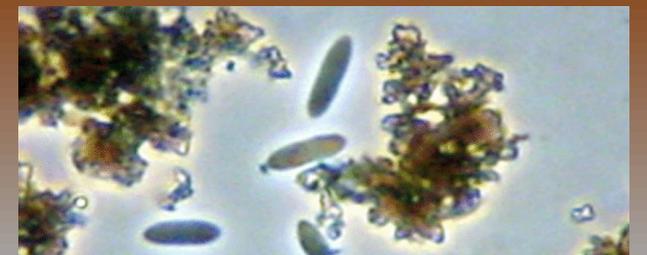


Large-scale study on groundwater dissolved organic matter reveals strong heterogeneity and a complex microbial footprint

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Groundwater Ecology Group



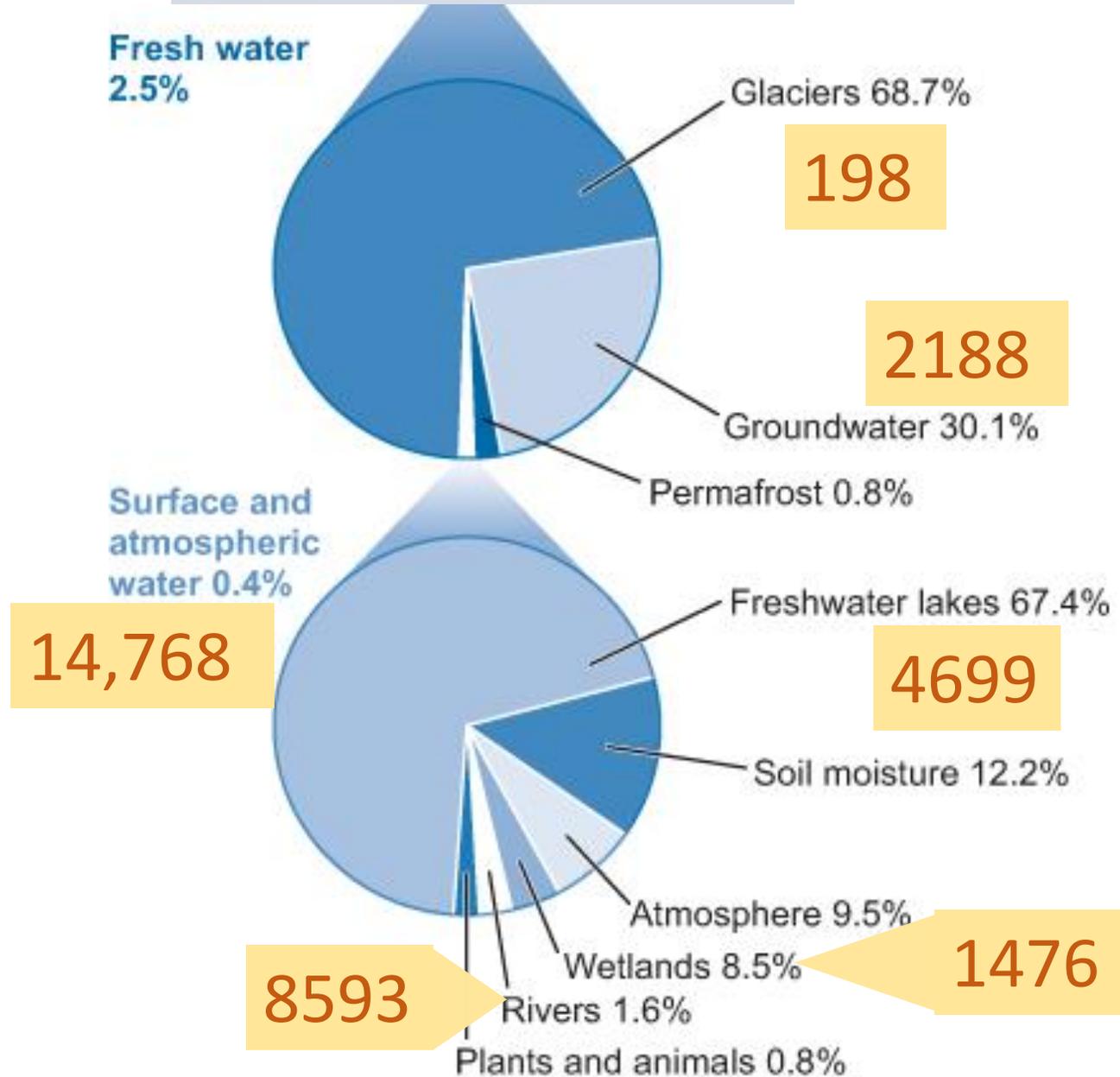
DOM in groundwater is understudied:

