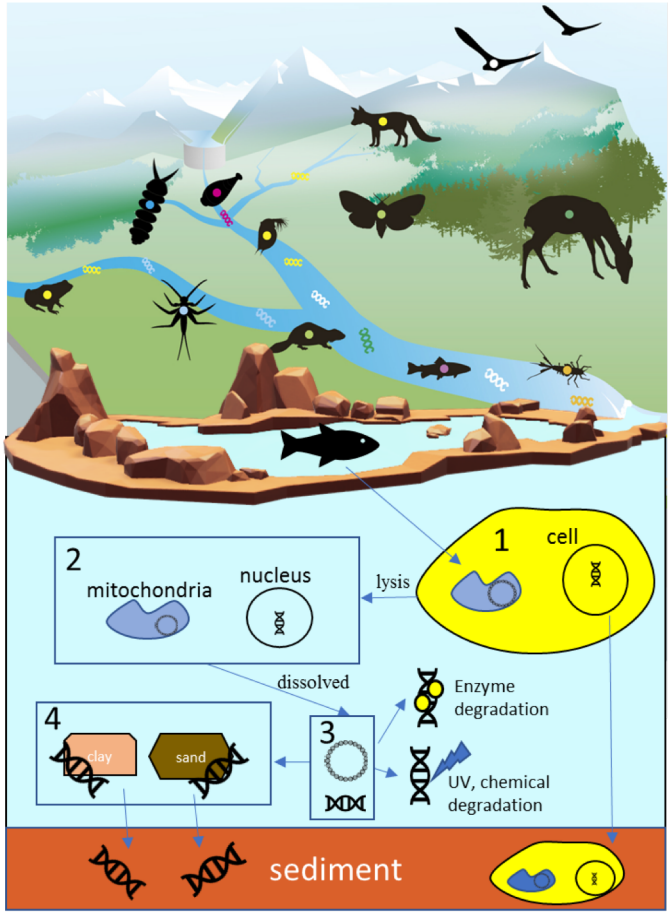
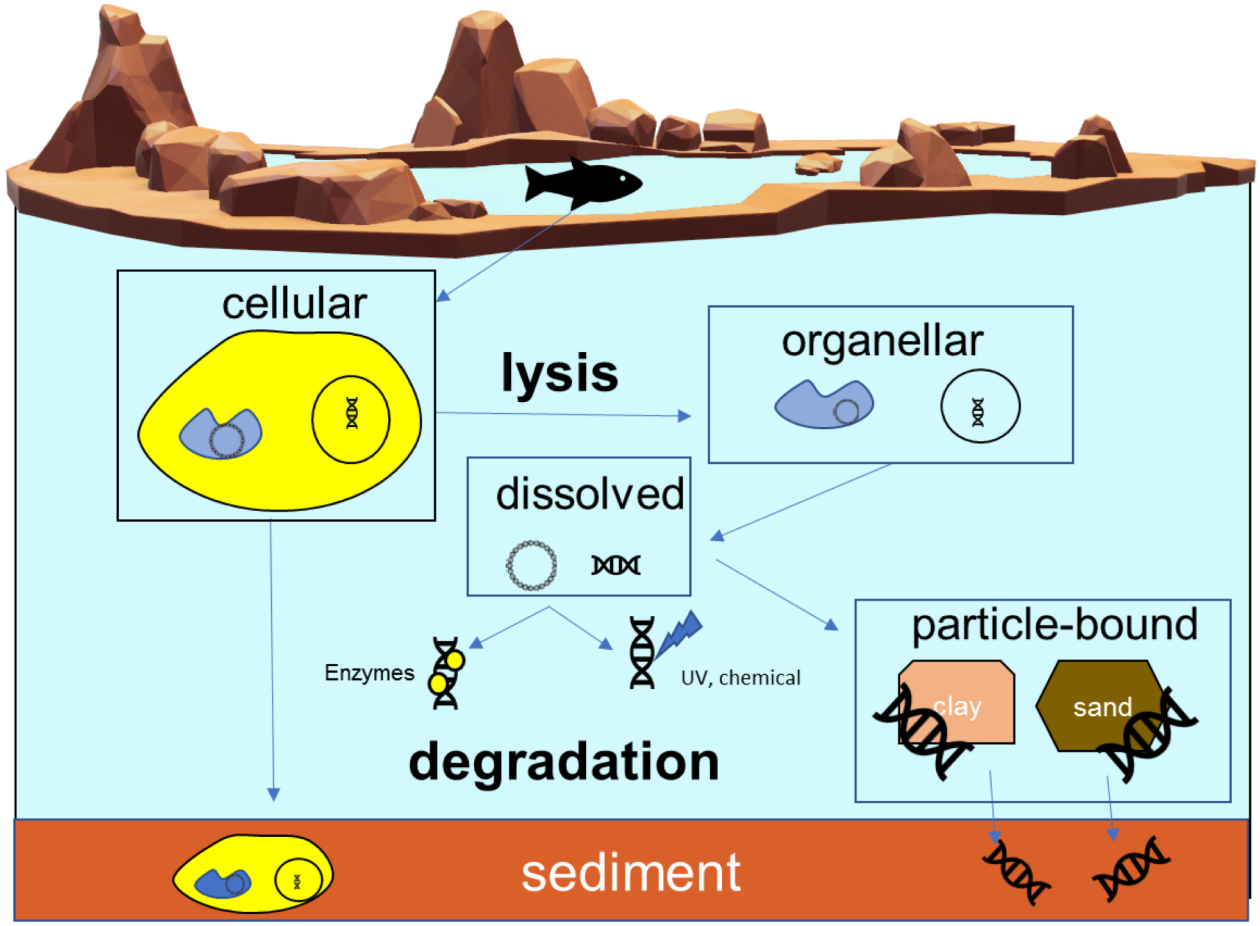


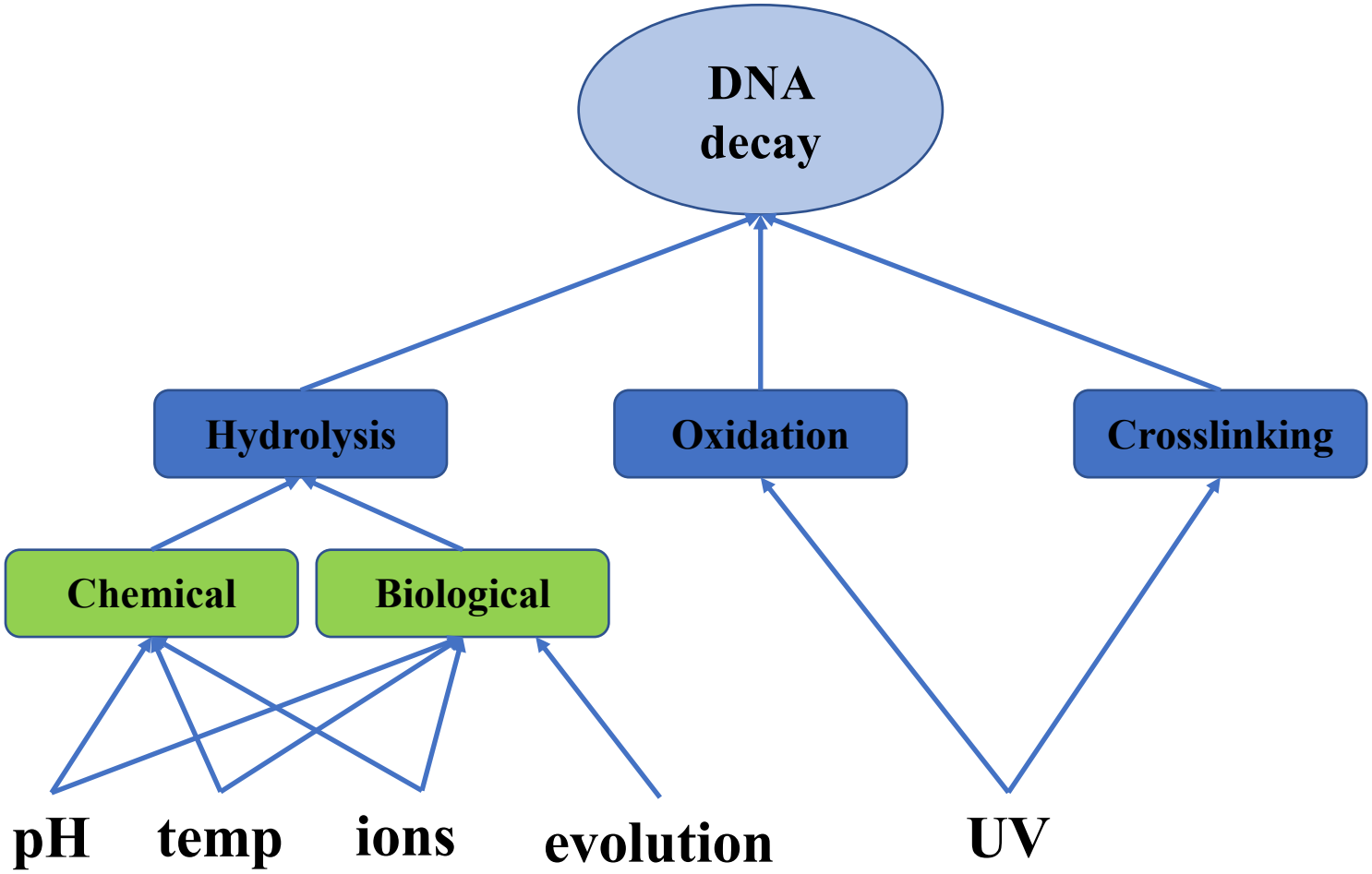
What is Environmental DNA?
How do we sample and sequence it?
What can we learn from it?

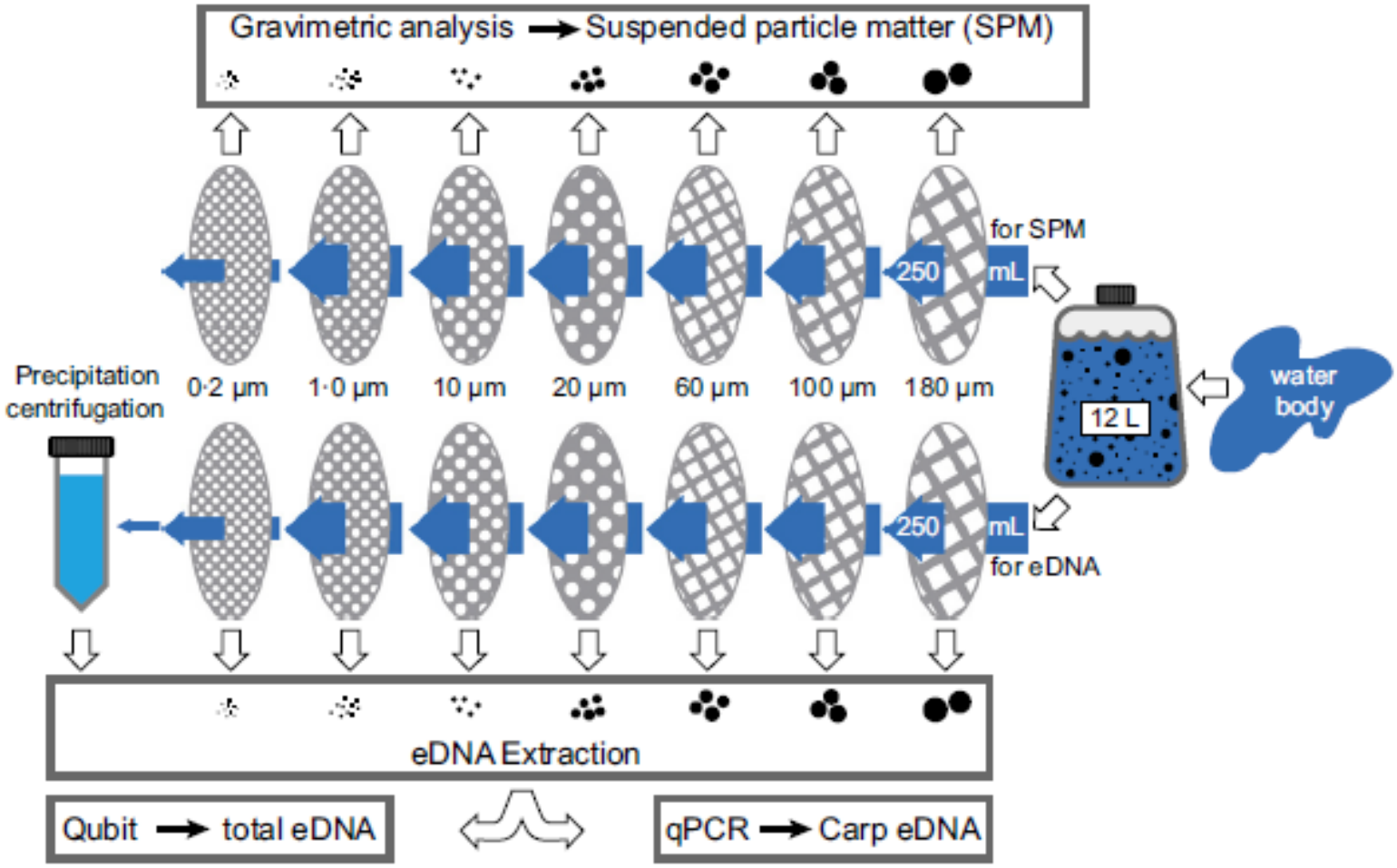
Kristy Deiner, soon (April1) to be Prof of Environmental DNA Ecology at ETH

What is Environmental DNA?

