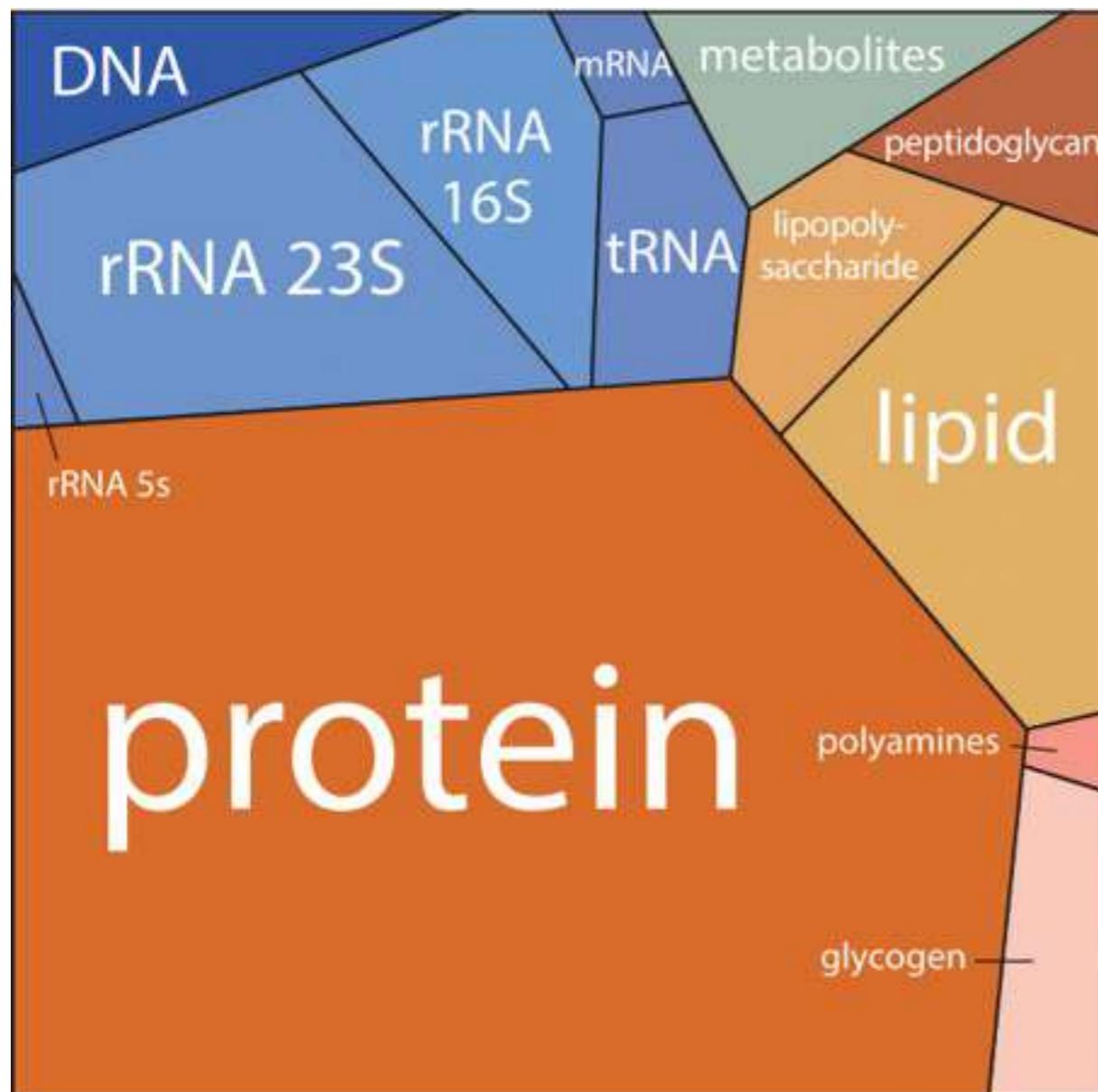
Growth rate (μ): the per-capita change in abundance (N) over time, or the slope of $\ln(N)$ versus time in the absence of mortality

Generation time (g): the time necessary for a population to double [$g = \ln(2)/\mu$], also referred to as doubling time; turnover time is mathematically equivalent to generation time

- ***Vibrio natriegens*** is capable of doubling every 7 min equivalent to a growth rate of over 140 d^{-1}
- ***Pelagibacter ubique***, a representative of the most abundant bacterial clade in the ocean, SAR11, has a generation time of nearly 2 days and growth rates of $0.4\text{--}0.6 \text{ d}^{-1}$ in laboratory pure cultures

Microbial Growth III

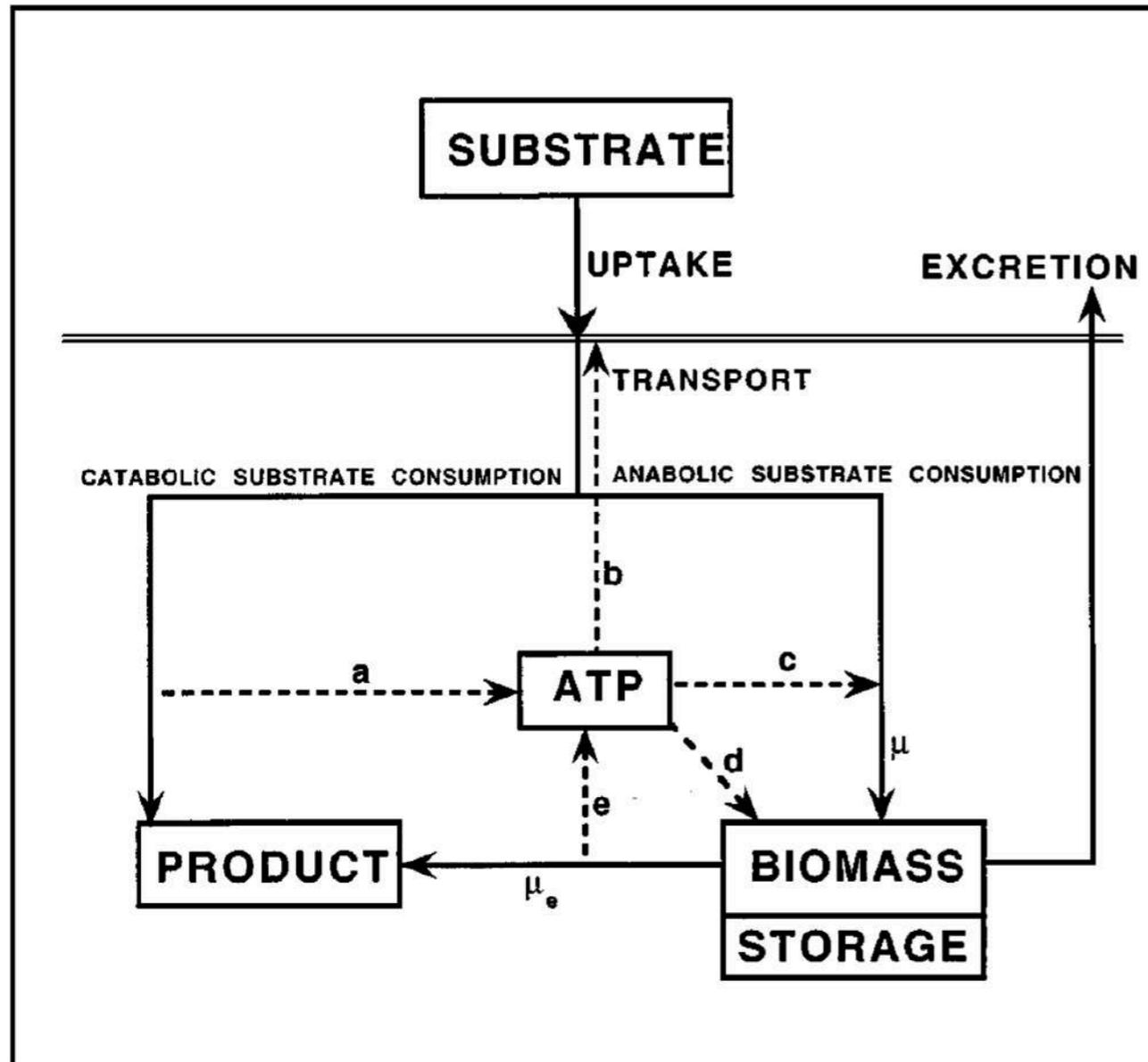
A Voronoi tree diagram of *E.coli* composition