Groundwater makes 95% of liquid, available Freshwater, DOM papers are scarce. Only 12% of DOM papers contain „Groundwater“. Results from search in Web of Science.

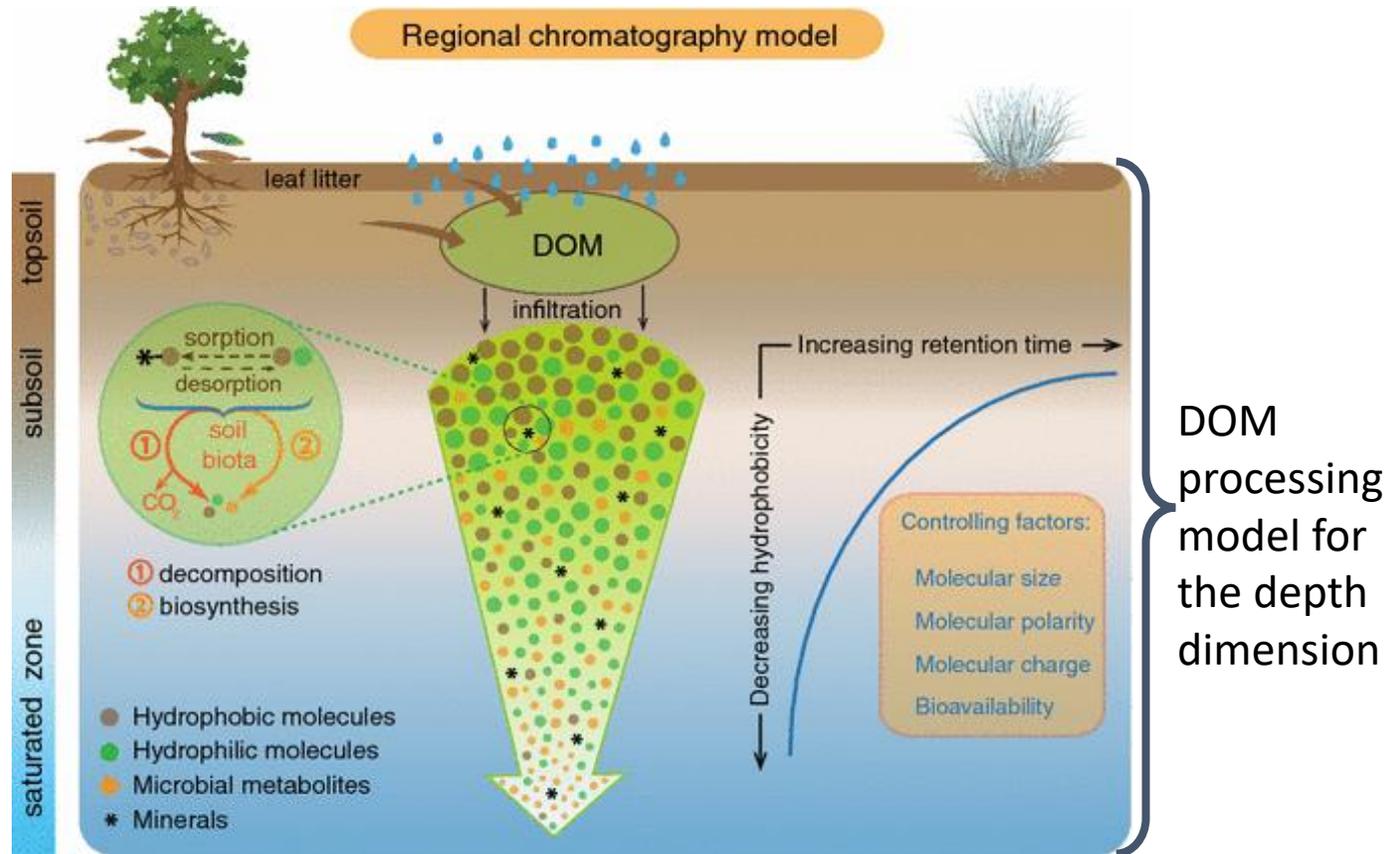
Publications on Dissolved Organic Matter (DOM)