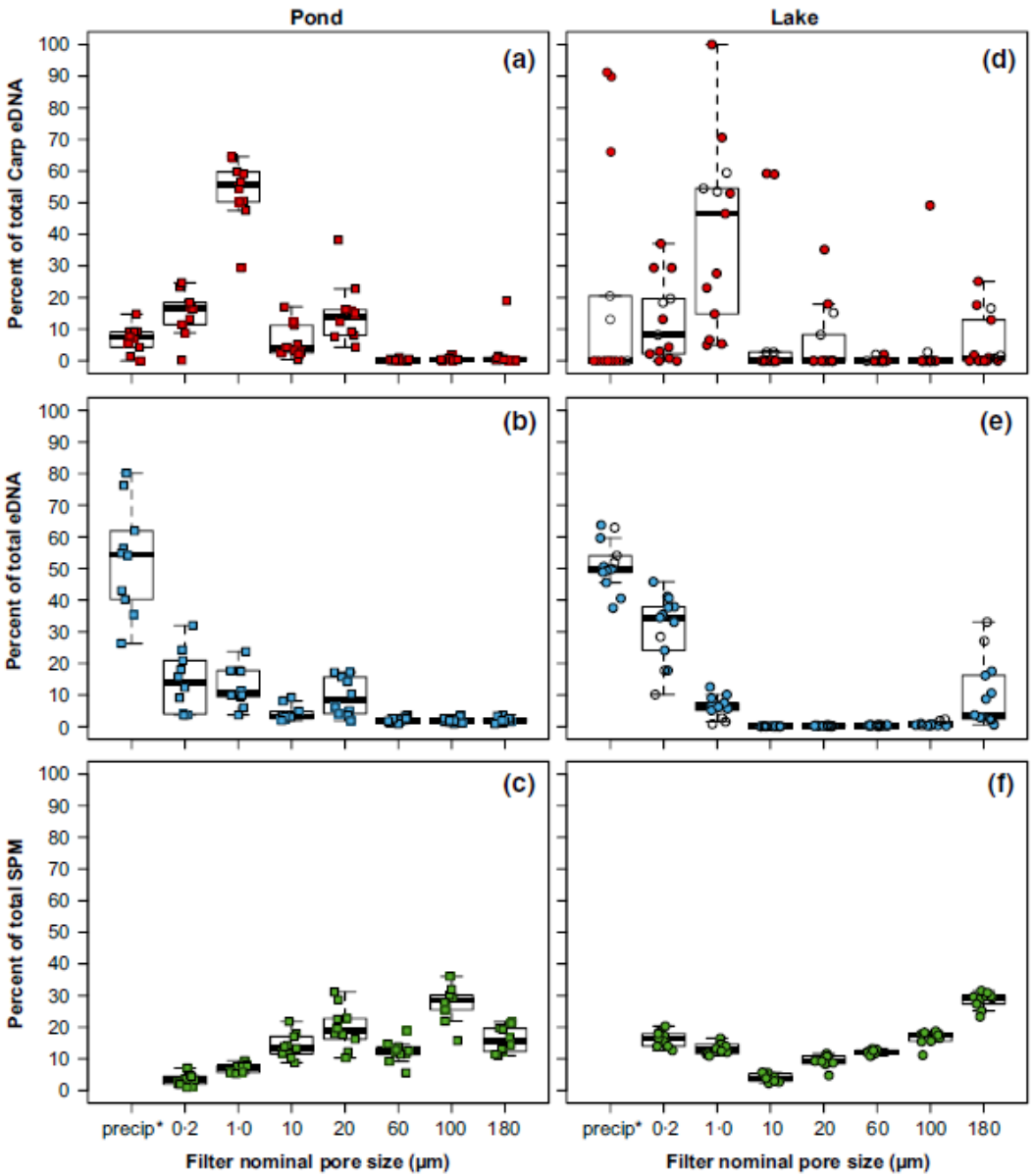
The complex state of ‘eDNA’ =Temporal inference




DNA
decay
Hydrolysis
Oxidation
Crosslinking
Chemical
Biological
pH
temp
ions
evolution
UV



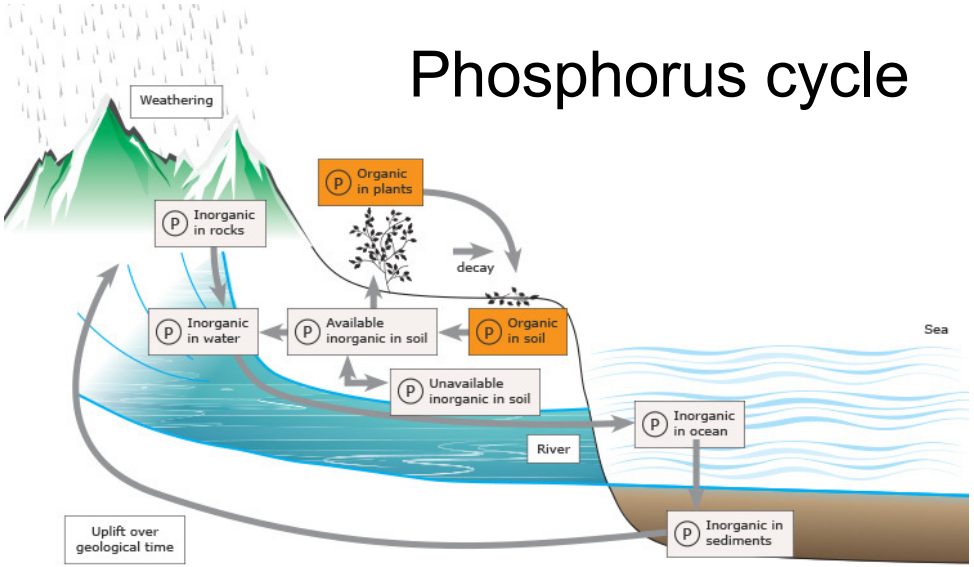
Turner *et al.* 2014 MEE



Turner et al. 2014 MEE

Degradation of eDNA as a function of…

Phosphorus cycle



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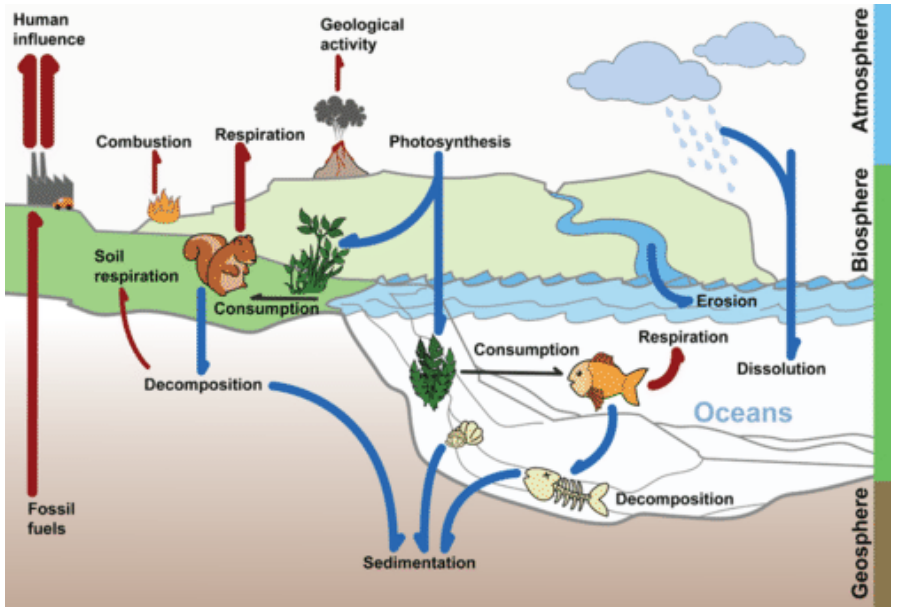
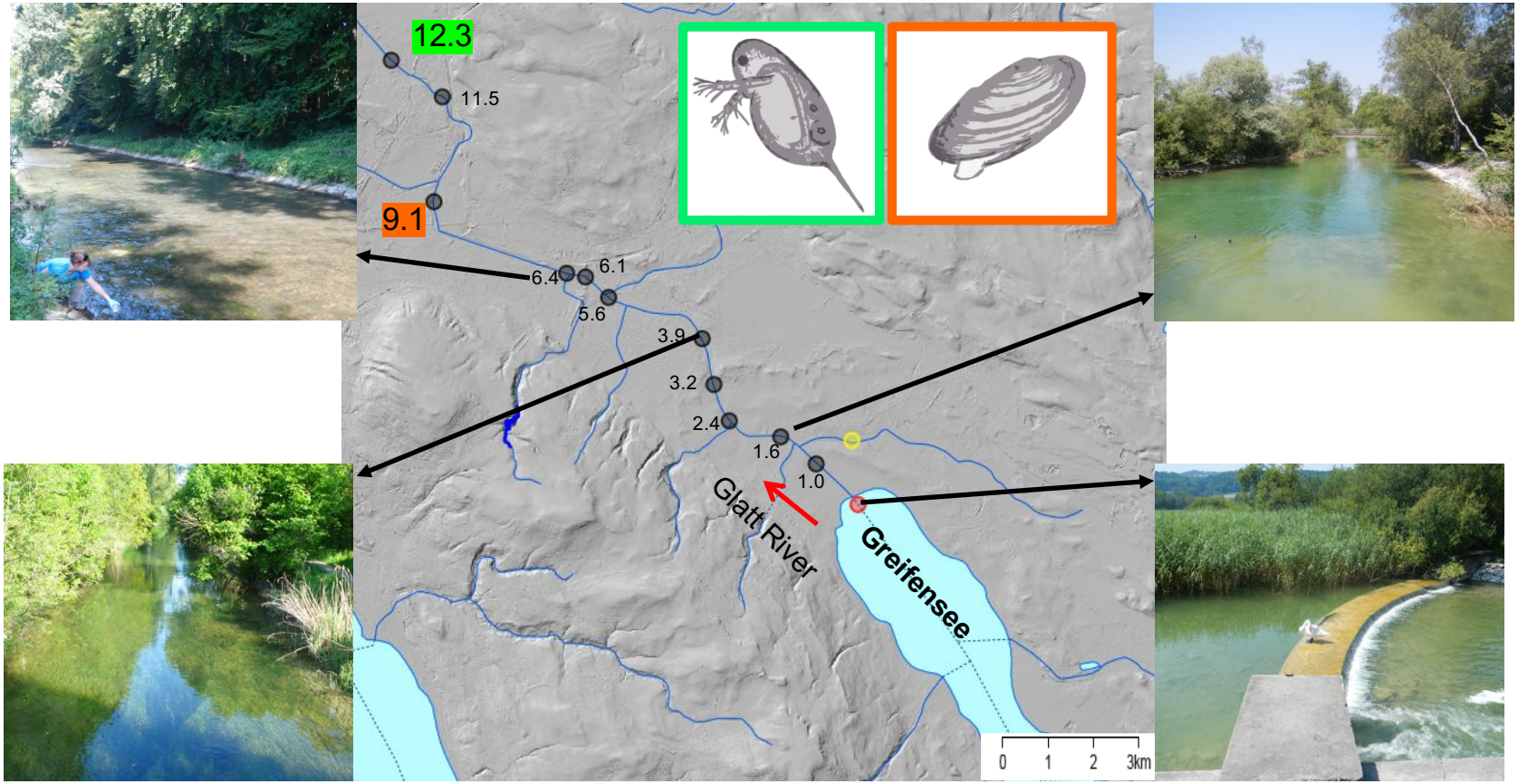
Carbon cycle



The complex transport of ‘eDNA’