40 min

- Each polygon area is the relative fraction of the corresponding cellular constituent (dry mass)
- Similar colors = related functional role
- Steady-state mean cell size (large circles) scales exponentially with nutrient-determined growth rate

Microbial Growth IV

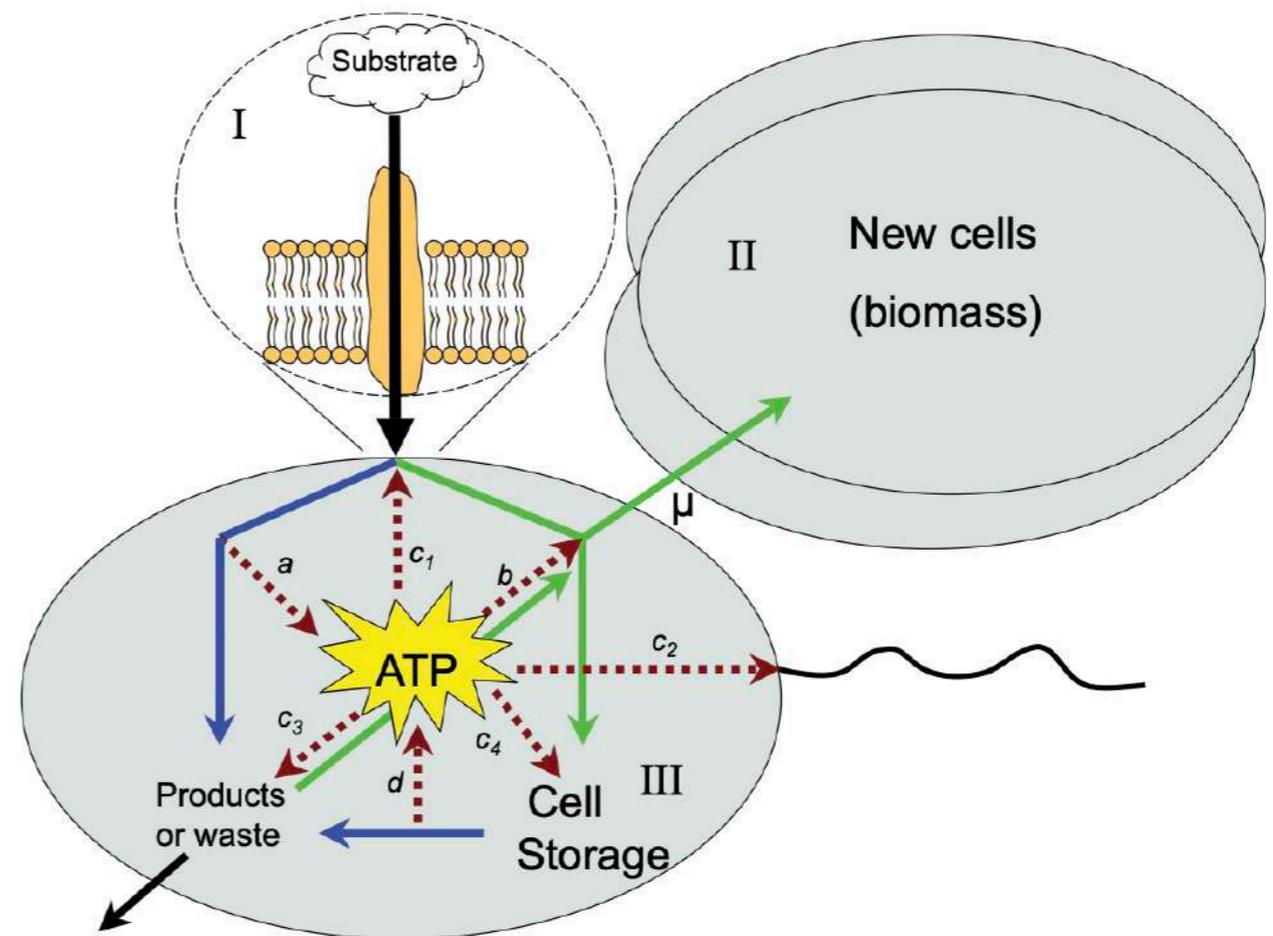


- The oxidation of organic compounds contributes to the energy pool as ATP at a rate ***a***
- Active transport of substrates into the cell requires energy from this ATP pool at a rate ***b***
- Anabolic reactions utilize ATP at a rate ***c*** and result in a growth rate μ
- The anabolic pathways result not only in increases in biomass but also in storage products and organic compounds that may be excreted back to the medium
- Maintenance expenditures consume ATP at rate ***d***
- In the absence of exogenous substrates, minimum maintenance energy requirements must be supported by degradation of biomass through endogenous metabolism (μ_e), which supplies ATP at a rate ***e***