Earth's Freshwater distribution

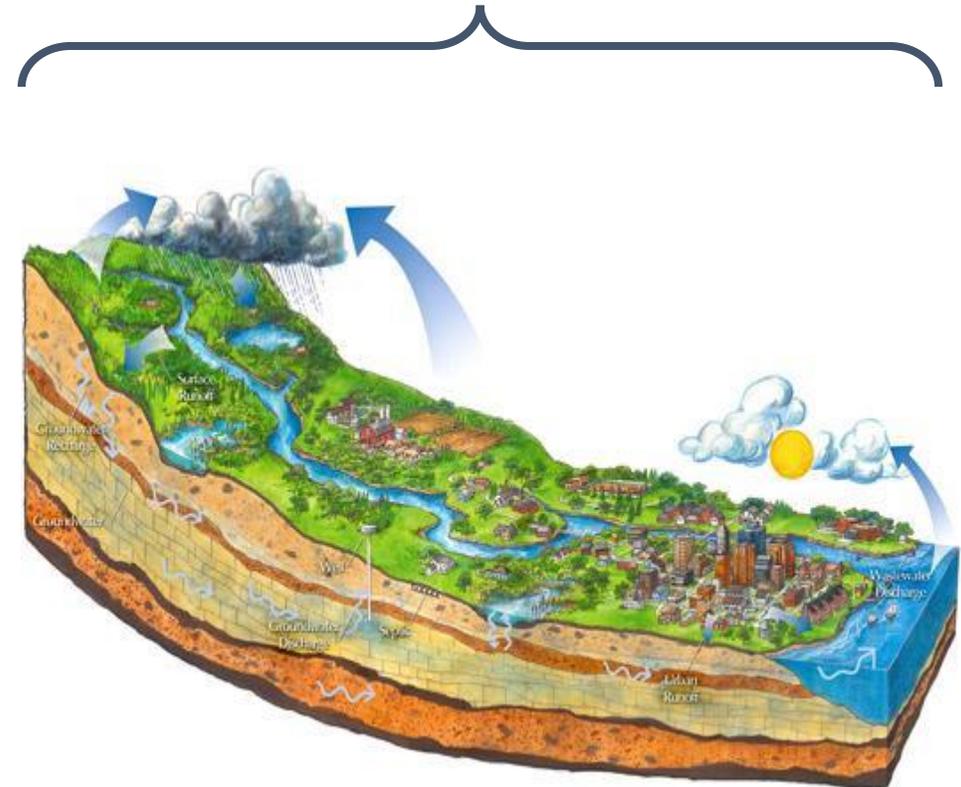


What do we know about DOM quality in groundwater bodies on the large scale?



Processing of DOM in the subsurface. With depth DOM molecules get smaller and are mainly composed of microbial metabolites (Shen et al. 2015).

What do we know about this dimension?
DOM quality across groundwater bodies,
geology, land use...