= Spatial inference

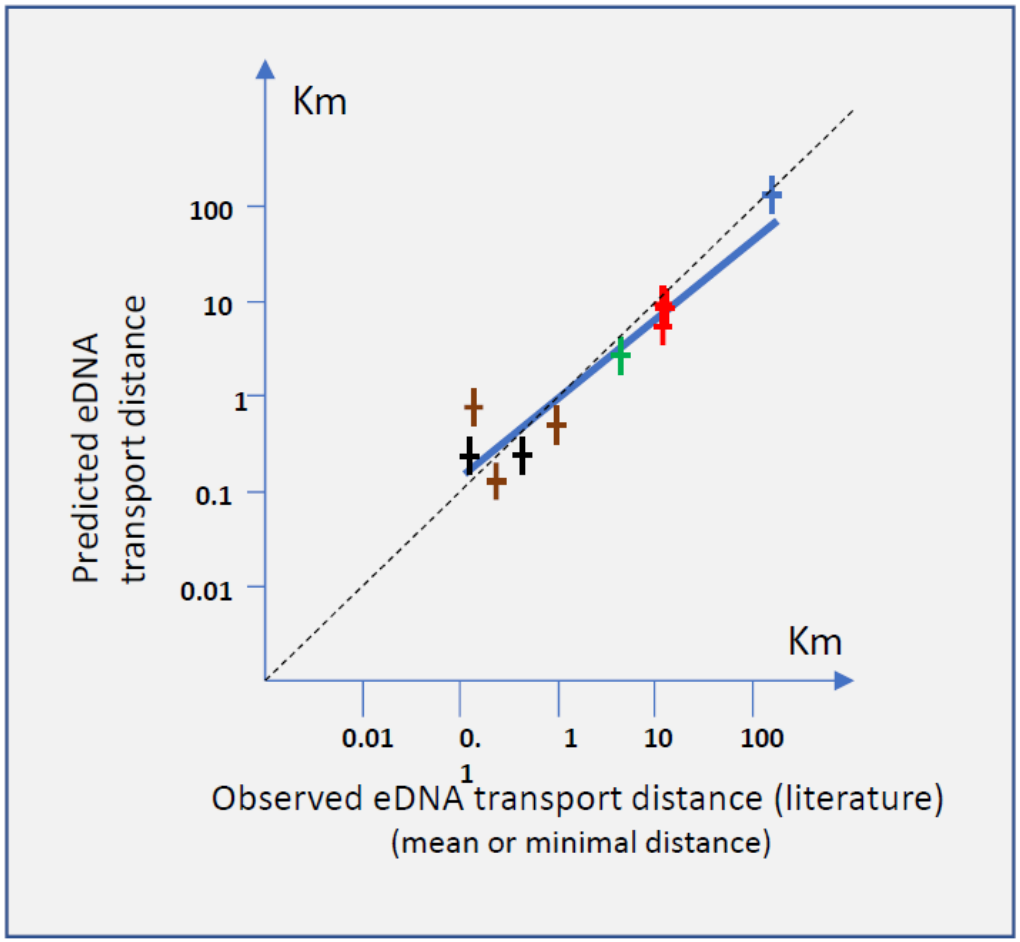
Transport of Environmental DNA



15-hour minimal transport time

Deiner & Altermatt 2014 *PLoS One*

Relating flow and treating eDNA like **FPOM** allows predicted distance downstream

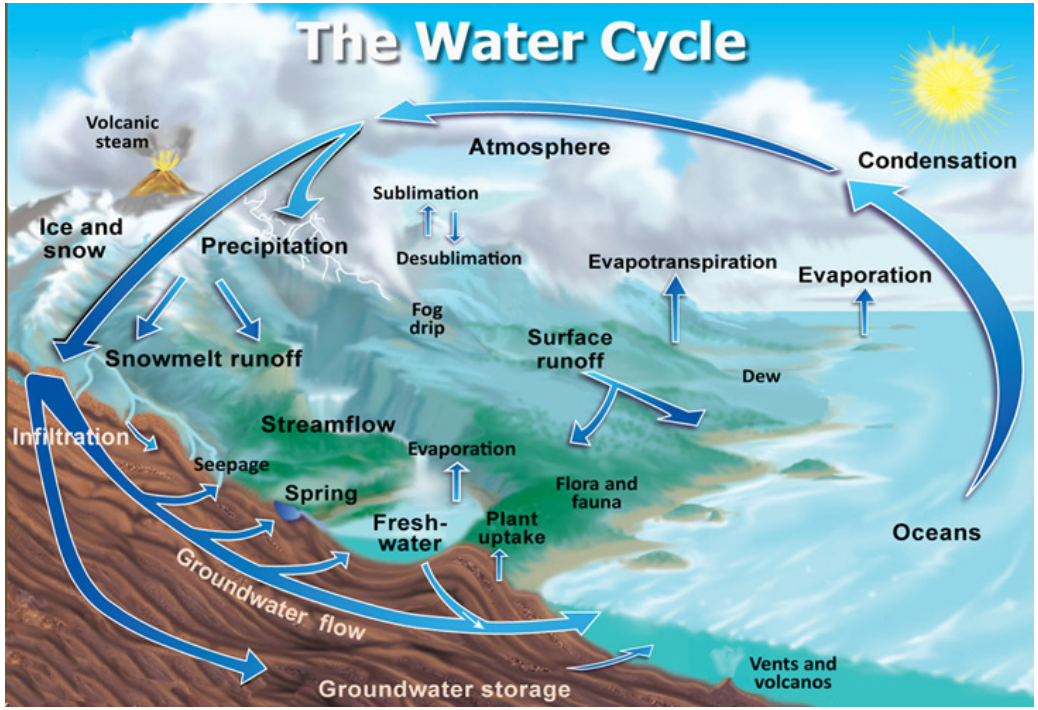


Deiner et Altermatt (2014)
Jane et al. (2015)
Wilcox et al. (2016)
Civade et al. (2016)
Pont et al. (2018)

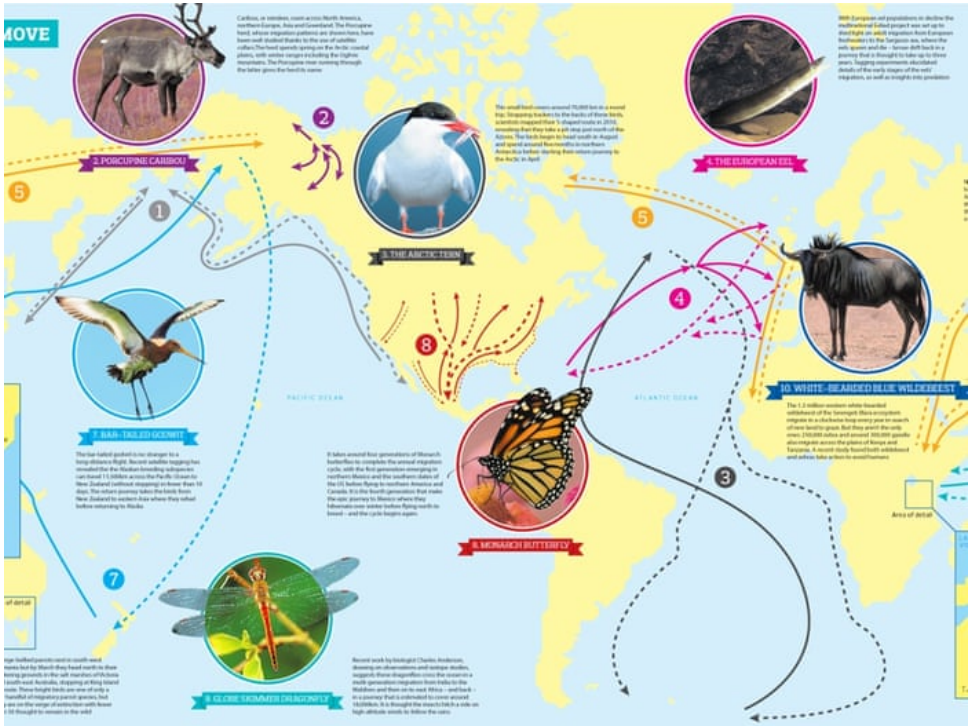
Ponte *et al.* 2018 *Scientific Reports*

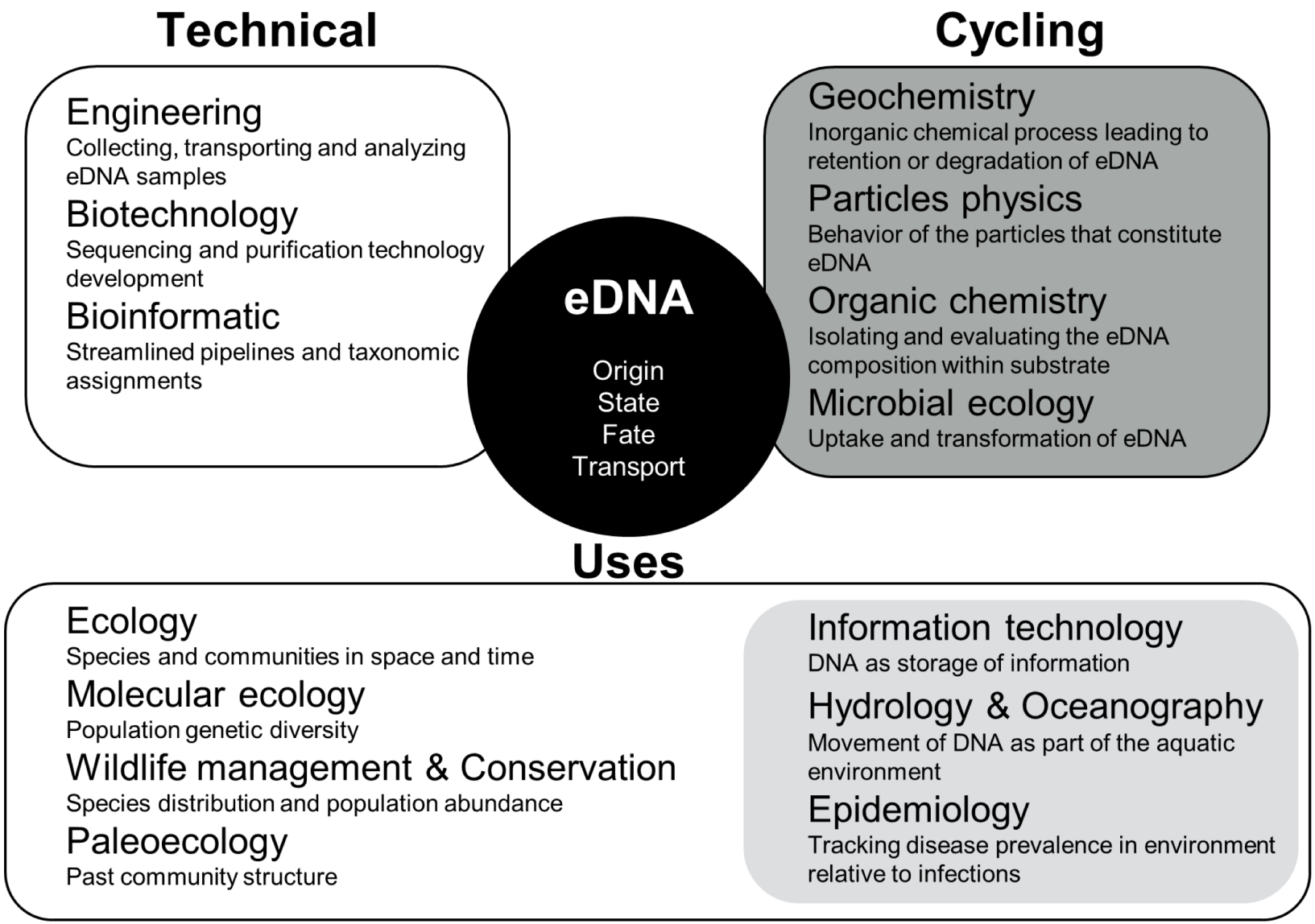
Transport of eDNA is a function of….

abiotic



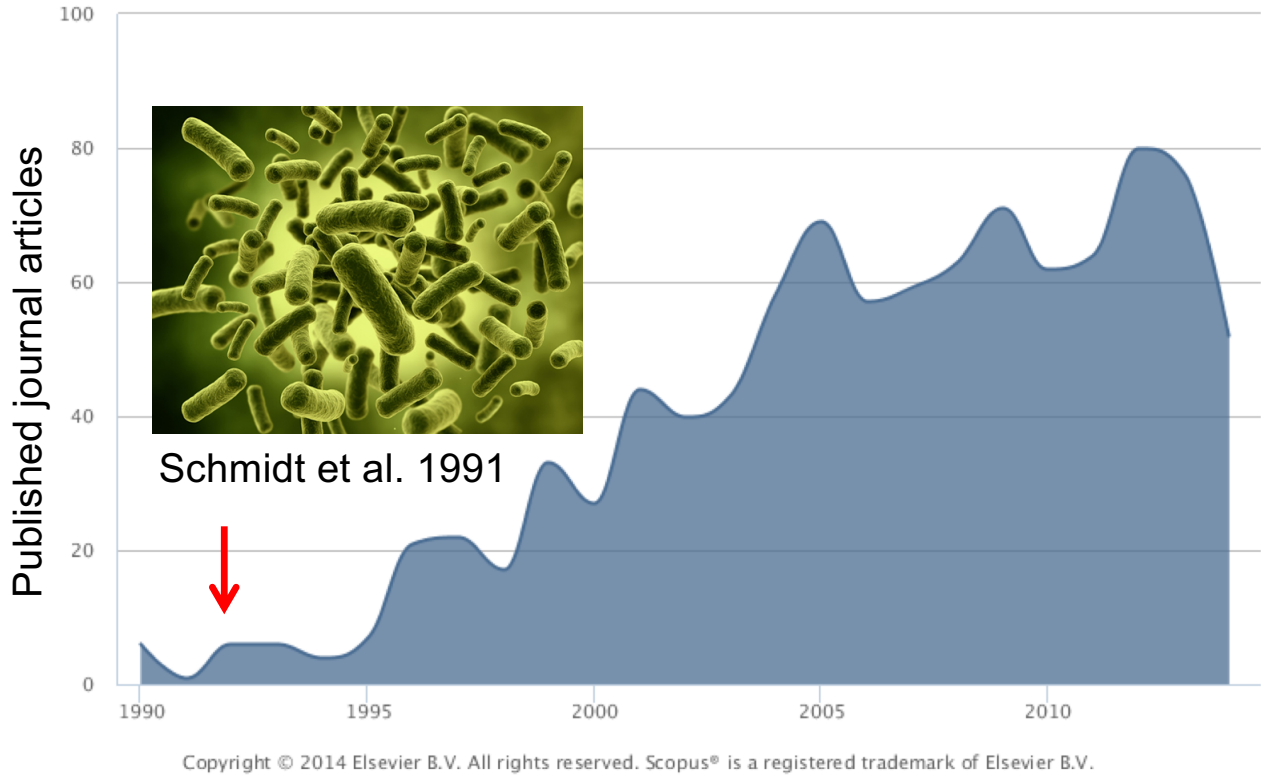
biotic





Lacoursière-Roussel & Deiner 2019 *J. Fish Biol.*

“Environmental DNA”