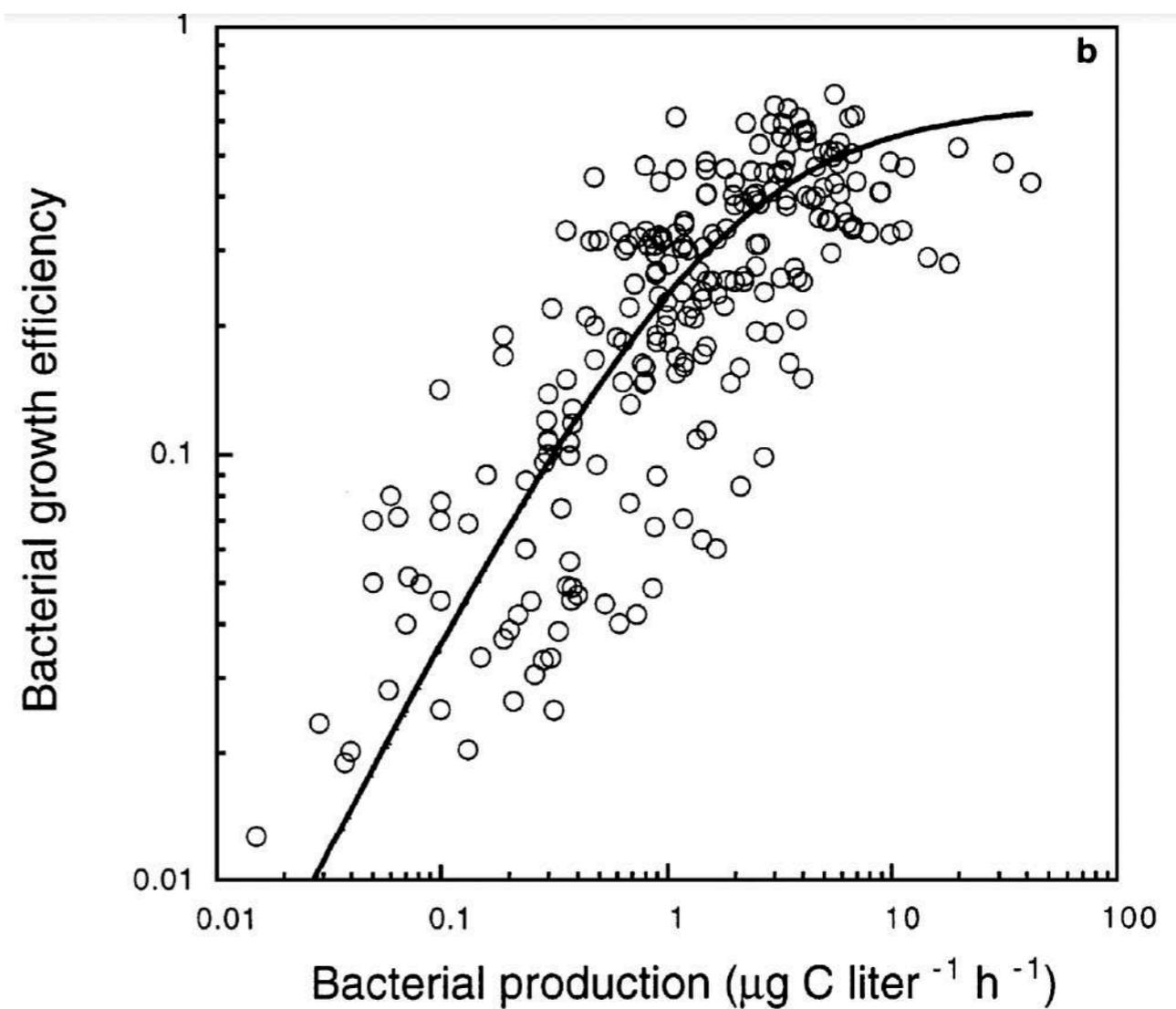
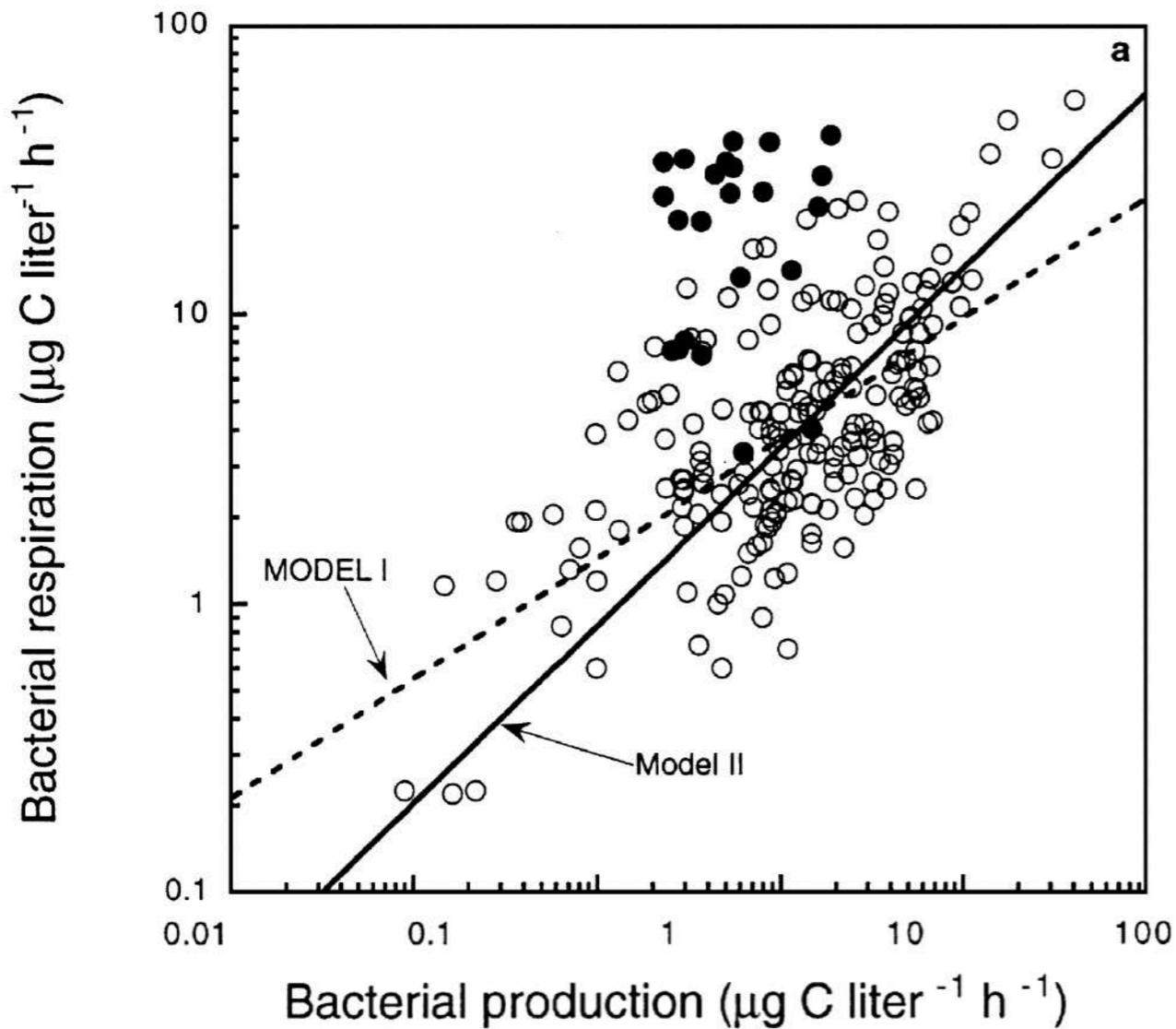
Endogenous metabolism is defined here as the state when no growth is possible, and by definition BGE is 0 under these conditions

Microbial Growth V

- Substrate < 700 Da are actively taken up via membrane proteins (I)
- As the substrate enters the cell via active uptake, it either enters into **catabolic pathways (blue lines)** or **anabolic pathways (green lines)**
- Monomers for anabolism can come preformed from the environment or as products of catabolism
- The red-hashed lines represent the flow of energy to and from these metabolic pathways. **Energy is conserved via substrate catabolism and ATP is produced at a rate a**
- As ATP is hydrolyzed, energy is released and utilized at rate b to drive anabolic processes such as production of new cells (growth; II) and production cell storage products (III)
- Energy is also utilized at various rates to support processes that are independent of anabolism. This maintenance energy is used at rates c1 to activate uptake systems, c2 to fuel cell motility, c3 to actively eliminate waste, and c4 to repair cellular machinery
- In the absence of exogenous organic substrates, the cell can yield ATP at rate d by catabolizing storage material (endogenous substrates)