Hypotheses

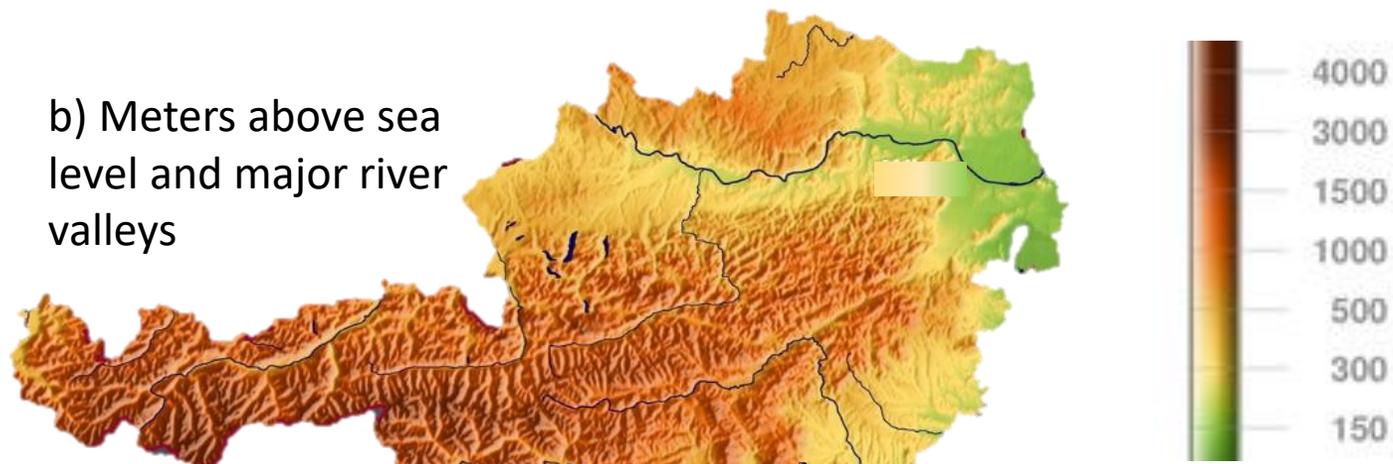
1. Ground water DOM shows an equally high chemical diversity as surface waters, however, due to sorption and longer residence times higher microbial imprint on DOM.
2. DOM quality is related to bacterial abundance in groundwater -> higher content of proteins and degraded DOM points towards higher bacterial abundance.
3. DOM quality can be related to land use and topology -> e.g. Agricultural watersheds have imprint on groundwater DOM.

Study Area

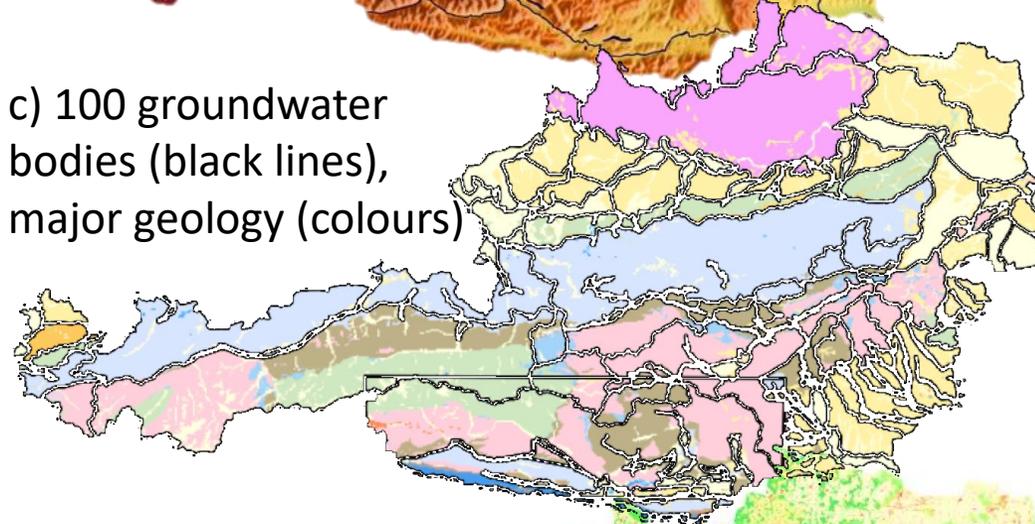
a) Land use map of Europe with Study area (b-d) in black