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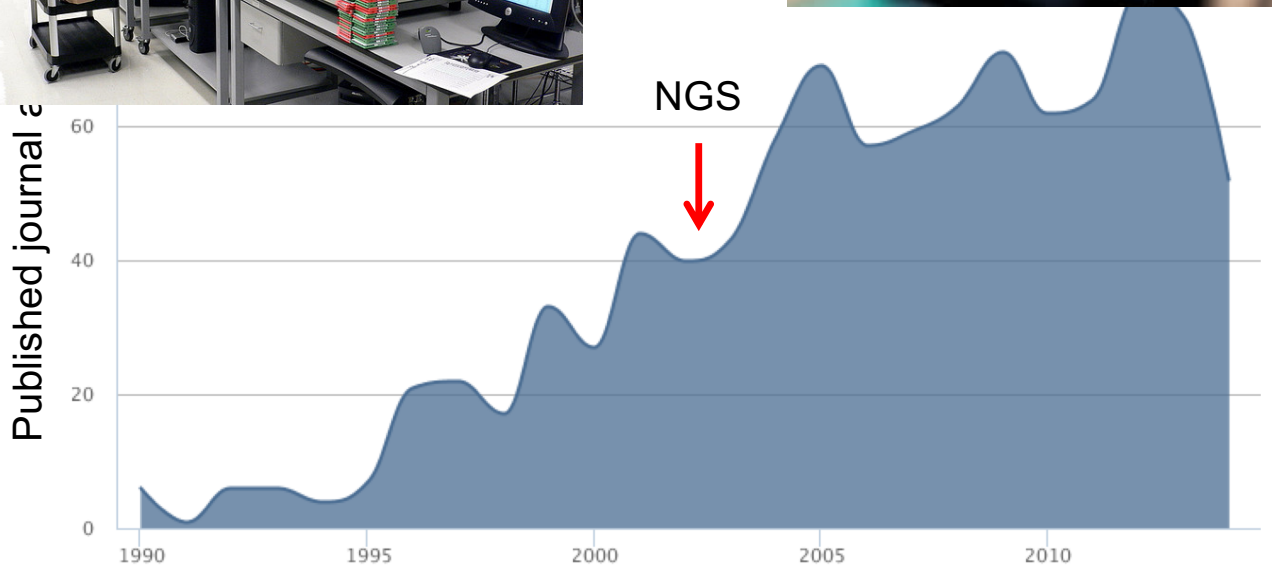
“Environmental DNA”



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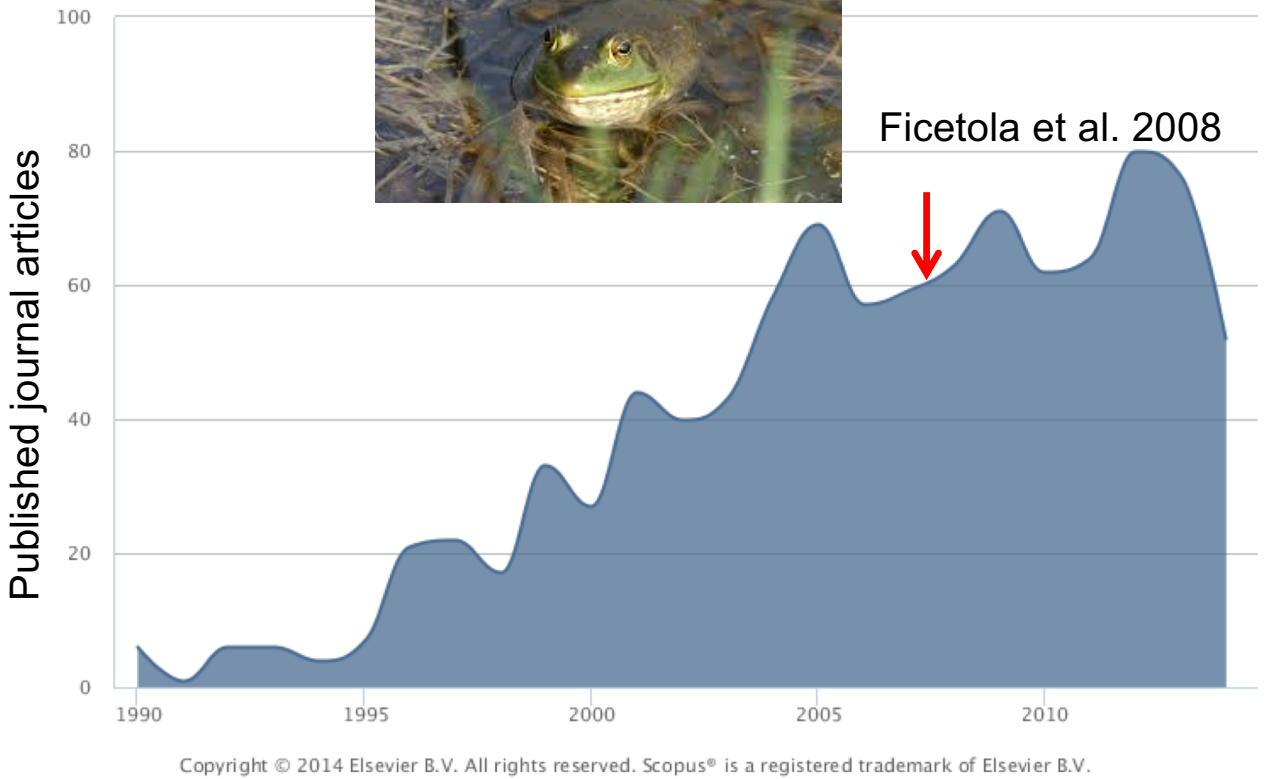
“Environmental DNA”



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How do we sample and sequence it?

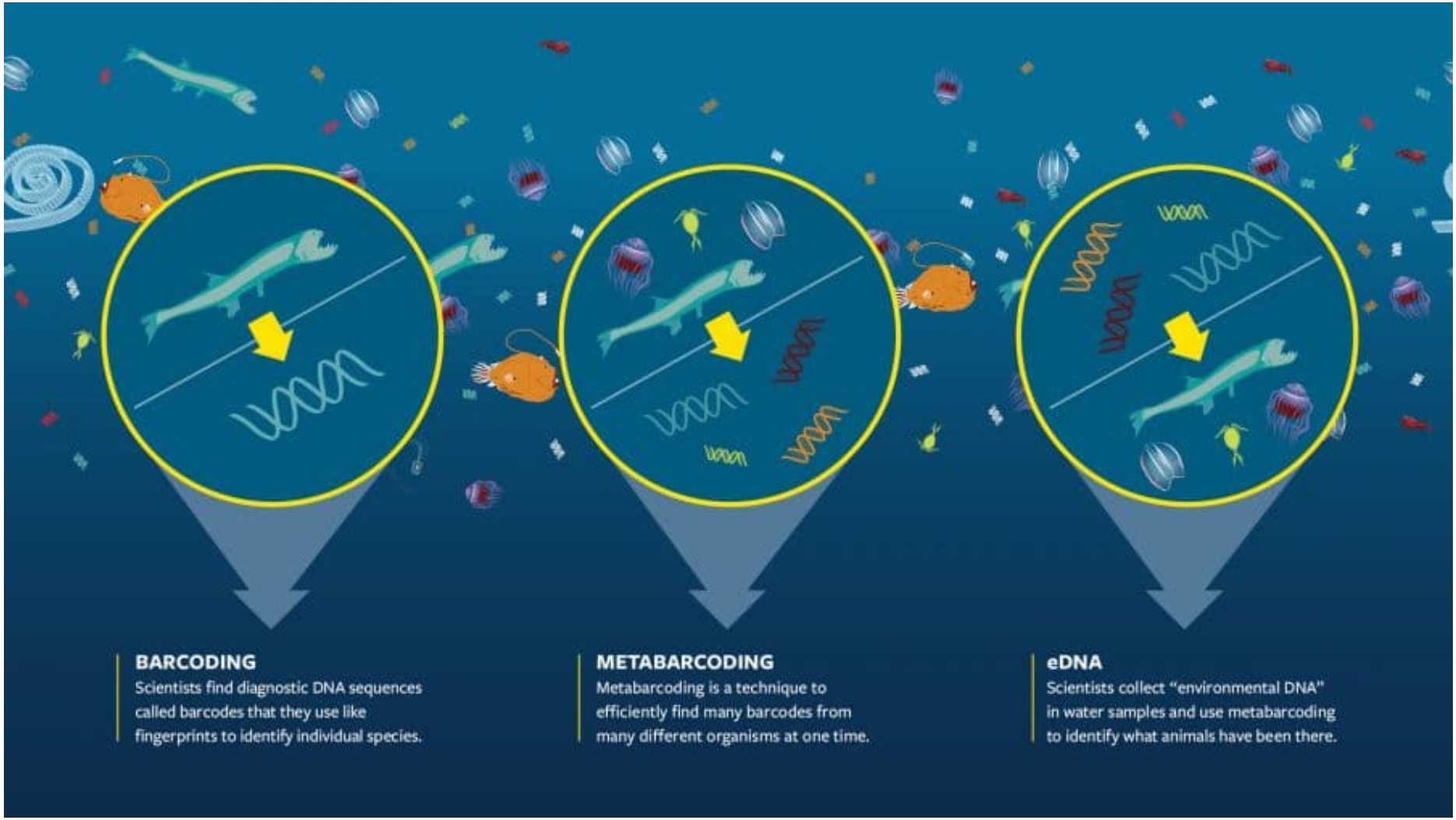
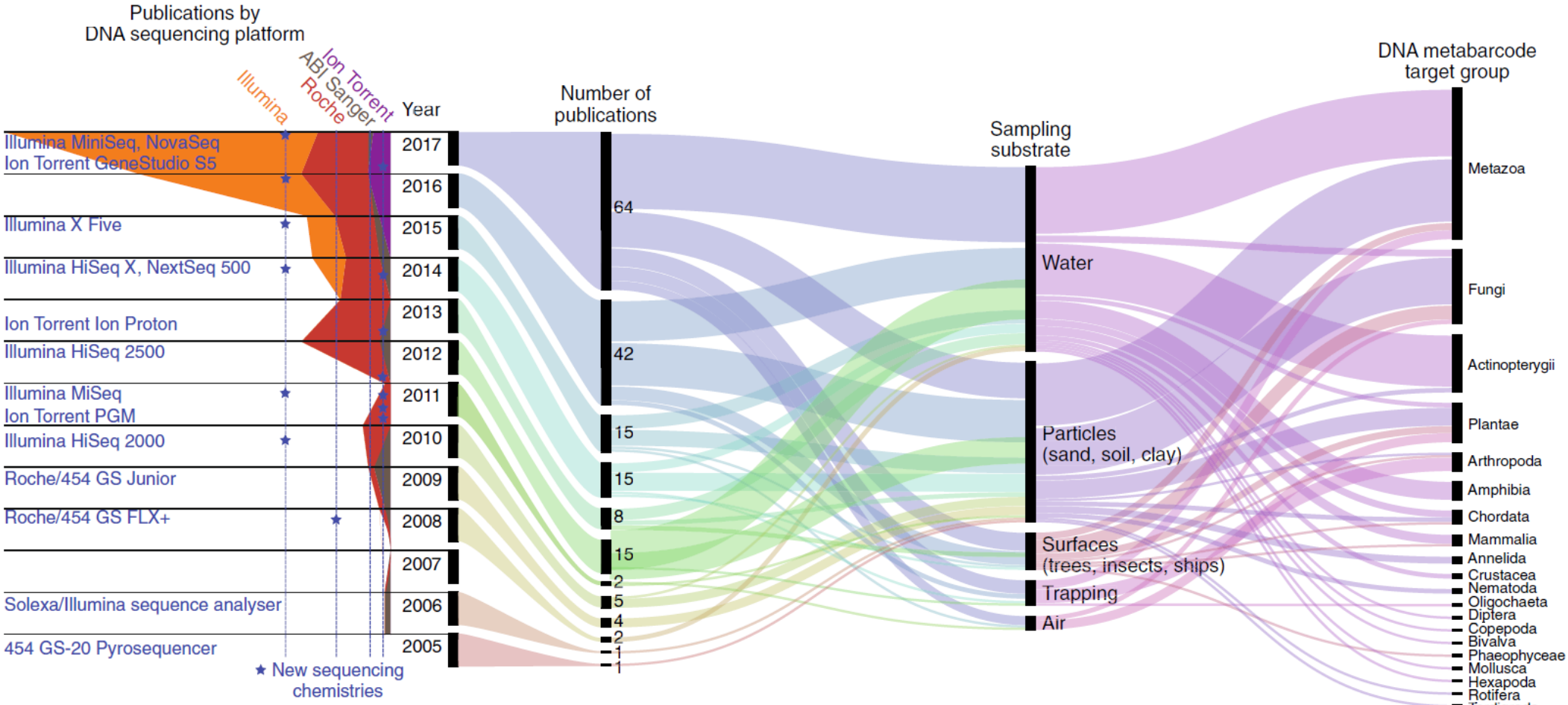


Illustration by Natalie Renier and Eric S. Taylor, WHOI Creative

a
b



Jarmine et al 2017 *Nat Ecol & Evol*

WORKFLOW

Study design

Basic science or applied? (e.g., environmental biomonitoring)

What is your study goal?
- presence/absence
- diversity assessment
- absolute quantification

What taxa will you target?

