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# **Microbial Ecology**

- Study of the interactions of microorganisms with their environment (including organic matter), each other, and plant and animal species (other organisms)
   —> symbioses, biogeochemical cycles, climate change
- Marine sediment ecosystem
- Ocean ecosystem
- Freshwater ecosystem

### **Microbial abundance**



Spheres

# MARINE SEDIMENTS OCEAN FLOOR

# Marine sediments as a microbial ecosystem

- Global view of Earth: a heterogeneous mixture of rocks, water, gases, organisms is maintained in a non equilibrium state by sunlight and volcanism
- Hydrothermal vents, cold seeps are oasis of primary production and microbial and biochemical diversity



Vadigan et al. 2018



- Approximately 1x10<sup>9</sup> microbes in 1 g of upper sediment
- Biofilm

### Microbial metabolic pathways shaping Earth ecosystem



**Figure 20.20** Diversity of marine systems and associated microbial metabolic processes. Decreasing electron acceptor availability with depth into the sediment or with increasing distance into an oxygen minimum zone is indicated by red wedges. Sulfate becomes limiting only at greater depths in marine sediments. The indicated metabolic diversity is covered in Chapter 14.

Madigan et al. 2018

## **Terrestrial** *vs* marine fingerprint



**Figure 4.11** Composition of ocean sediments: percent derived from continents (terrigenous) versus autochothonous biotic sources. The six boxes in the key (numbered 1–6) show values from <10% (1, lightest shading) to >90% (6, darkest shading) terrigenous sediment. (From Lisitzin, A.P. 1996. *Oceanic Sedimentation*, pp. 28–29. American Geophysical Union, Washington, DC. Copyright 1996, American Geophysical Union. Reproduced by permission of American Geophysical Union.)

- Marine sediments are influenced by terrestrial ecosystem and water column ecosystem
- Marine sediments: clay-silt-sand
- Marine sediments are very dynamic

#### **Ocean surface photosynthesis dictates** sedimentation



Marine snow

- Chlorophyll data from satellite to inform on photosynthesis —> organic matter production by microscopic algae (phytoplankton) and cyanobacteria
- Life and death in the upper ocean • create a down-ward flux of organic matter and associated minerals



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More

ANME archaea Facultative anaerobes (bacteria and fungi) Chloroflexi 'Ca. Atribacteria' 🚫 Methanogens 8

OATZ

SMTZ

Strict aerobes

Sulfate reducers

🔪 Thaumarchaeota

SO,2

Less

More

## Sedimentation rate

- Redox conditions in seafloor and subseafloor ecosystems are linked to sedimentation rate and distance from shore
- Ultra-low rates of organic matter sedimentation under oligotrophic gyres lead to deep O<sub>2</sub> penetration (left), and consequently, the survival of microbial communities is limited by availability of electron donors from organic matter
- Faster sedimentation rates closer to shore cause anoxic conditions shortly below the seafloor owing to the rapid consumption of oxygen for organic matter oxidation at the sediment surface (right), terminal electron acceptors—> sulfate



### Sediment: Organic matter decomposition/respiration —> nutrient recycling

 Microbes able to respire in multiple ways will always choose available acceptors with the biggest potential difference to the donor







- Microorganisms (purple) in the upper layers of marine sediments use oxygen (O<sub>2</sub>) that diffuses from sea water as an acceptor of electrons, which they produce in energygenerating metabolic reactions. As a result, other microbes (orange) in deeper, anoxic layers (where oxygen is scarce or absent) have to use other electron acceptors such as sulphate (SO<sub>4</sub><sup>2-</sup>) for growth. Transfer of electrons to oxygen results in the formation of water, whereas electron transfer to sulphate produces hydrogen sulphide (H<sub>2</sub>S), which is poisonous to many organisms
- Long bacterial filaments could transport electrons generated when hydrogen **sulphide is converted into sulphur (S)** at the bottom of the sediments and use them to consume oxygen in the upper layers
- During classical sulfur (S) oxidation, bacteria consume both O2 and H2S, and, as a result, both half-reactions of the overall redox reaction occur in the same location
- During electrogenic sulfur oxidation, long-distance electron transport (LDET) allows multicellular cable bacteria to harvest H<sub>2</sub>S in deeper horizons of the seafloor, while still keeping access to O<sub>2</sub>, which is available only in the top millimetres of the sediment. By separating the two half-reactions in space, cable bacteria can outcompete classical sulfide oxidizing bacteria