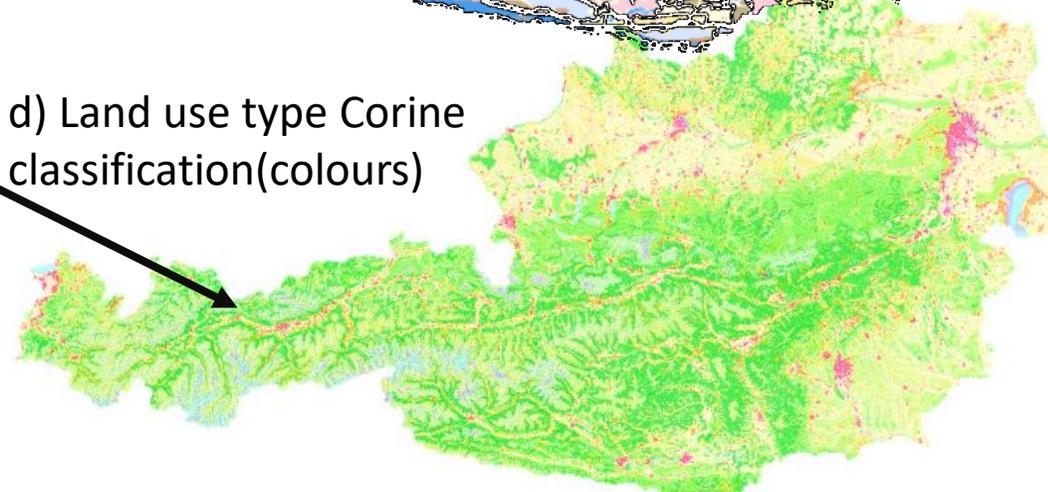
b) Meters above sea level and major river valleys



c) 100 groundwater bodies (black lines), major geology (colours)

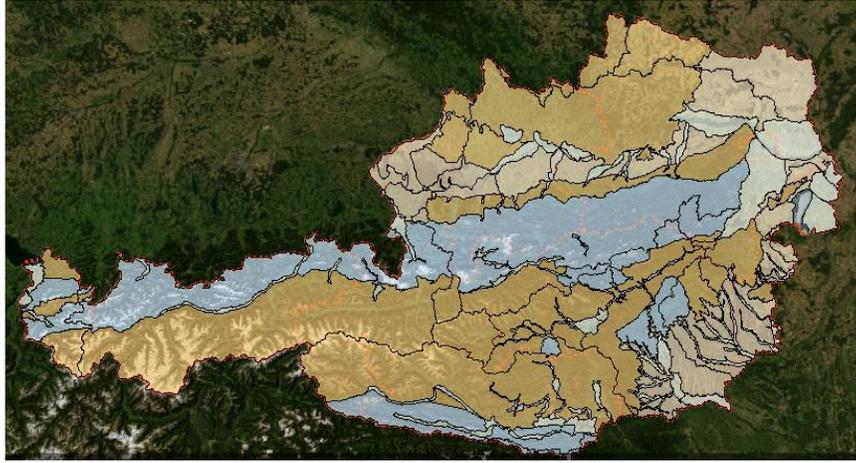


d) Land use type Corine classification (colours)