Is the scale of inference for your sample type appropriate to your question?

Can you compare complementary data types? (e.g. traditional vs. eDNA)

Does your sampling/ replication scheme provide good statistical power?

In the field

What type of sample is needed? (water, soil, air)

What metadata should you collect?

How many replicates will you collect?

Does your sampling protocol minimize/ control for :
- contamination (e.g., positive and negative controls)
- any known biases (e.g., inhibitors, sample volume)

In the laboratory

Sample Handling Phase

What extraction method? (physical vs. chemical)

How much sample?

What locus and primers?

Do you need to generate reference sequence data?

Are technical replicates needed?

What library preparation method will you use?

How many samples will you index and pool?

What sequence depth is needed per sample ?

What read length will you use?

DNA Processing Phase

What sequencing platform will you use?

Do you need paired end sequencing?

Have you included appropriate quality assurances? (e.g., mock community, qPCR, bioanalyzer traces)

Does your laboratory protocol minimize/ control for:
- contamination (e.g., positive and negative controls)
- any known biases (e.g., primer bias, coverage, taxonomic resolution)

At the keyboard

How complete is the reference database?

Do you have adequate sequencing coverage across samples?

Are you using appropriate choices for software tools, parameters?

Are your biological conclusions upheld using alternative parameters and workflows?

Are you including appropriate quality filtering of your data? (see Box 2)

Deiner et al.
2017 *Mol. Ecol.*

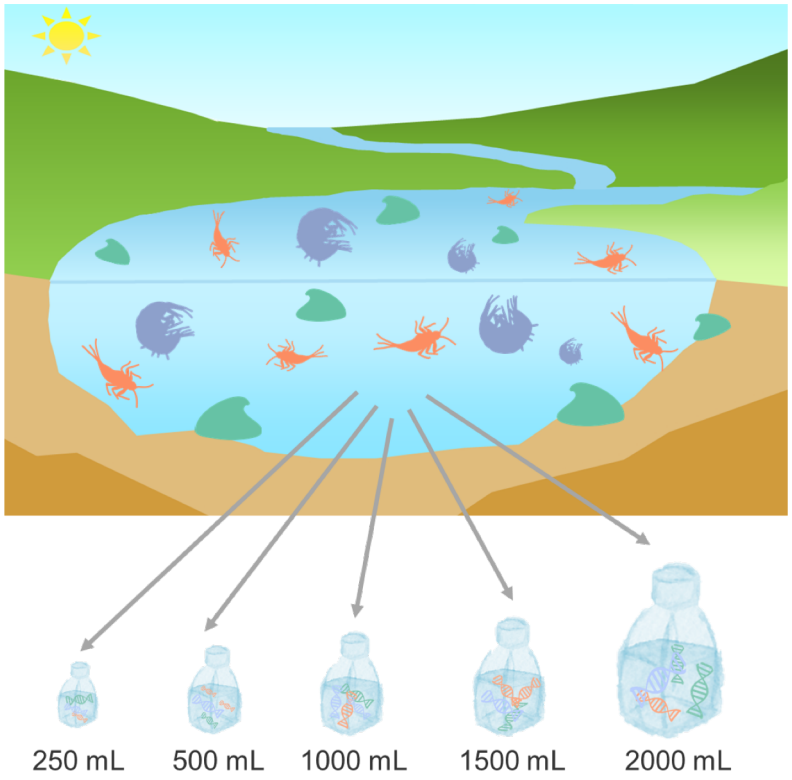
Environmental Science & Technology

Article

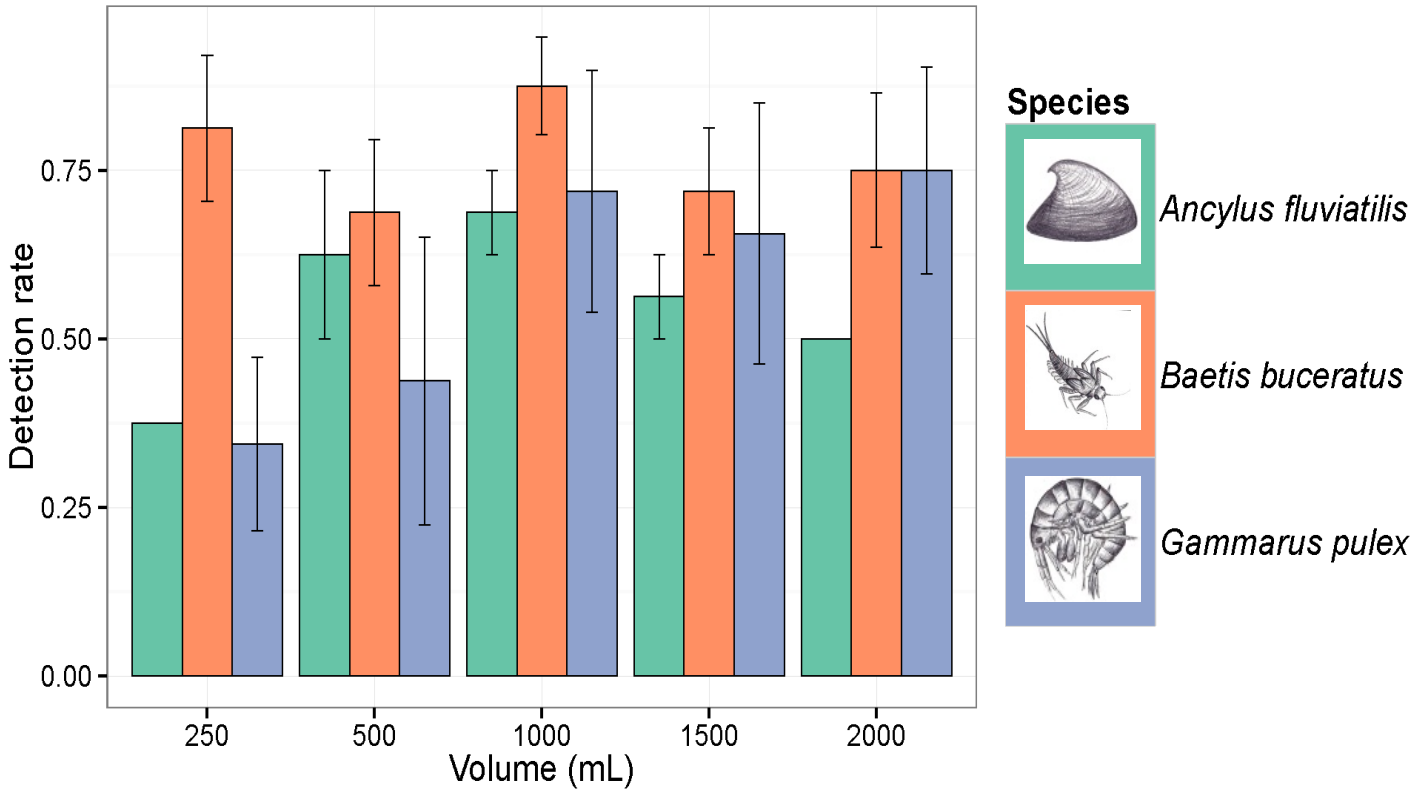
pubs.acs.org/est

Fishing in the Water: Effect of Sampled Water Volume on Environmental DNA-Based Detection of Macroinvertebrates

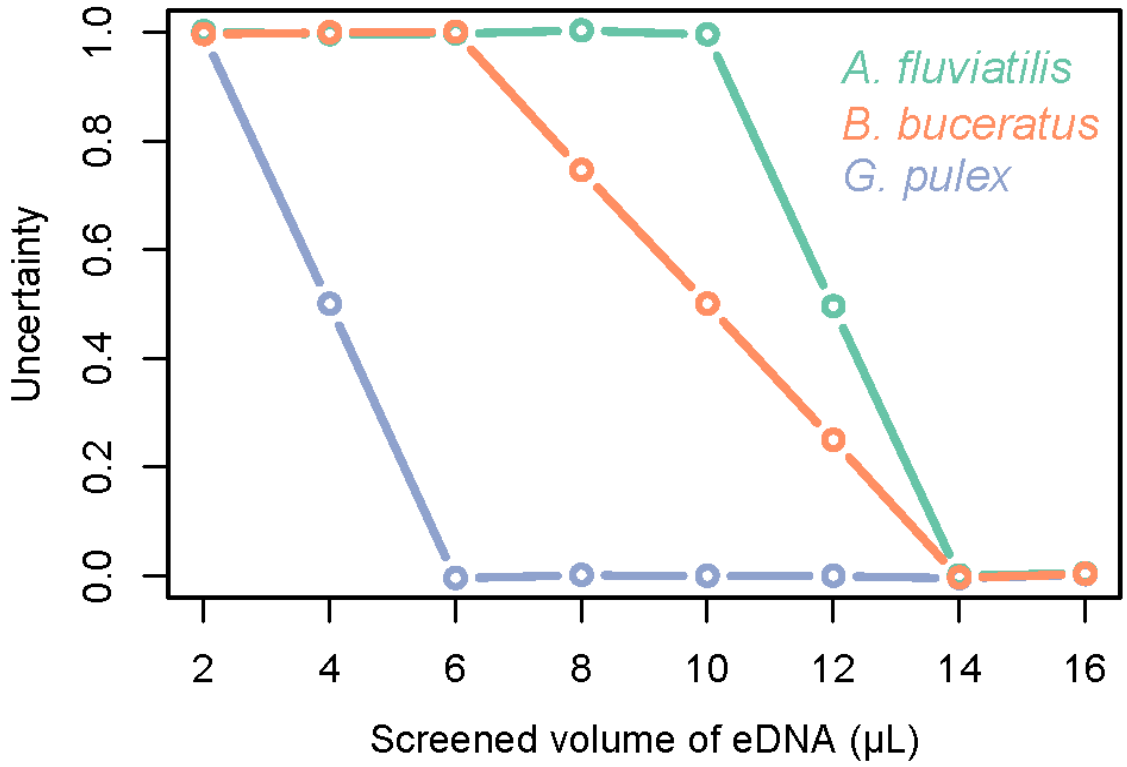
Elvira Mächler,[*,†,‡] Kristy Deiner,[†,§] Fabienne Spahn,[∥] and Florian Altermatt[†,‡]