BP, BR, BGE relationship



BGE

- The amount of biomass produced per unit of organic C consumed is the **bacterial growth efficiency (BGE)** (Sherr and Sherr, 1996; del Giorgio and Cole, 1998)
- BGE is a measure of the coupling between **catabolic (energy-yielding) reactions to anabolic (biosynthetic; energy requiring) reactions** and is expressed by the formula: **$BGE = BP / (BP + BR)$**
- **BP is bacterial production**
- **BR is bacterial respiration**
- BP was positively and significantly correlated to primary production (PP) across a wide range of aquatic ecosystems, and concluded that net BP averaged about <20% - 30%-50% of PP
- **The flux of carbon needed to support a given estimate of BP is referred to as bacterial carbon demand (BCD)**. It is possible to derive BCD, and thus total carbon consumption (BR plus BP), by combining measurements of BP with estimates of BGE, such that, **$BCD = BP / BGE$**
- **From cells to C per cell: 12.6 fg C cell⁻¹**

Microbial life is tough !

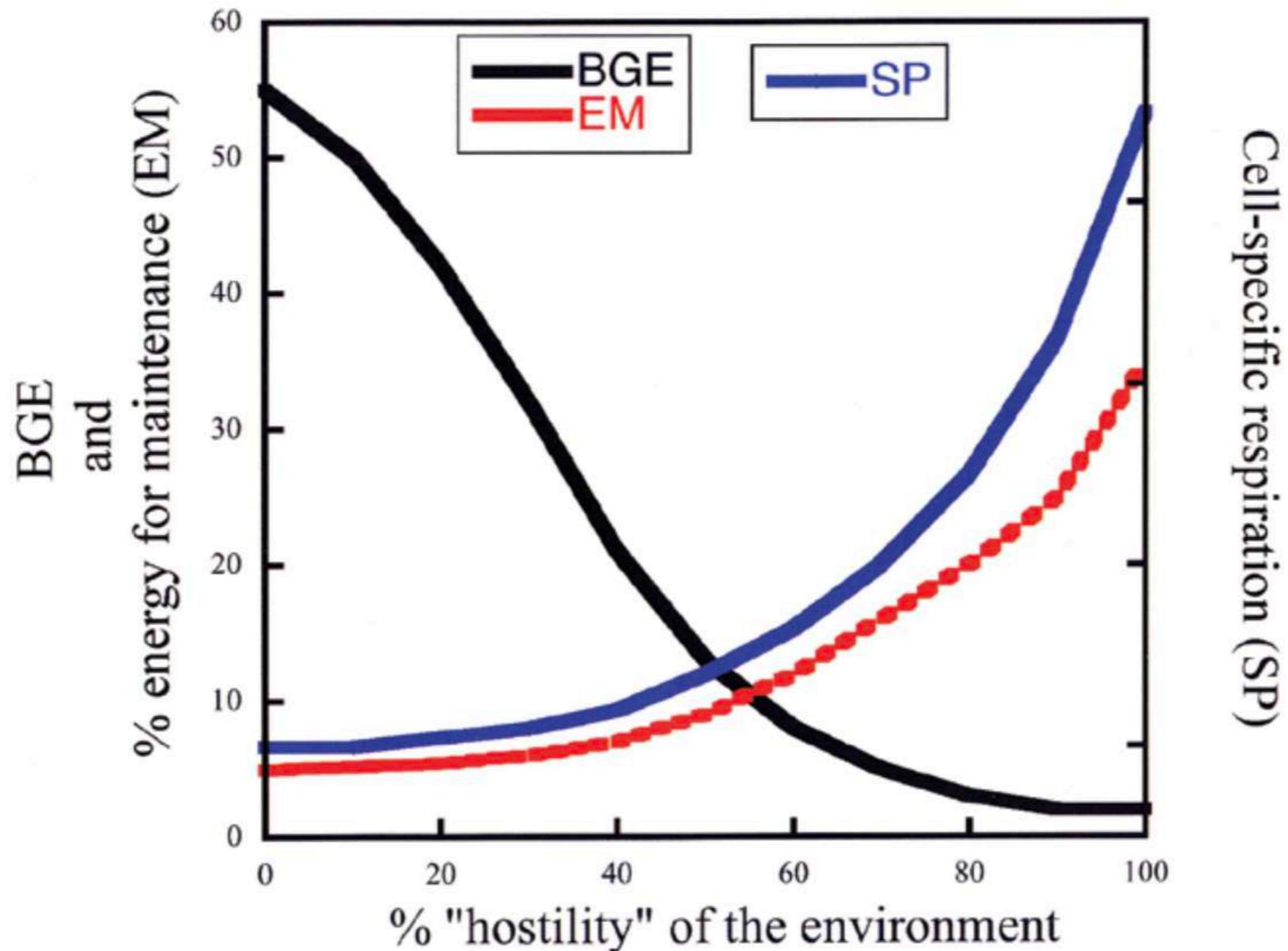
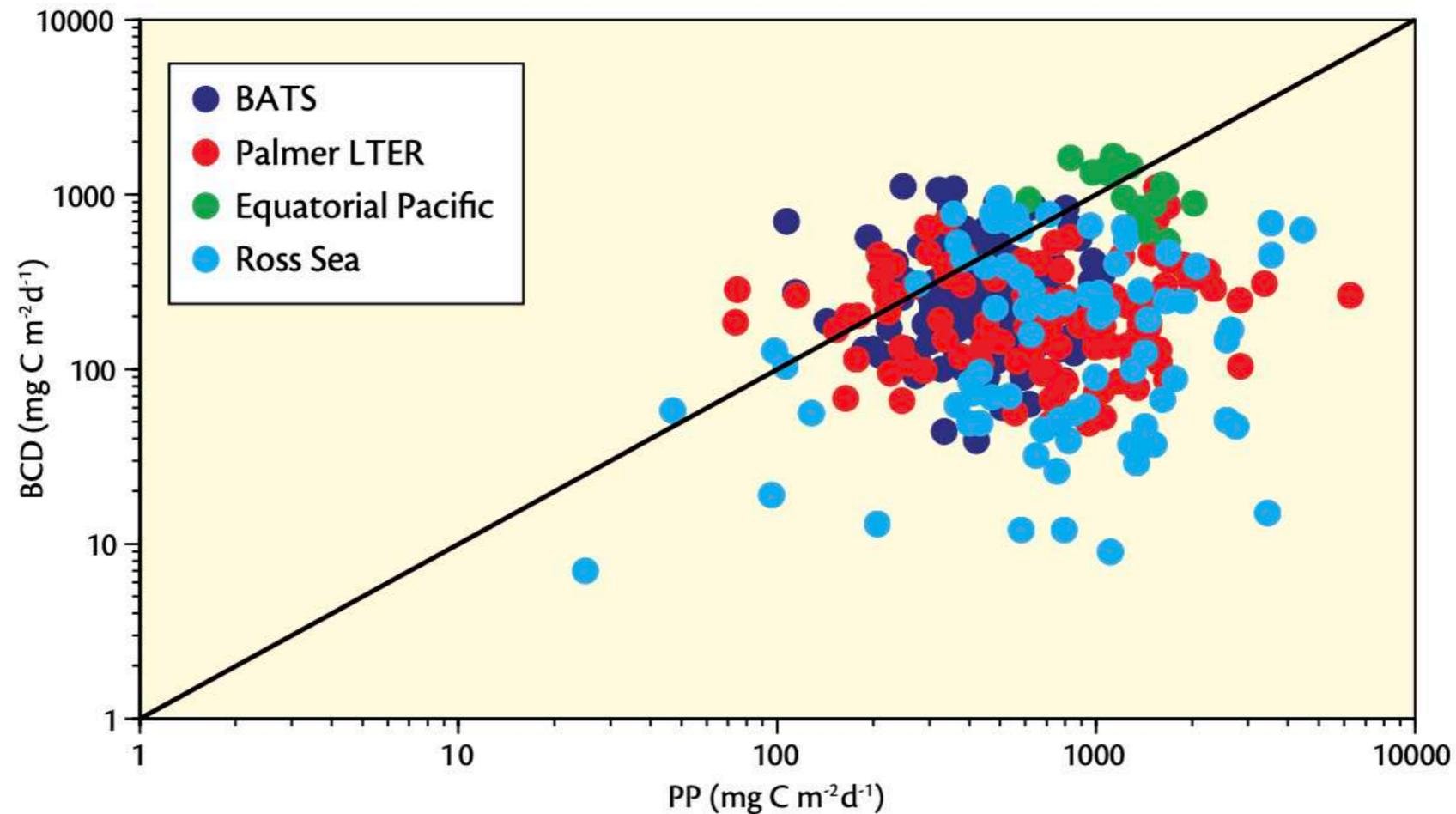