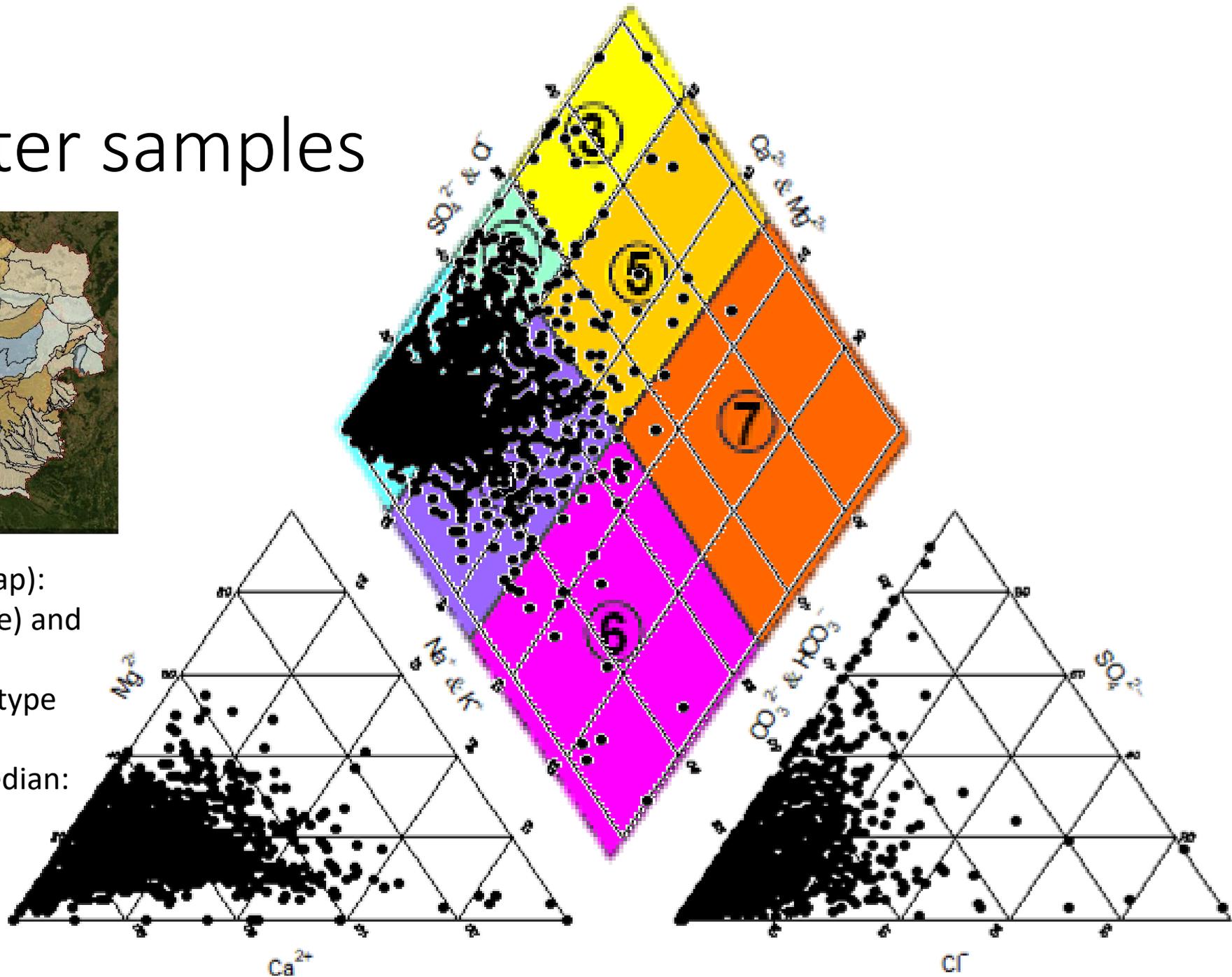


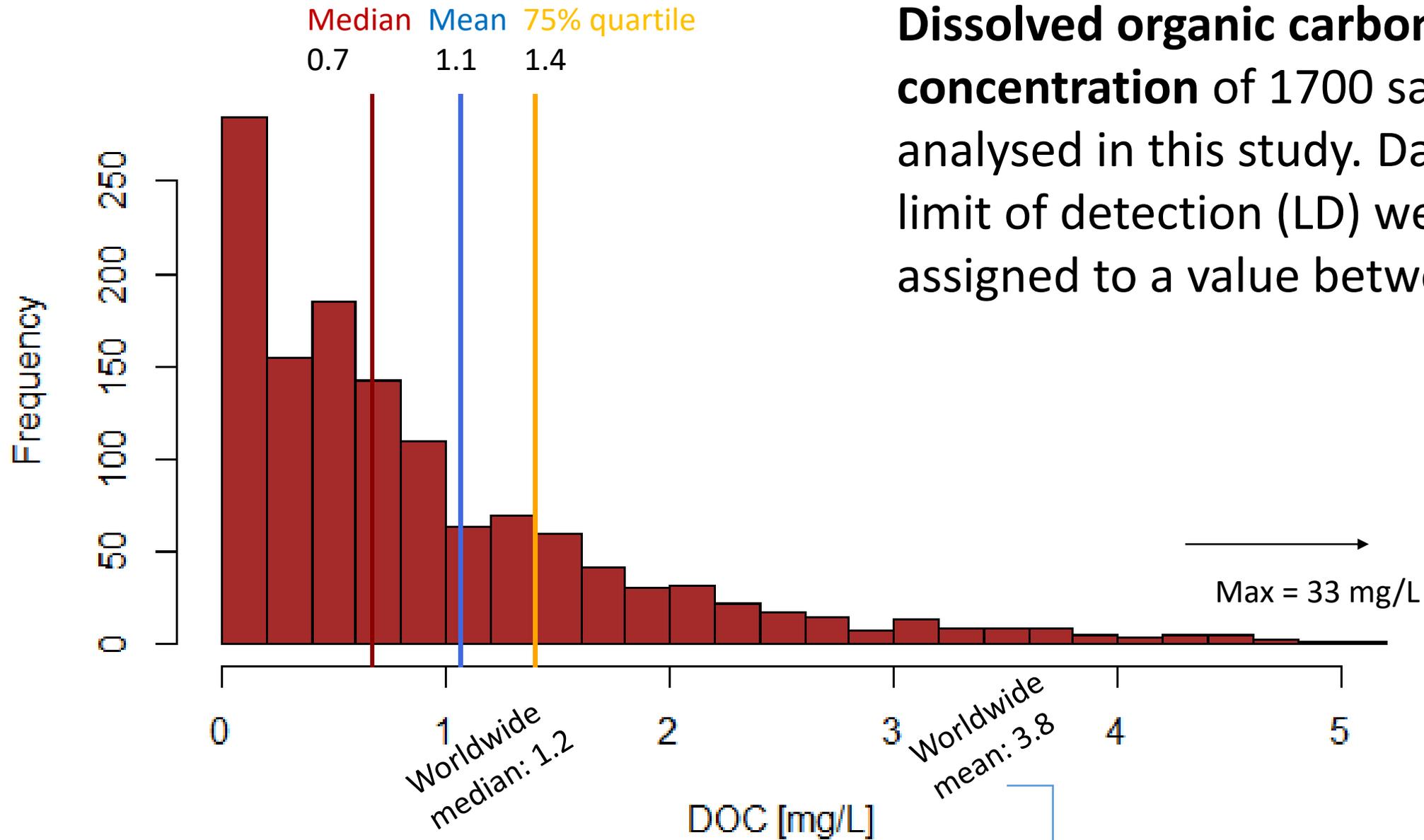
- | | |
|--|------|
| | 4000 |
| | 3000 |
| | 1500 |
| | 1000 |
| | 500 |
| | 300 |
| | 150 |
-
- | | |
|--|-------------------------|
| | Quarternary |
| | Tertiary Basins |
| | Bohemian Massif |
| | Helvetian Zone |
| | Penninic |
| | Calcarious Alps |
| | Greywacke |
| | Austroalpine cristaline |
-
- | | |
|--|---------------------------------|
| | Artificial areas |
| | Arable land and permanent crops |
| | Pastures and mosaics |
| | Forested land |
| | Semi-natural vegetation |
| | Open spaces/bare soils |
| | Wetlands |
| | Water bodies |

Groundwater samples



- 100 ground water bodies (see map): porous (light brown), karstic (blue) and fractured (dark brown)
- 90% Carbonate water chemistry type (blue area in the piper diagram)
- 0.5-80 meters below surface (median: 14 m)
- 100-2000 meters above sea level
- <5-50 years ground water age