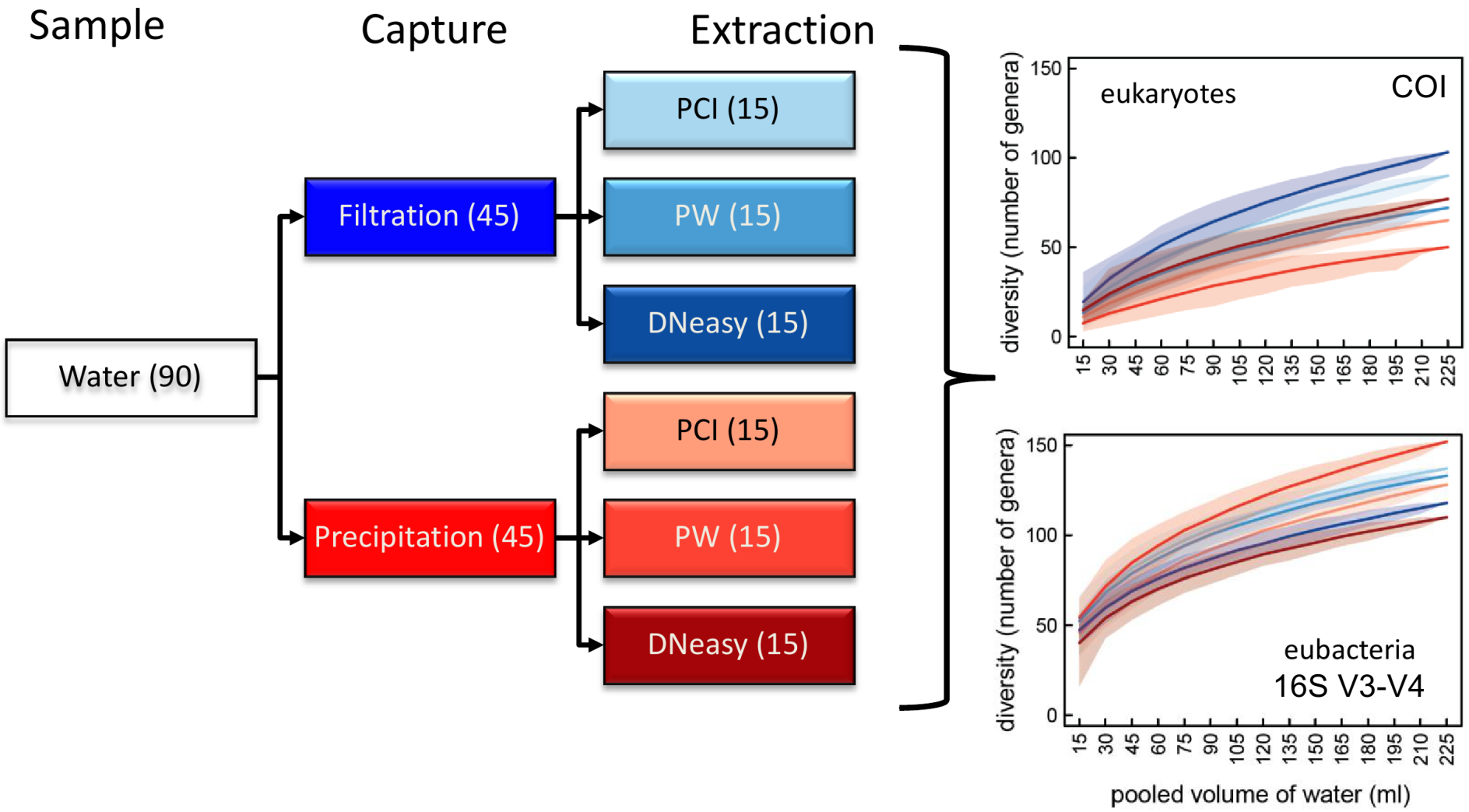


Detection rate of different species in different volumes of water

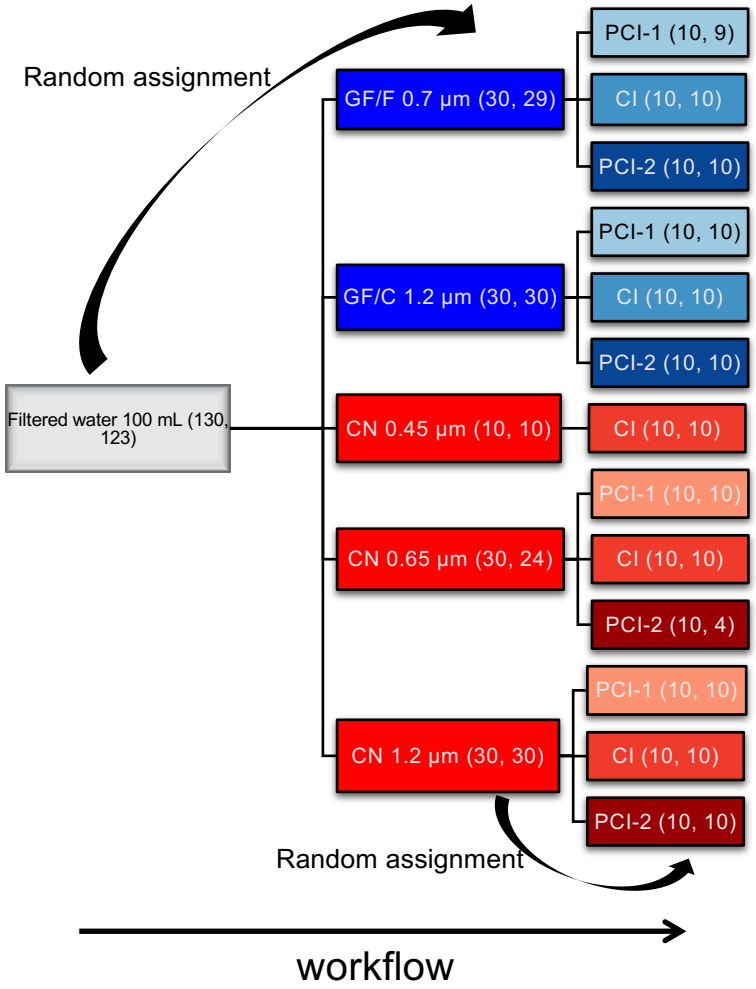


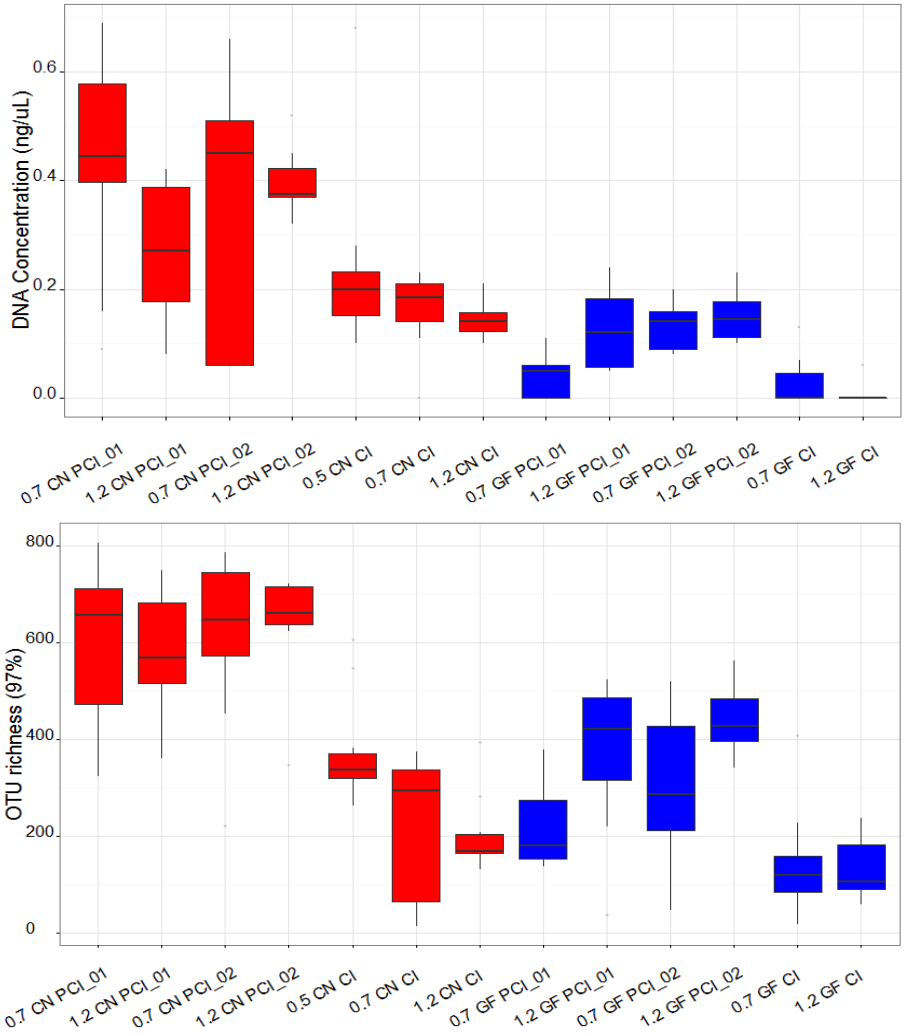
Uncertainty in detection rate of with different PCR replicates



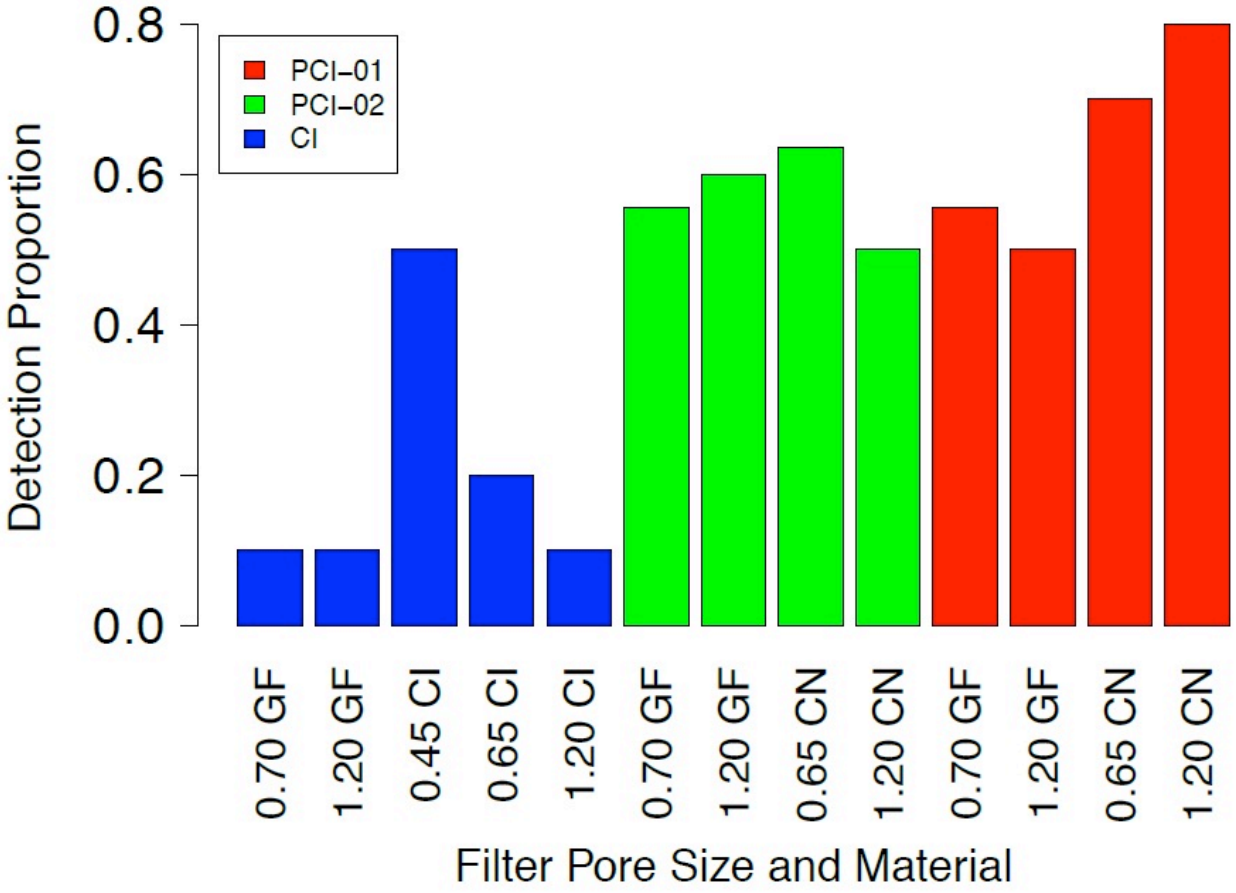


Deiner et al. 2015 *Biol Cons*

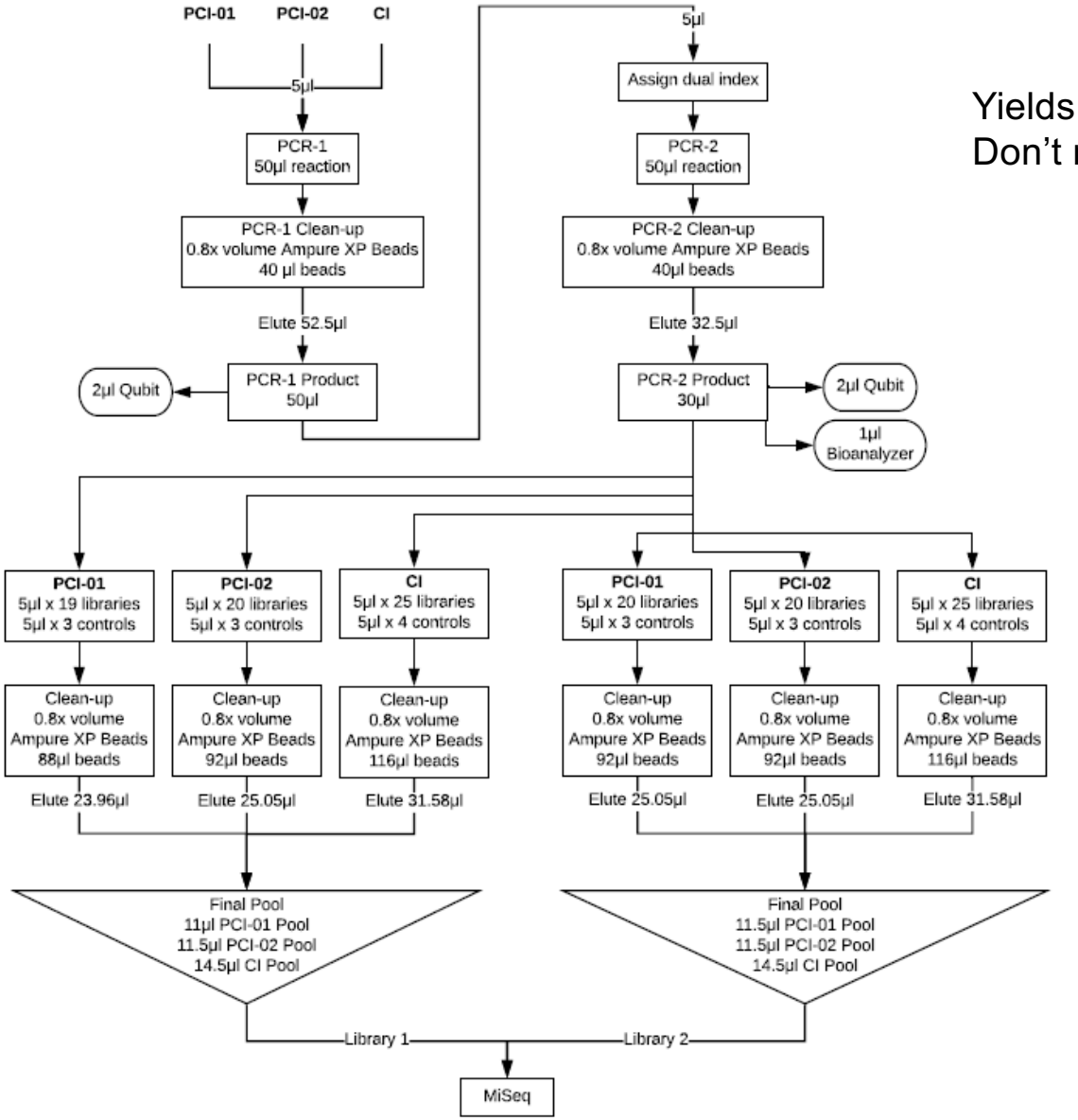




Deiner et al. 2018 *Metabar & Metagen*



Deiner et al. 2018 *Metabar & Metagen*



Yields might matter!
Don’t remove effect of experiment by normalizing…

Deiner et al. 2018 *Metabar & Metagen*

What can we learn from it?