# Microbial abundance and diversity in the sediment

- Structured 3D environment at high pressure at low temperature
- Hydrothermal vents, cold seeps, brines, carcasses as oasis of metabolic diversity (e.g. symbiosis, temperature, relatively fresh organic matter)







# SEAWATER: COASTAL And OPEN OCEAN

### Key features related to the ocean microbiome



Tara Ocean Foundation et al., 2023

#### Upper ocean water column

#### SeaSCAPES microbial dynamics





Malfatti, Jaffe, Countway

### Fate of fixed carbon in ocean ecosystem



#### Seasonality —> high and low production

#### Marine microbial environment: impressionistic painting



Azam, F. 1998 Science 280:694-696

## Marine microbial microenvironment



• Disparate processes contribute to make the ocean a sea of gradients, from the vantage point of microorganisms

Stocker, 2012

## Microbial abundance in ocean

- Photic zone 0-200 m
- More than 96% of ocean is dark and with a constant temperature (~ 4°C)
- Microbes in coastal ocean 1X10<sup>6</sup> cells/mL
- Archaea dominate at depth
- Primary production sustain ocean interior > organic matter degradation and sediment too!









Bacteria/Archaea represent 90% the total biomass in the ocean Bacteria/Archaea are the most productive organisms in the ocean and contribute up to 90% to biota 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)

#### Ocean: Organic matter decomposition/respiration —> nutrient recycling

- Microbial degradation of organic matter in the water column generate nutrient fluxes available for primary producers (limiting nutrients based on cell demands)
- Microbial action with organic matter at the microscope structure ocean-basin scale



#### Seasonality in the ocean: factors affecting the mixed layer depth and C fixation in the food web and C biogeochemical cycle

heating, precipitation and slight winds



Ganachaud et al., 2011

Influence of the thermocline and mixed layer depth on transfer of nutrients to surface waters for C fixation into organic matter by photoauthotrophs



Ganachaud et al., 2011

# Seasonality in C fixation



https://datalab.marine.rutgers.edu/ooi-lab-exercises/lab-7-identifyfactors-that-control-primary-production-in-the-western-temperateatlantic-ocean/lab-7-instructor-guide/

#### Organic matter continuum: a unifying concept to understand how bacteria perceive and interact with organic matter



- Ocean is replete with transparent gel particles (~ million/liter) creating a gel-like milieu
- Primary production is the source of the great chemical and structural complexity

# Microbial adaptive strategies for growth at small scale



Azam and Malfatti, 2007 Nature Reviews Microbiology 10:782

- Motility, environmental sensing (interacting with molecules)- MCPs
- Significance of spatial coupling hydrolysis-uptake (permease) on the cell
- Cell surface hydrolases; 10<sup>2</sup>-10<sup>4</sup> x variability in cell-specific activity
- Coordinated behavior and biochemistry in space and time

### Nutrient patch evolution and sources



- Low Reynolds number —> high viscosity
- Modelling in time and space in a turbulent flow (L= 5.65 cm)
- Nutrient sources: living phytoplankton, dying phytoplankton, dying copepods, marine snow plume, fecal pellets

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# Marine bacteria can exploit ephemeral nutrient patches



- Motility as an adaptive strategy to respond to nutrient patches
- Exploration: non motile 0.45 pL (Brownian motion) vs motile 0.5  $\mu$ L every (motility 50  $\mu$ m/s) 10 min
- Daily exploration: non motile ~ 430  $\mu$ m cube vs motile ~ 1 cm cube chemotactic
- Copiotrophic populations outcompete nonmotile, oligotrophic populations during diatom blooms and bloom collapse conditions (more motile cells)

## **Bacteria-phytoplankton interactions**



- The phycosphere, defined as the region surrounding a phytoplankton cell that is enriched in organic substrates exuded by the cell, is an important microenvironment for planktonic aquatic bacteria
- The phycosphere is defined as the region where the concentration is >50% above background

## Ocean & sea ice diversity



## Polar winter

- Primary producers (microalgae, Euk) experience from lower or none solar irradiance
- Sea Ice formation
- Shift from autotrophy to mixotrophy
- Forming spores or resting structure
- Slowing down metabolisms and using storage reserve
- Producing cryoprotectant molecules to protect from freezing damage and desiccation
- Heterotrophic prokaryotes are active but slow



Circles of yellow (left) indicate duration of the midnight sun period. Circles of blue (right) indicate duration of the polar night period with illumination at winter solstice defined as polar twilight (outer), civil polar night (middle), and nautical polar night (inner). Illustration from ScienceAdvances

> https:// norwegianscitechnews. com/2018/01/sheddinglight-zooplankton-dark/

### **Biosphere model**



# FRESHWATER: LAKE RIVER

# Hydrological cycle-soil-subsurface



Madsen, 2016

**Figure 4.12** Conceptual flow system for understanding the role of soil and subsurface habitats in the hydrologic cycle. (Reprinted from Madsen, E.L. 1995. Impacts of agricultural practices on subsurface microbial ecology. *Adv. Agron.* **54**:1–67. Copyright 1995, with permission from Elsevier.)