Figure 3. Conceptual diagram demonstrating the relationship between environmental stressors or environmental "hostility" and the partitioning of energy within a bacterial cell, the resulting bacterial growth efficiency (BGE), and cell specific respiration. As environmental hostility increases, more energy is partitioned into maintenance energy (EM). Thus, bacterial growth efficiency decreases and cell-specific respiration (SP) increases. Some combination of both physical (temperature, pH, salinity) and chemical (toxins, substrate availability) factors contribute to environmental hostility

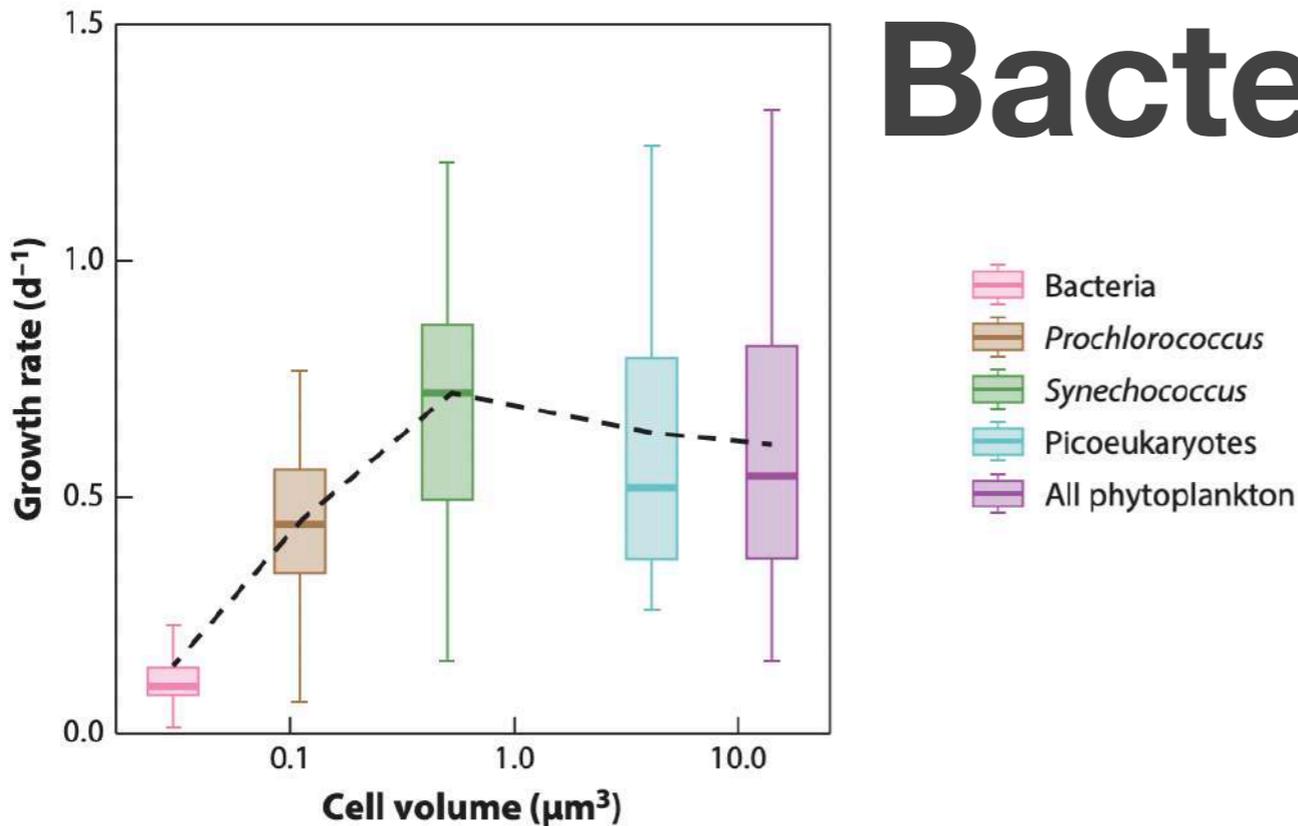
BCD and PP relationship



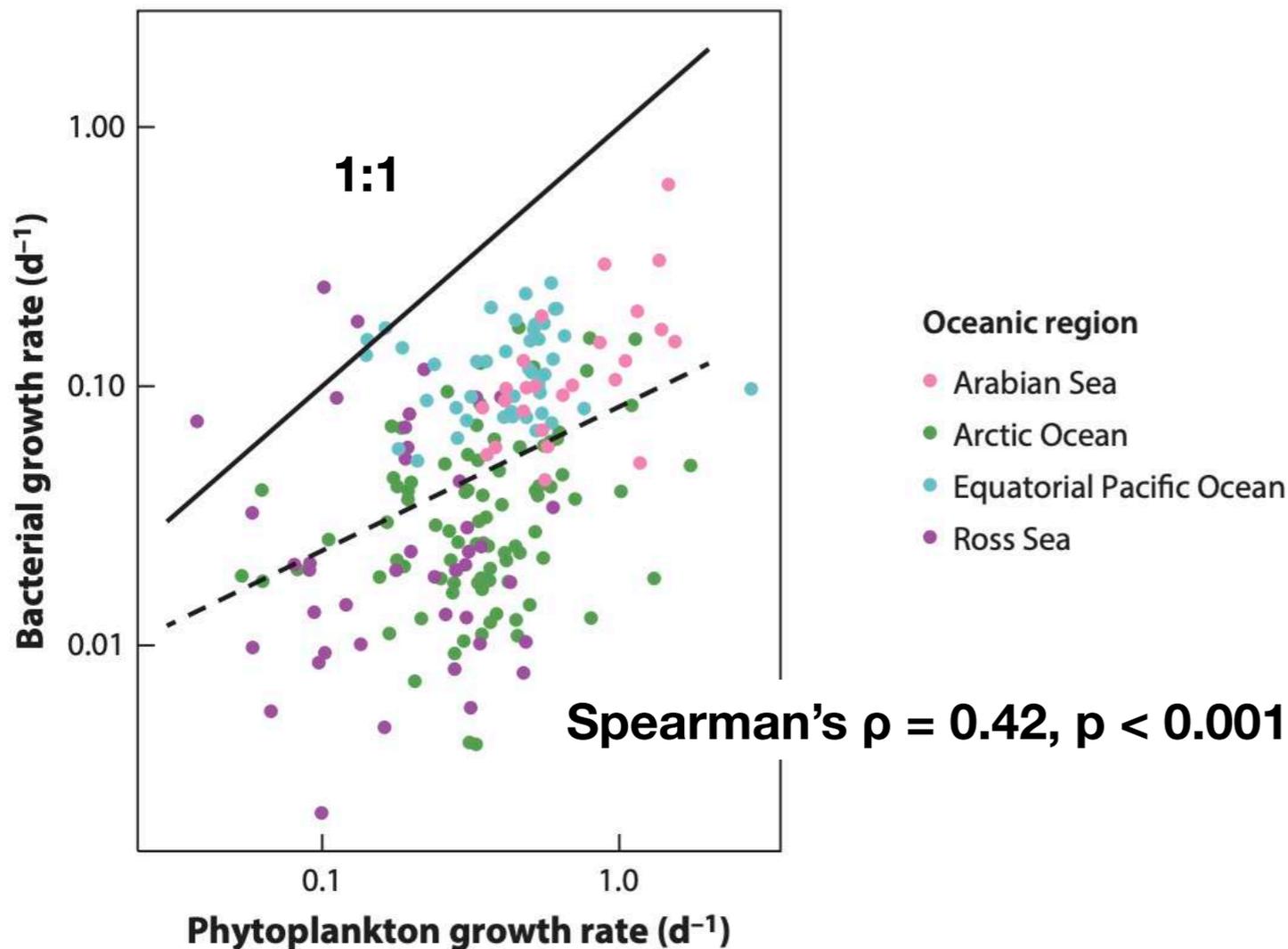
Carlson et al. 2007

Figure 4. Relationship between integrated bacterial carbon demand (BCD) and primary production (PP) within the euphotic zone of representative ocean sites. All data are derived from paired measurements of bacterial production and PP integrated within the euphotic zone from each study site. A common bacterial growth energy of 0.1 was used to estimate BCD. The