Science of the Total Environment 709 (2020) 135727

Contents lists available at ScienceDirect

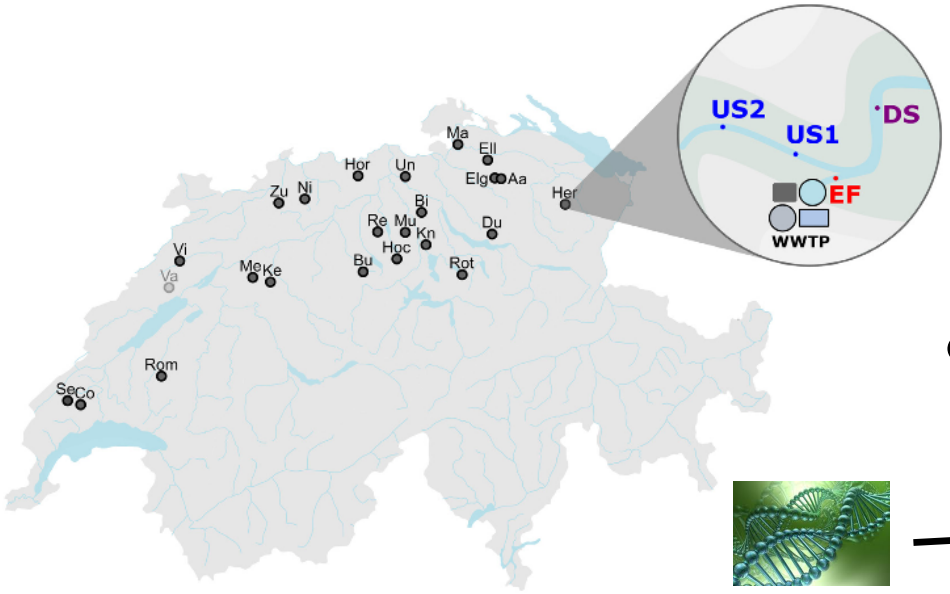
Science of the Total Environment

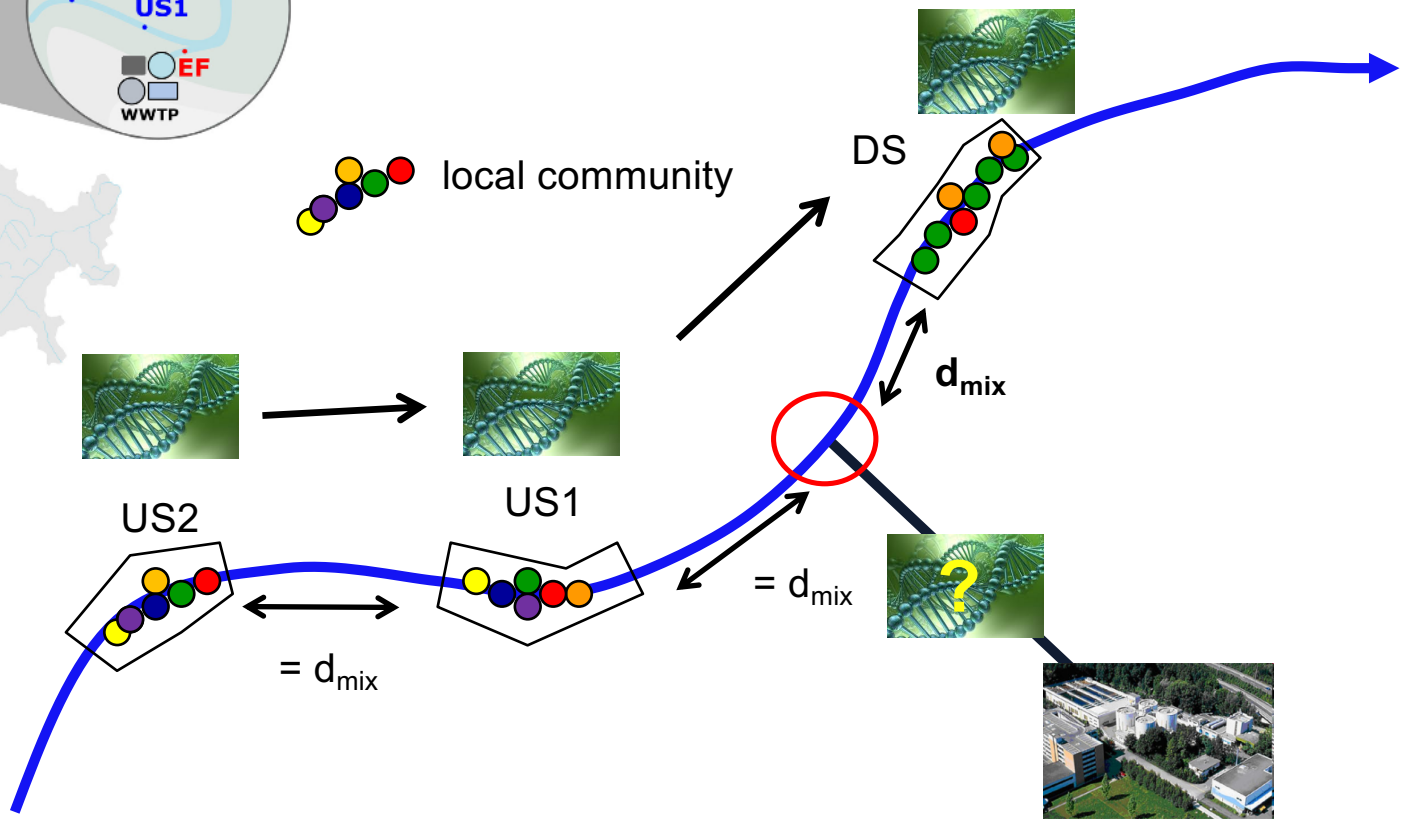
journal homepage: www.elsevier.com/locate/scitotenv

Microbial community shifts in streams receiving treated wastewater effluent

Cresten Mansfeldt [a,1], Kristy Deiner [a,b,*,1], Elvira Mächler [a,c], Kathrin Fenner [a,d,e], Rik I.L. Eggen [a,d], Christian Stamm [a], Urs Schönenberger [a], Jean-Claude Walser [d], Florian Altermatt [a,c]

[a] Eawag: Swiss Federal Institute of Aquatic Science and Technology, Dübendorf, Switzerland
[b] Natural History Museum London, London, UK
[c] Department of Evolutionary Biology and Environmental Studies, University of Zürich, Zürich, Switzerland
[d] Department of Environmental Systems Science, ETH, Zürich, Switzerland
[e] Chemistry Department, University of Zürich, Zürich, Switzerland



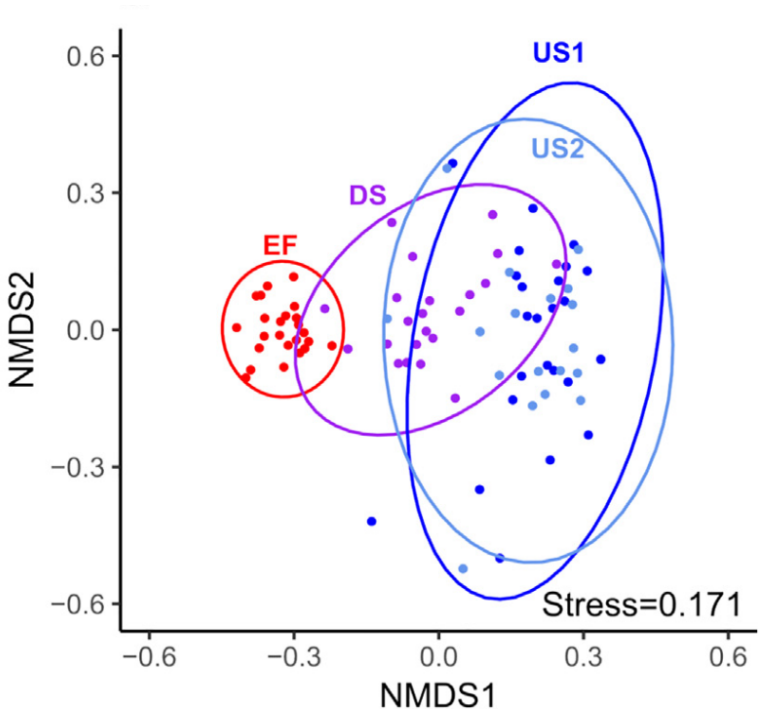


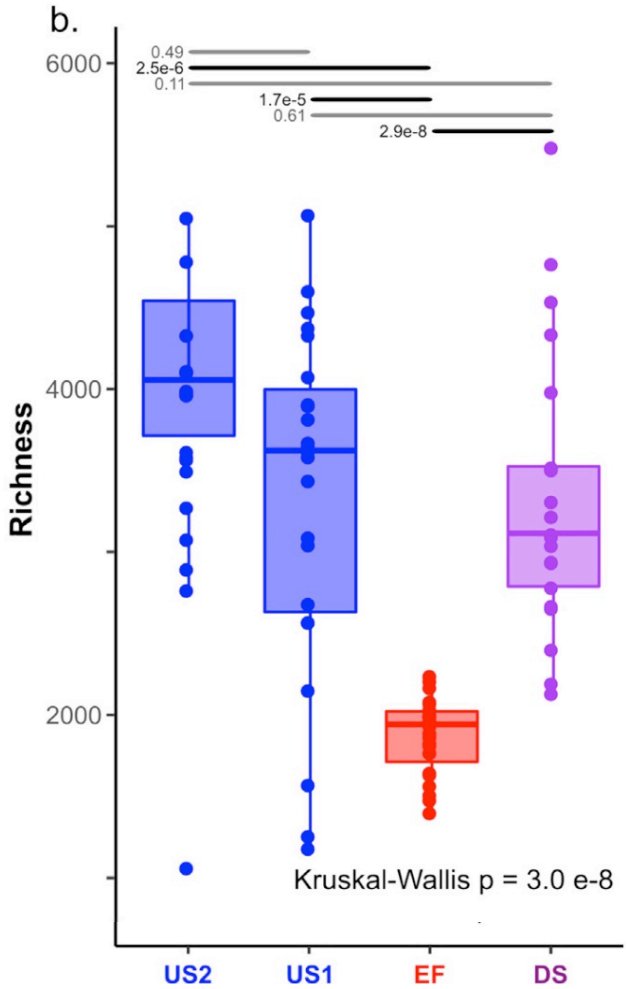
Hypothesis

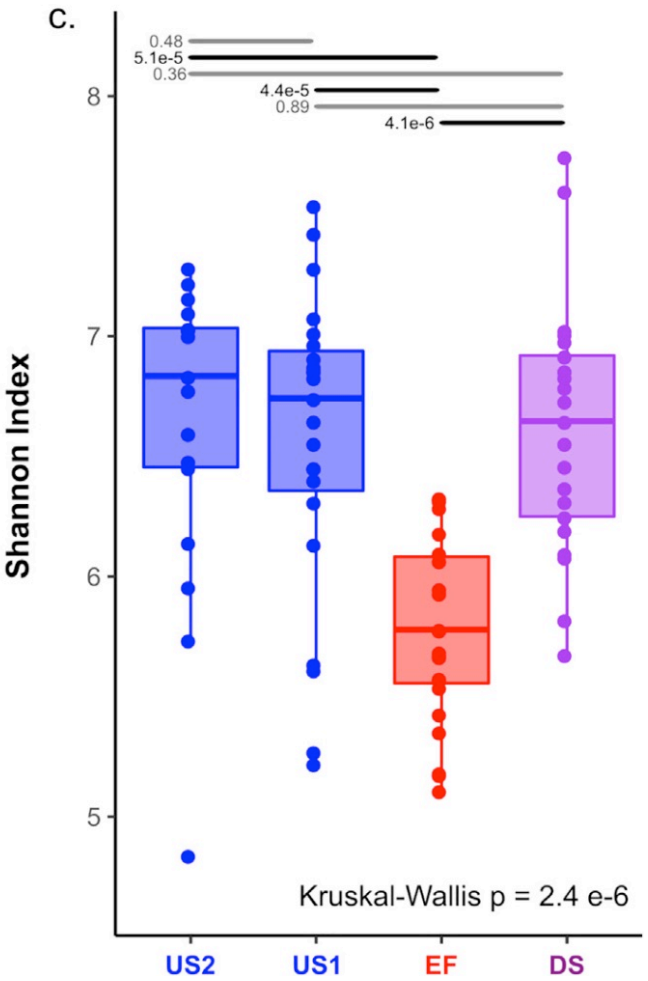
- Bacterial community @ DS is hydraulic mixture of US and EF

d_{mix}: Mixing distance required to achieve full mixing of waste water into the stream water (site-specific; ranging between approx. 50 to 500 m).