- Water connects upper microbiomes (soil) with subsurface microbiome
- Lower microbiome at ~ 4 km at 125°C
- Underneath there are saturated and unsaturated subsurface zone
- Subsurface zone associated microbiome is important in dictating chemical composition of the water flowing in it (beside the influence of the soil and geological strata)

About **30%** of all freshwater is **terrestrial ground water**, whereas the world's **lotic (streams and rivers)** and **lentic (lakes)** systems contribute only **0.3%** 

About 2% of Earth's water resources occur as groundwater, of which half is saline and the other half fresh

Groundwater flow is governed largely by **recharge and discharge rates** and the **hydraulic properties of the saturated rocks** 

Groundwater-flow paths are derived from **hydraulic gradients between areas of recharge** (e.g., precipitation in high topographic regions) **and discharge** (e.g., springs in low topographic regions)

# Groundwater environment are characterised by continuous darkness and limited nutrients —> slow growing microbes

Groundwater ecosystems deliver services that are of immense **societal** and **economic value**, such as:

- 1) **purification** of water and its storage in good quality for decades and centuries,
- 2) active **biodegradation** of anthropogenic contaminants and inactivation and elimination of pathogens,
- 3) nutrient recycling,
- 4) mitigation of floods and droughts

### Groundwater connection with ocean

#### Diffusive seepage through unconsolidated sediments

Point-sourced discharge, as in karstic or volcanic aquifers



#### Ruiz-González et al., 2021

Groundwater direct connection to the ocean through permeable sediments or rocks allows the discharge of groundwater, a process known as submarine groundwater discharge, but also the entrance of seawater

#### Aquatic surface and subsurface microbial ecosystems



Arrows depict the flow of water carrying energy and matter through the subsurface, with boxes next to arrows indicating typical groundwater residence times.

Circles highlight transition zones between habitat types. Curly braces indicate the distribution of different microbial groups in the subsurface. CF, capillary fringe;GWT, groundwater table

#### Low microbial abundance in low C system

Group	Habitat	Contamination	Abundance (cells cm <sup>-3</sup> )	Reference
Prokaryota				
Bacteria	Water from karst and cave systems	No	$10^2 - 10^5$	Gounot (1994); Farnleitner et al. (2005)
	Sediment from cave waters	No	$10^4 - 10^8$	Gounot (1994); Rusterholtz & Mallory (1994)
	Water from granite and basalt systems	No	$10^2 - 10^5$	Stevens & McKinley (1995); Pedersen (1997)
	Ground water	No	$10^{3}-10^{6}$	Ghiorse & Wilson (1988); Madsen &
		Yes	$10^{3}-10^{7}$	Ghiorse (1993); Pedersen (2000); Griebler (2001)
	Groundwater-saturated	No	$10^{5} - 10^{8}$	
	porous sediment	Yes	up to 10 <sup>10</sup>	
	Vadose zone sediment	No	$10^4 - 10^8$	Brockman et al. (1992); Kieft et al. (1993)
Archaea		Yes or no	up to 20% of total cell counts	Detmers et al. (2004)
Protozoa				
Heterotrophic Flagellata	Ground water	No	$10^{0}-10^{2}$	Hirsch <i>et al.</i> (1992); Madsen & Ghiorse (1993); Novarino <i>et al.</i> (1997)
		Yes	up to $10^5$	Novarino et al. (1997): Zarda et al. (1998)
	Groundwater-saturated	No	$10^{3}-10^{5}$	Novarino et al. (1997)
	porous sediment	Yes	up to 10 <sup>8</sup>	Novarino et al. (1997)
Amoebae	Ground water	Yes or no	$10^{-1} - 10^{1}$	Hirsch <i>et al.</i> (1992); Madsen & Ghiorse (1993); Novarino <i>et al.</i> (1997)
Ciliata Heliozoa	Ground water (near surface) Ground water (near surface)	Yes or no No	$10^{-1} - 10^{0}$ $10^{-1} - 10^{0}$	provences and an