black line represents the 1:1 line. Data points that lie above this line indicate that bacterial carbon demand was greater than local primary production at the time of sample collection. The Bermuda Atlantic Time-series Study (BATS) data represent monthly values from 1991–2003 ($n = 155$; see Steinberg et al., 2001 for details; data available at <http://bats.bbsr.edu/>). Paired BP and PP from the Equatorial Pacific ($n=16$) and Ross Sea, Antarctica, ($n=77$) were calculated according to Ducklow (1999) (data available at <http://usjgofs.whoi.edu/jg/dir/jgofs/>). All data from the Palmer Peninsula, Antarctica, ($n=112$) provided by H. Ducklow and the Palmer LTER program.

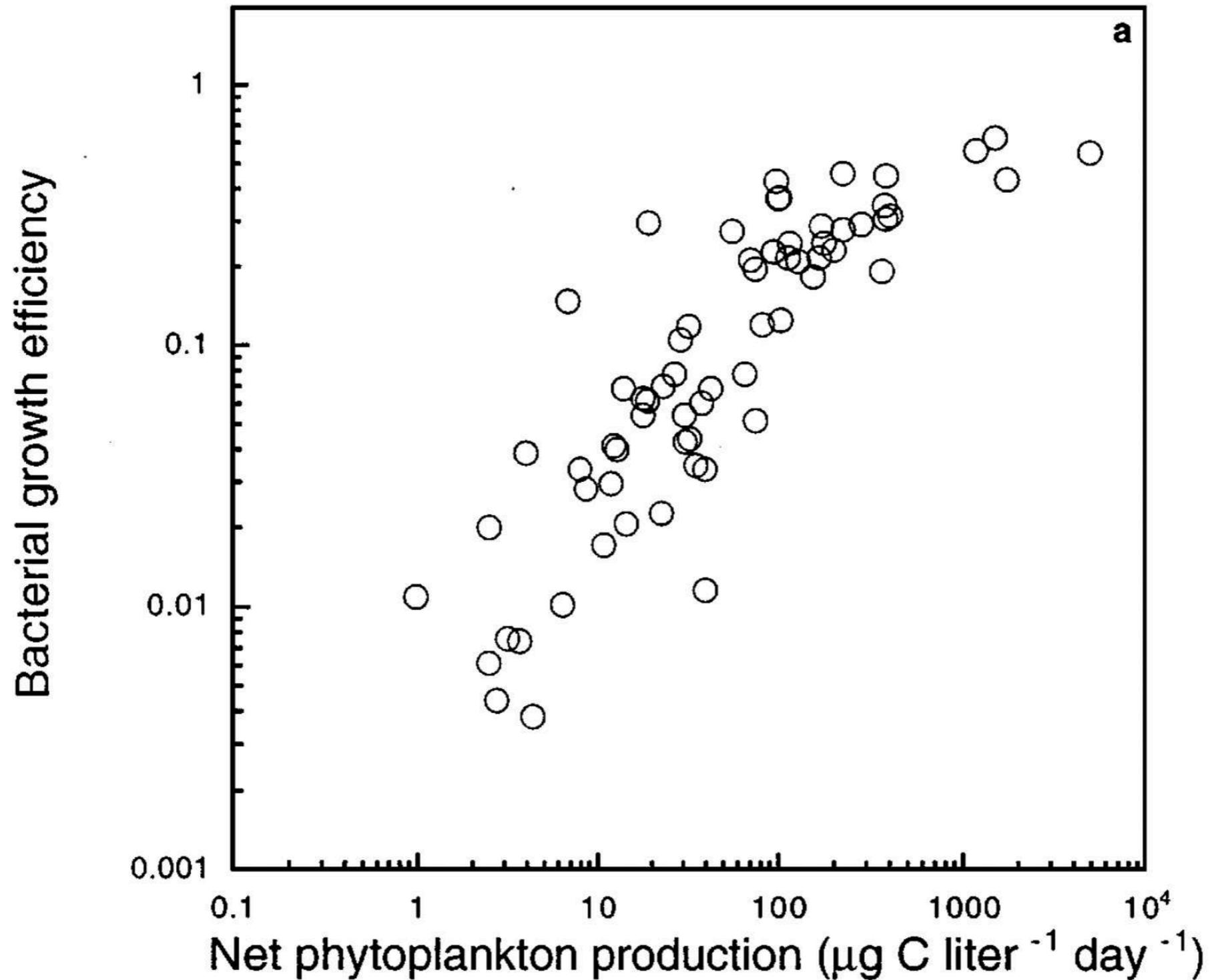
Bacteria-phytoplankton



- Heterotrophic bacteria $\mu <$ than Cyanobacteria and phytoplankton
- Bulk growth rate of the heterotrophic bacterial community is approximately 1/6 of the the phytoplankton community growth rate
- Phytoplankton rates can constrain growth rates of heterotrophic bacteria
- Bacterial growth rate is correlated with phytoplankton growth rate