Using phyloseq:

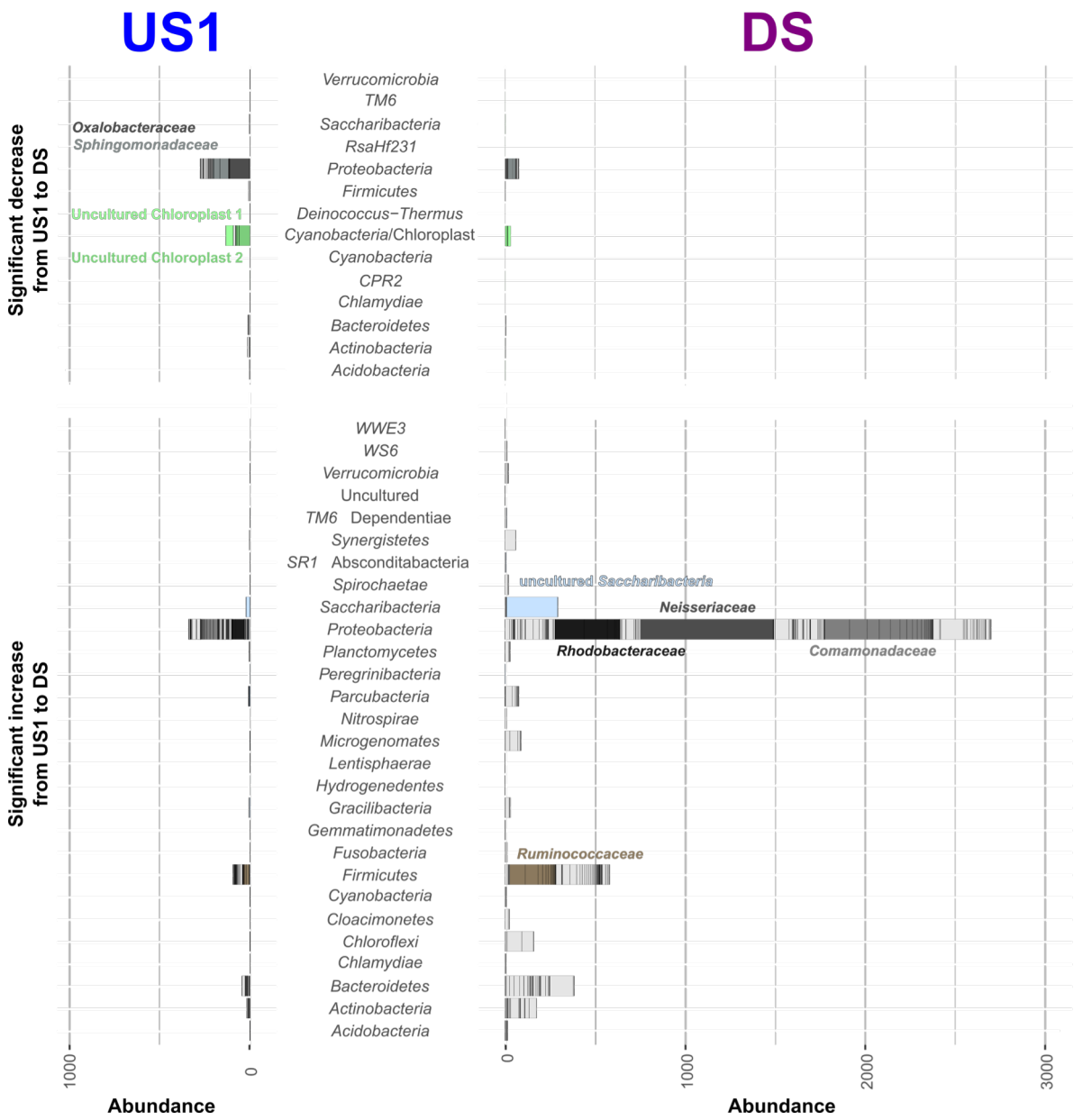




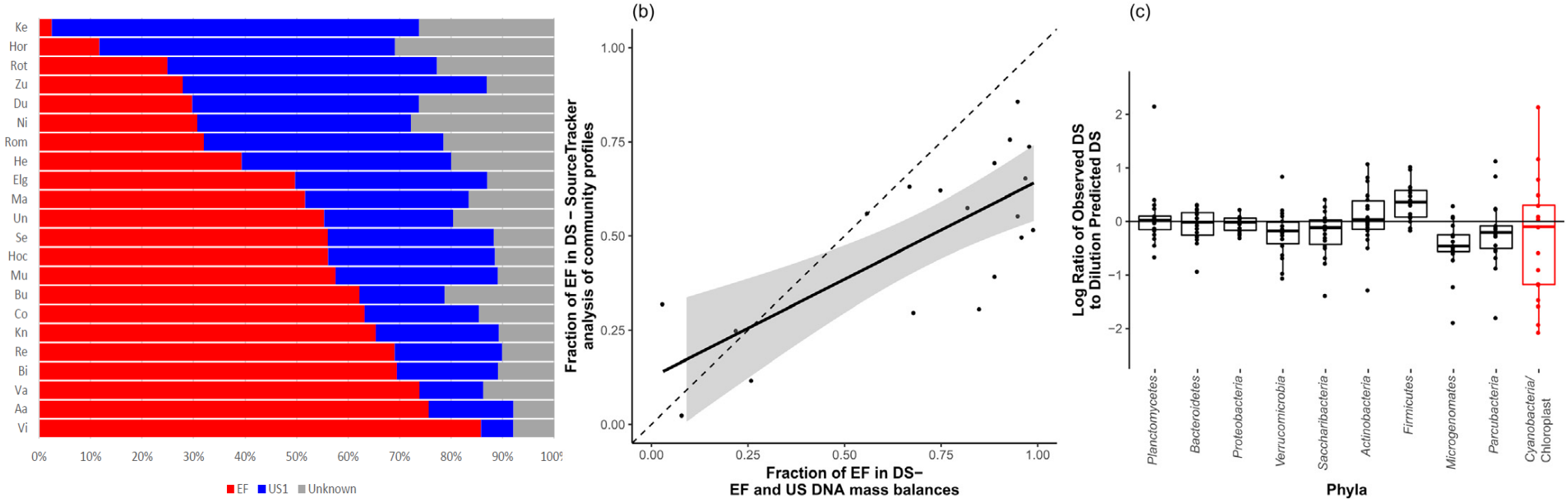


Unweighted UniFrac dissimilarity (considering identity and phylogentic relatedness)

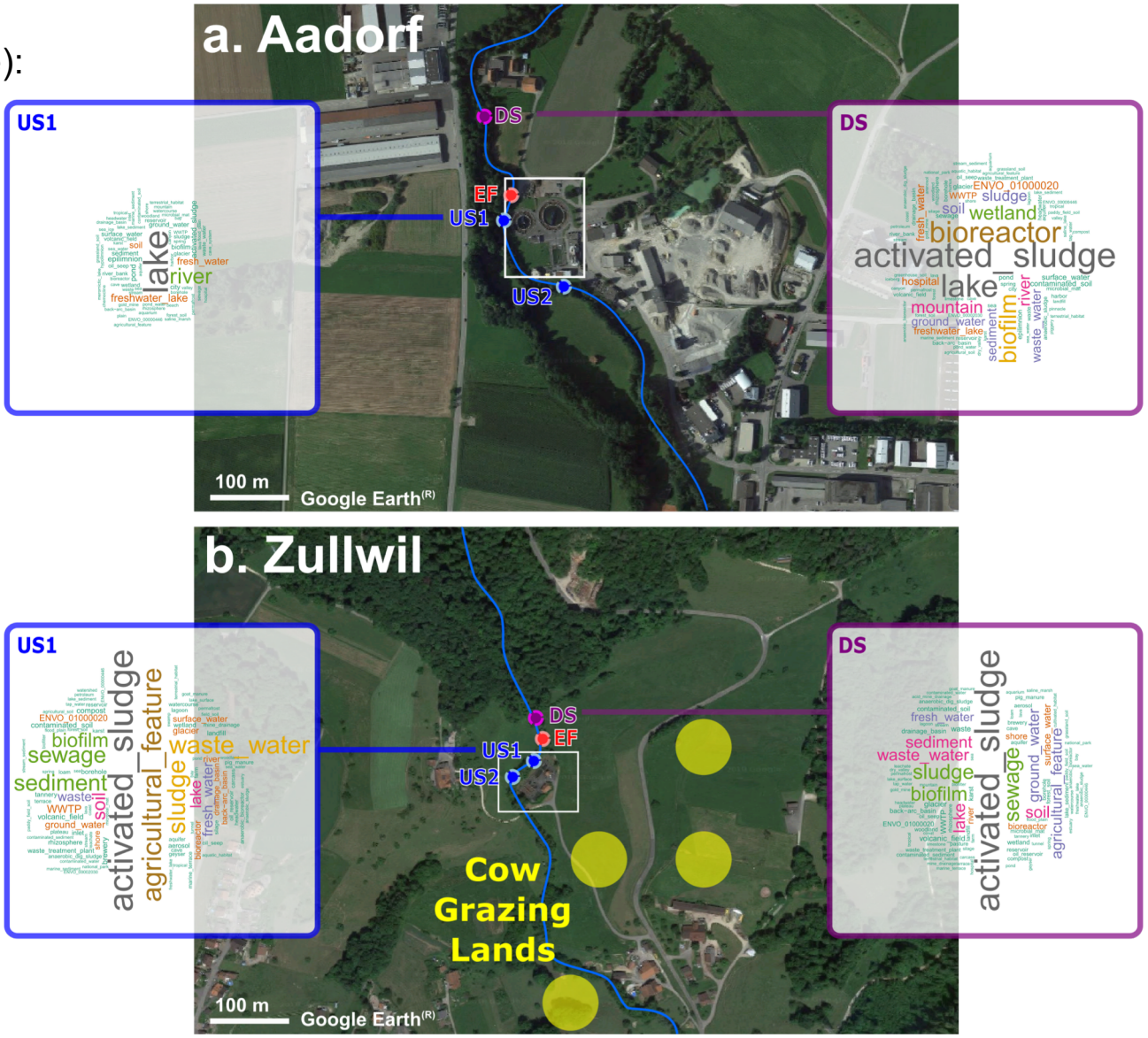
Using DESeq2: tested for differential ESV abundance



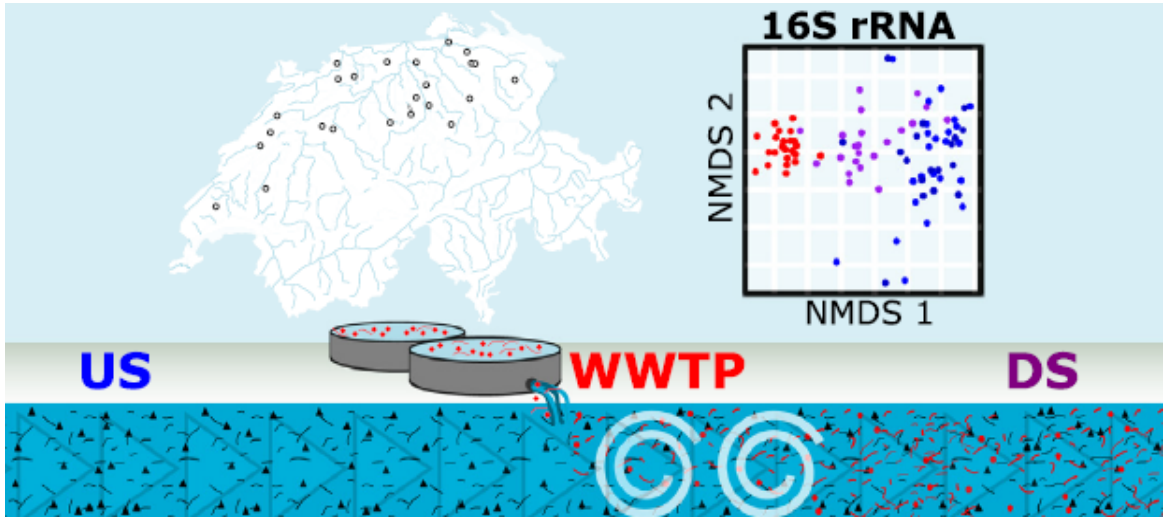
Using SourceTracker and hydrological equation to predict downstream mixing:



Using SeqENV (text mining pipeline):



Microbial community shifts in streams receiving treated wastewater effluent



- Mixing between the stream and the wastewater effluent predicted downstream community composition for most taxa
- 14 sites showed greater than 50 % of the bacteria taxa were from the wastewater
- Decreases in *phototrophic taxa* could not be explained by mixing alone
- Human-gut related bacteria are indicators of natural streams impacted by wastewater effluent
- Functional effects of these community shifts need further investigation

What is Environmental DNA?

- A molecule in some state (cell, particle bound, etc.) in the environment

How do we sample and sequence it?

- Purify DNA from soil, water air,
- methods are as diverse as your sample and they all have trade-offs! Try to understand and mitigate for these through good study design

What can we learn from it?

- Has and will continue to fundamentally change how we study the biosphere