 Table 2
 Abundance of microbial groups in different subsurface habitats

© 2008 The Authors, Journal compilation © 2008 Blackwell Publishing Ltd, Freshwater Biology, 54, 649-677

### Large marine ecosystems and watersheds

- On land watersheds don't comunicate with other watersheds
- Characteristic chemical and biological fingerprint of each watershed



### Lake as a complex microbial ecosystem



**Figure 4.5** Major lakes of the world showing an approximate comparison of the surface areas of many of the larger inland waters, all drawn to the same scale. The Aral Sea has experienced catastrophic reductions in area (more than half of that depicted here) because of diversion of water for agriculture. (Reprinted from Wetzel, R.G. 2001. *Limnology: Lake and River Ecosystems*, 3rd edn. Academic Press, San Diego, CA. Copyright 2001, with permission from Elsevier.)

- Nutrient inputs from terrestrial vegetation and human activities drive microbial activities in production and decomposition of organic matter
- Microalgal bloom (thus including Cyanobacteria)
- Organic matter decomposition can create anoxia (lack of O<sub>2</sub>)

• Seasonality in the water column



# Steep microbial gradients in lake and sediment shape microbial community and metabolism



#### The redox potential is used to describe a system's overall reducing or oxidizing capacity

The redox potential is measured in millivolts (mV) relative to a standard hydrogen electrode and is commonly measured using a platinum electrode with a saturated calomel electrode as reference In well-oxidized water, with oxygen concentrations > ~1 mg O<sub>2</sub> L<sup>-1</sup>, the redox potential will be > 300–500 mV In reduced environments, such as in the deep water of stratified lakes or the sediment of eutrophic lakes, the redox potential will be < 100 mV or even negative

## Reduction potential ranges of microbial respiration

- The achievable energy yield of ETC depends on the difference in electrical potential between electron donor and acceptor
- Microbes able to respire in multiple ways will always choose available acceptors with the **biggest potential difference** to the donor (e.g., *E. coli* O<sub>2</sub> > NO<sub>3</sub>-> fumarate)





# Anaerobic respiration

#### Microbially mediated reactions

#### Microaerophiles

 $4Fe^{2*} + 10H_2O + O_2 \rightarrow 4Fe(OH)_3 + 8H^+$ Gallionella spp., Leptothrix spp., Mariprofundus spp., Sideroxydans spp.

#### Photoferrotrophs

 $\frac{\text{HCO}_3^- + \text{Fe}^{2+} + 10\text{H}_2\text{O}}{(\text{CH}_2\text{O}) + 4\text{Fe}(\text{OH})_3 + 7\text{H}^+}$ 

Rhodopseudomonas palustris TIE-1 Rhodobacter sp. SW2 Chlorobium ferrooxidans (KoFox) Thiodictyon sp. F4

#### NO3-reducing Fe(II)-oxidizers

 $10Fe^{2+} + 2NO_3^- + 24H_2O \rightarrow$ 10Fe(OH)\_3 + N\_2 + 18H^+

Acidovorax spp., KS, 2002 Thiobacillus denitrificans

#### Fe-ammox

Anaerobic respiration

 $NH_4^+$  + 6FeOOH + 10H<sup>+</sup>  $\rightarrow$  $NO_2^-$  + 6Fe<sup>2+</sup> + 10H<sub>2</sub>O

#### Unknown

#### Fe(III)-reducing organic C and/or H<sub>2</sub>-oxidizers

 $4FeOOH + CH_{3}CHOHCOO^{-} + 7H^{+} \rightarrow 4Fe^{2+} + CH_{3}COO^{-} + HCO_{3}^{-} + 6H_{2}O$  $2Fe(OH) + H_{2} \rightarrow 2Fe^{2+} + 2H_{2}O$ 

Geobacter spp., Shewanella spp. Albidoferax ferrireducens, Geothrix spp.