BGE and PP relationship



Oligotrophic-copiotrophic life style

Table 2 Characteristics of oligotrophic and copiotrophic bacteria in the oceans

Property	Oligotrophs	Copiotrophs	Comment
Maximum growth rate	<0.2 d ⁻¹	>1 d ⁻¹	See main text
Variation in growth	Small	Large	See main text
Abundance	High	Low	
Genome size	Small	Large	Divide at 2 Mb?
Versatility	Low	High	E.g., organic carbon use
Hydrolases	–	+	E.g., chitinase
Periplasmic proteins	++	+	E.g., transporters
Siderophores	–	+	
Motility	–	+	
Chemotaxis	–	+	
Quorum sensing	–	+	
CRISPR/Cas	–	+	Phage defenses

The –, +, and ++ indicate the absence, presence, and enhanced presence of the indicated property, respectively. Table based on studies by Lauro et al. (2009) and Yooseph et al. (2010) and arguments presented in the main text.

The Biological Productivity of the Ocean

- **Primary productivity** is calculated by measuring the uptake of CO₂, or the output of O₂
- Production rates are expressed as grams of organic carbon per unit area per unit time
- Grams of carbon per meter squared per year is used: g C m⁻² yr⁻¹.
- The annual primary productivity of the oceans is estimated to be approximately 50 × 10¹⁵ grams (50 × 10⁹ metric tons) of carbon per year, roughly half of the global total primary production
- The energy they produce is called **primary production**, and the rate at which they produce the energy is called **productivity**
- **Light**
- **Nutrient availability**
- **Temperature**
- **Predator and competitors**