#### Microbial guilds and potential community functions



## Freshwater microbial diversity



• 16S rRNA gene based diversity

## **Biogeochemical processes**

	Table 1   Examples of physiological processes catalysed by microorganisms in biosphere habitats							
Process		Nature of process	Typical habitat					
	Carbon cycle							
	Photosynthesis	Light-driven CO <sub>2</sub> fixation into biomass	Ow, Fw, FwS, Og	s				
C respiration		Oxidation of organic C to CO <sub>2</sub>	All					
Cellulose decomposition		Depolymerization, respiration	SI					
	Methanogenesis	CH <sub>4</sub> production	Sw, FwS, Os					
,	Aerobic CH <sub>4</sub> oxidation	CH <sub>4</sub> becomes CO <sub>2</sub>	All	2				
Anaerobic CH <sub>4</sub> oxidation		CH <sub>4</sub> becomes CO <sub>2</sub>	Os					
				000				
	Biodegradation			Per				
	Synthetic organic compounds	Decomposition, CO <sub>2</sub> formation	All					
	Petroleum hydrocarbons	Decomposition, CO <sub>2</sub> formation	All					

SI, Sw, Gw

SI, Sw, Gw

SI, Sw, Gw

SI, Sw, Gw

As, arsenic; C, carbon;  $CH_4$ , methane;  $CO_2$ , carbon dioxide; Fe, iron; FeS, iron sulphide; Fw, freshwater; FwS, freshwater sediment; Gw, groundwater;  $H_2$ , hydrogen; Hg, mercury; Hg<sup>2+</sup>, mercuric ion; MTBE, methyl tertiary butyl ether; N<sub>2</sub>, nitrogen; NH<sub>3</sub>, ammonia; NH<sub>4</sub><sup>+</sup>, ammonium; NO<sub>2</sub><sup>-</sup>, nitrite; NO<sub>3</sub><sup>-</sup>, nitrate; Os, ocean sediments; Ow, ocean waters; S<sup>0</sup>, elemental sulphur; S<sup>2-</sup>, sulphide; SI, soil; SO<sub>4</sub><sup>2-</sup>, sulphate; Sw, sewage; U, uranium.

Compounds are dechlorinated through respiration in anaerobic habitats

Decomposition, CO<sub>2</sub> formation

Decomposition, CO<sub>2</sub> formation

Decomposition

Fuel additives (MTBE)

Chlorinated solvents

Pharmaceuticals, personal

Nitroaromatics

care products

## **Biogeochemical processes**

Nitrogen cycle			
N <sub>2</sub> fixation	N <sub>2</sub> gas becomes NH <sub>3</sub>	SI, Ow	
$NH_4^+$ oxidation	NH <sub>3</sub> becomes NO <sub>2</sub> <sup>-</sup> , NO <sub>3</sub> <sup>-</sup>	SI, Sw	
Anaerobic $NH_4^+$ oxidation	$NO_2^-$ and $NH_3$ become $N_2$ gas	Sw, Os	
Denitrification	$\mathrm{NO_3^-}$ is used as an electron acceptor and converted to $\mathrm{N_2}$ gas	SI, Sw	
Sulphur cycle			10
S <sub>2</sub> oxidation	$S^{2-}$ and $S^{0}$ become $SO_{4}^{2-}$	Os, FwS	200
SO <sub>4</sub> <sup>2-</sup> reduction	$SO_4^{\ 2^-}$ is used as an electron acceptor and converted to $S^0$ and $S^{2^-}$	Os, Sw, Gw	sen,
Other elements			lads
H <sub>2</sub> oxidation	$H_2$ is oxidized to $H^+$ , electrons reduce other substances	Sw, SI, Os, FwS	2
Hg methylation and reduction	Organic Hg is formed and Hg <sup>2+</sup> is converted to Hg	FwS, Os	
(per)chlorate reduction	Oxidants in rocket fuel and other sources are converted to chloride	Gw	
U reduction	U oxyanion is used as an electron acceptor; therefore immobilized	Gw	
As reduction	As oxyanion is used as an electron acceptor; therefore toxicity is diminished	FwS, Gw	
Fe oxidation, acid mine drainage	FeS ores are oxidized, strong acidity is generated	FwS, Gw	

As, arsenic; C, carbon;  $CH_4$ , methane;  $CO_2$ , carbon dioxide; Fe, iron; FeS, iron sulphide; Fw, freshwater; FwS, freshwater sediment; Gw, groundwater;  $H_2$ , hydrogen; Hg, mercury;  $Hg^{2+}$ , mercuric ion; MTBE, methyl tertiary butyl ether;  $N_2$ , nitrogen;  $NH_3$ , ammonia;  $NH_4^+$ , ammonium;  $NO_2^-$ , nitrite;  $NO_3^-$ , nitrate; Os, ocean sediments; Ow, ocean waters; S<sup>0</sup>, elemental sulphur; S<sup>2-</sup>, sulphide; SI, soil;  $SO_4^{-2-}$ , sulphate; Sw, sewage; U, uranium.