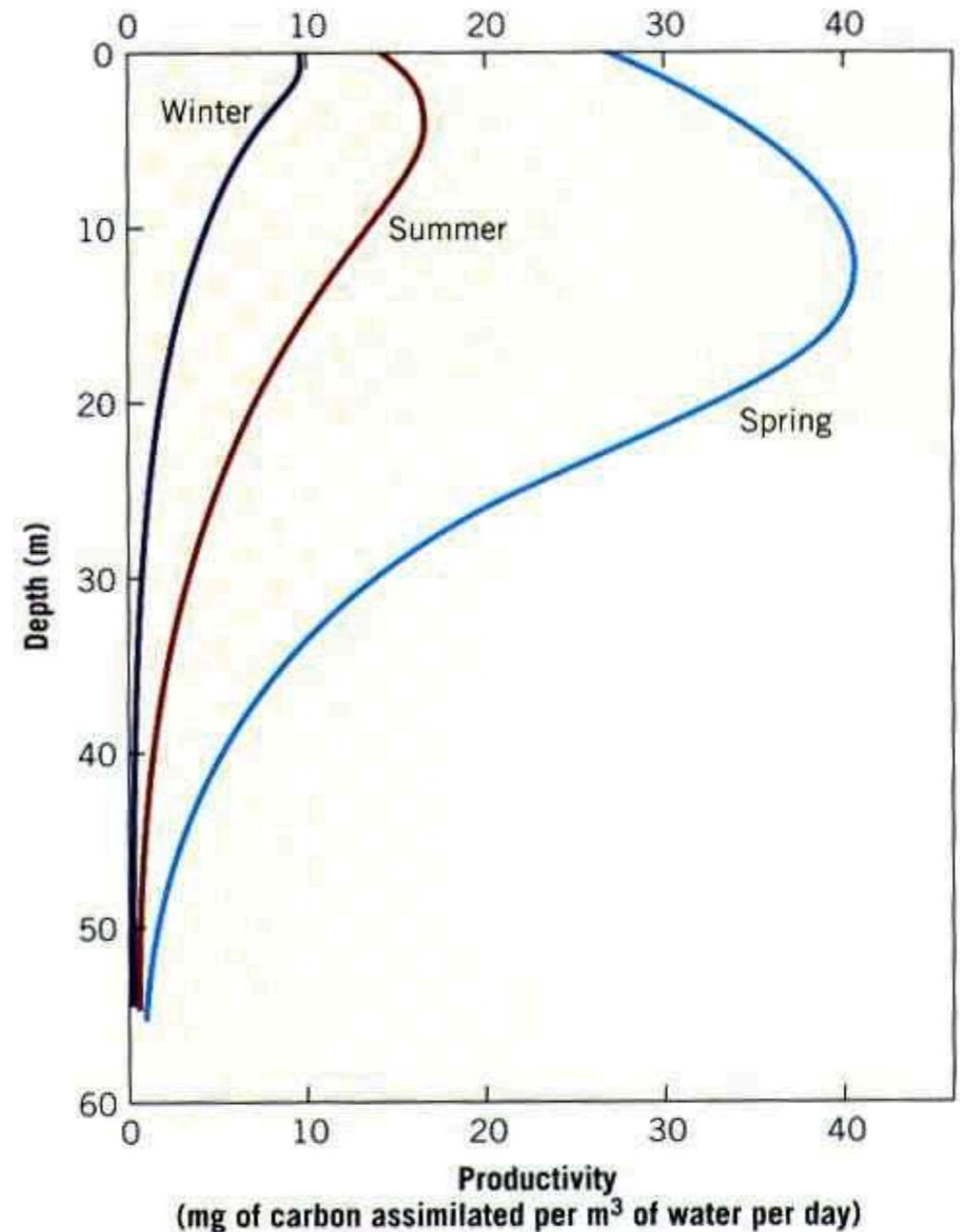


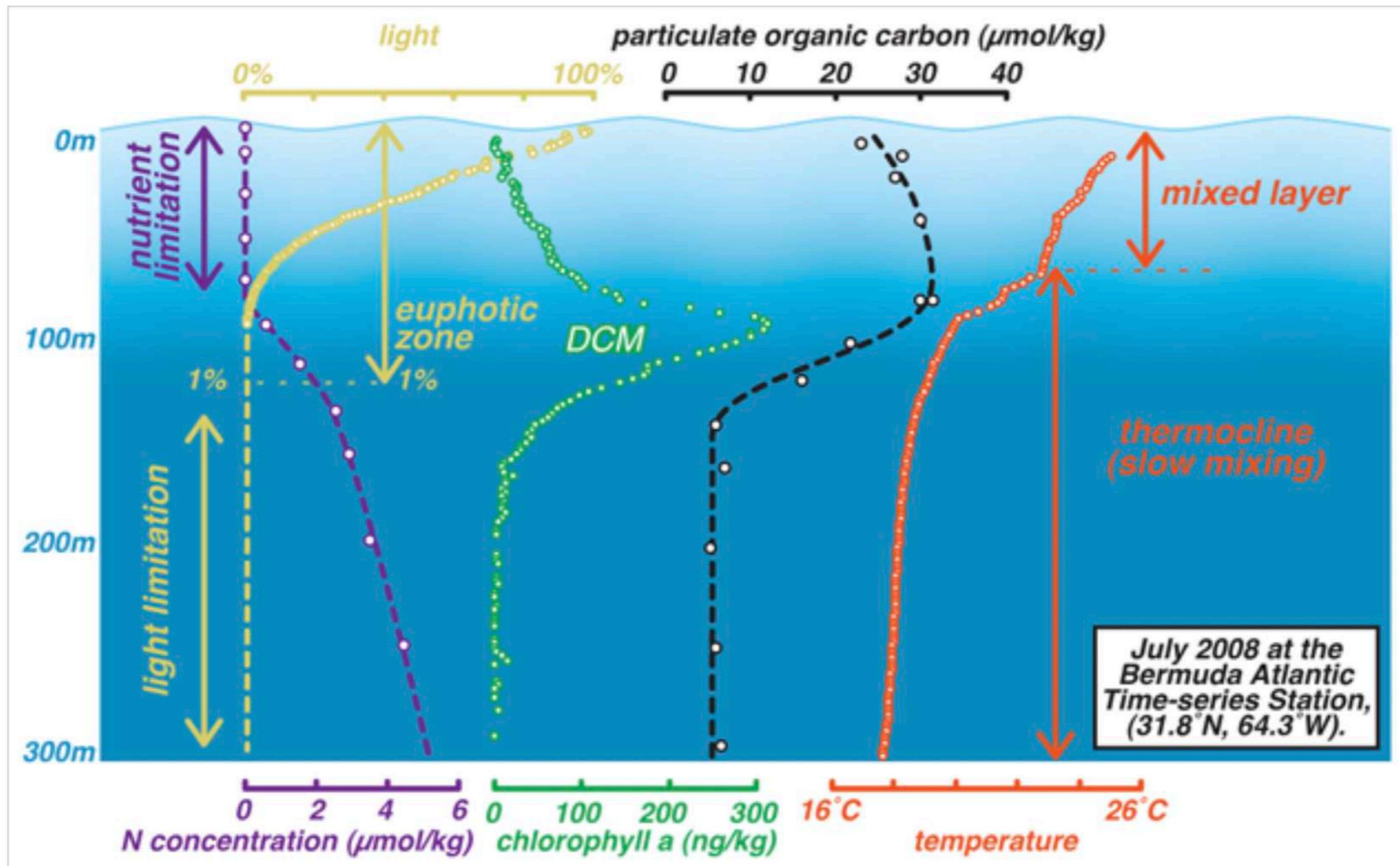
Figure 11.12

Typical variations in primary productivity with depth in the coastal waters of the Northeast Pacific Ocean. (Based on data from G. C. Anderson, 1964, *Limnology and Oceanography*. **9**; 294.)

GPP-NPP

“Gross primary production” (**GPP**) refers to the total rate of **organic carbon** production by **autotrophs**, while “**respiration**” refers to the energy-yielding oxidation of **organic carbon** back to carbon dioxide

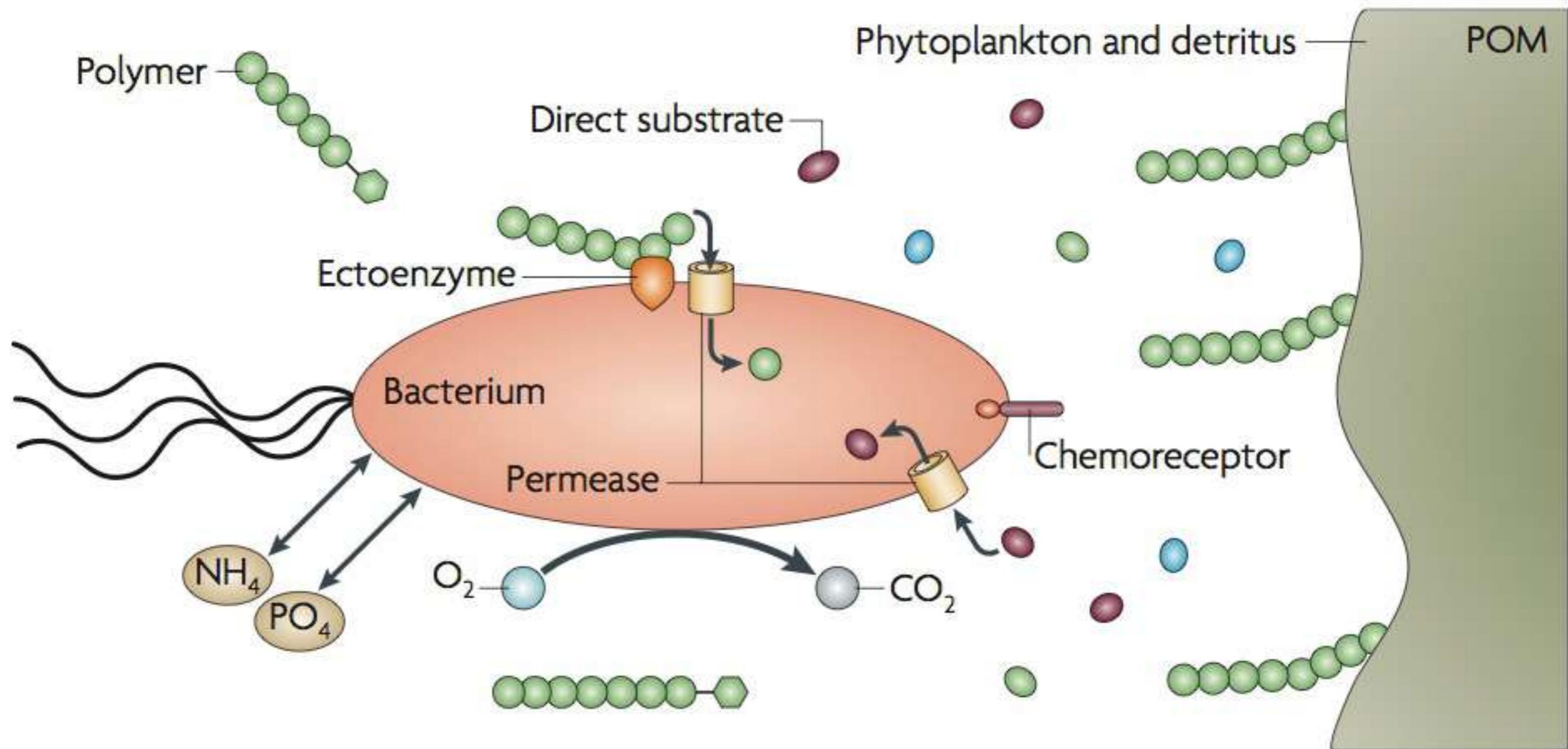
“Net primary production” (**NPP**) is **GPP** minus the **autotrophs'** own rate of **respiration**; it is thus the rate at which the full metabolism of **phytoplankton** produces biomass



Adaptive strategies of microbes in the ocean

- Two-component system
- Motility
- Hydrolysis-uptake coupling
- Genome architecture

Adaptive strategies of heterotrophic bacteria in the ocean



by Farooq Azam

Azam and Malfatti, 2007 Nature Reviews Microbiology 10:782

- Motility, environmental sensing, permeases and cell-surface hydrolases
- Adapted fine biochemical strategies to interact with organic matter natural and human-created