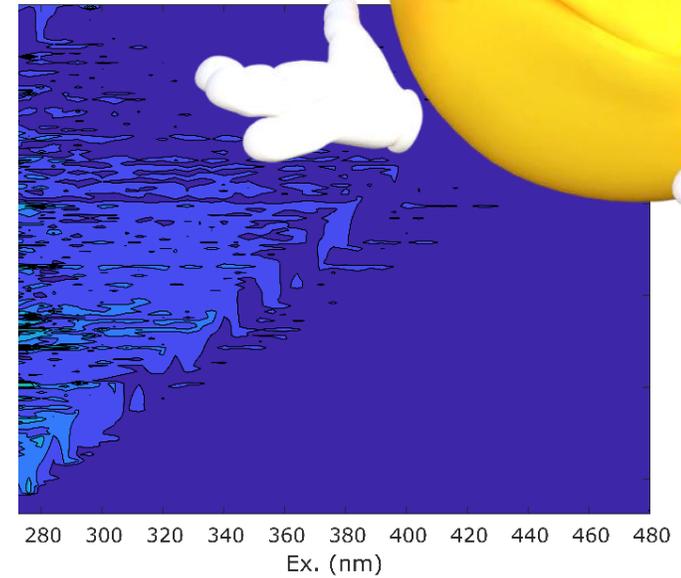
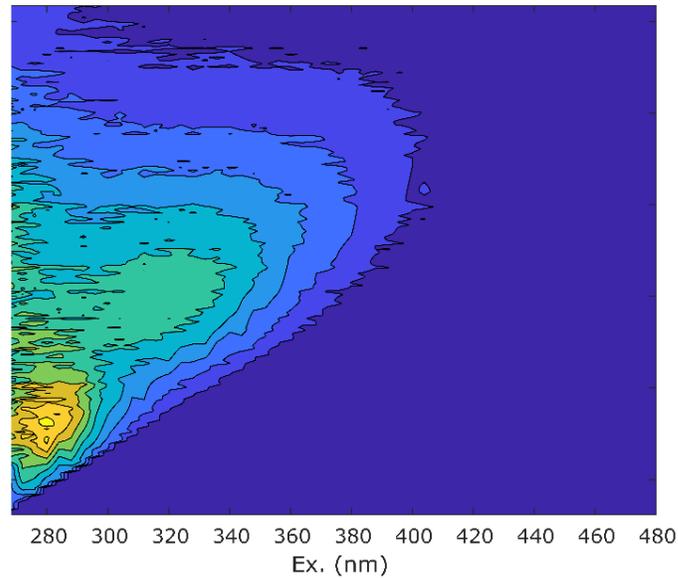
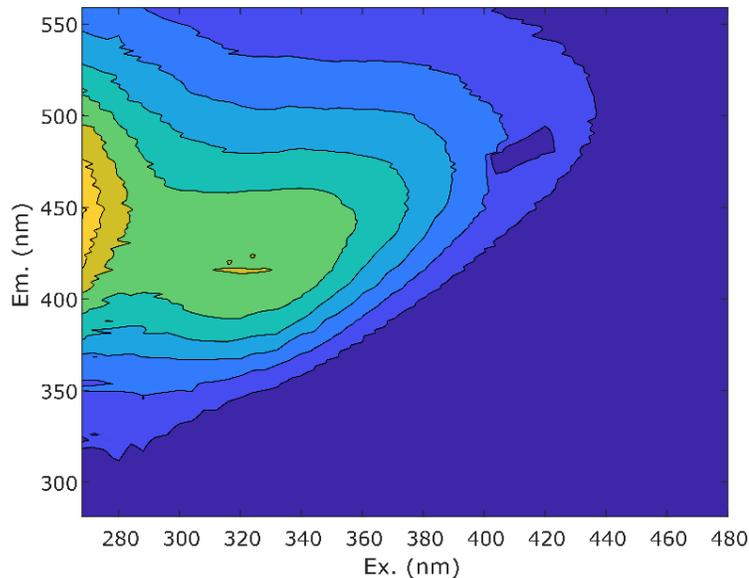


Dissolved organic carbon concentration of 1700 samples analysed in this study. Data below limit of detection (LD) were randomly assigned to a value between 0 and LD.

Data from McDonough et al. 2020.

Challenges for UV-Vis Fluorescence spectroscopy:

- 1700 Samples -> automated measuring procedure
- Some with very low concentrations -> low signal to noise ratio
- Unknown mixtures/clustering of samples as challenge for PARAFAC modeling of whole data set



Data treatment

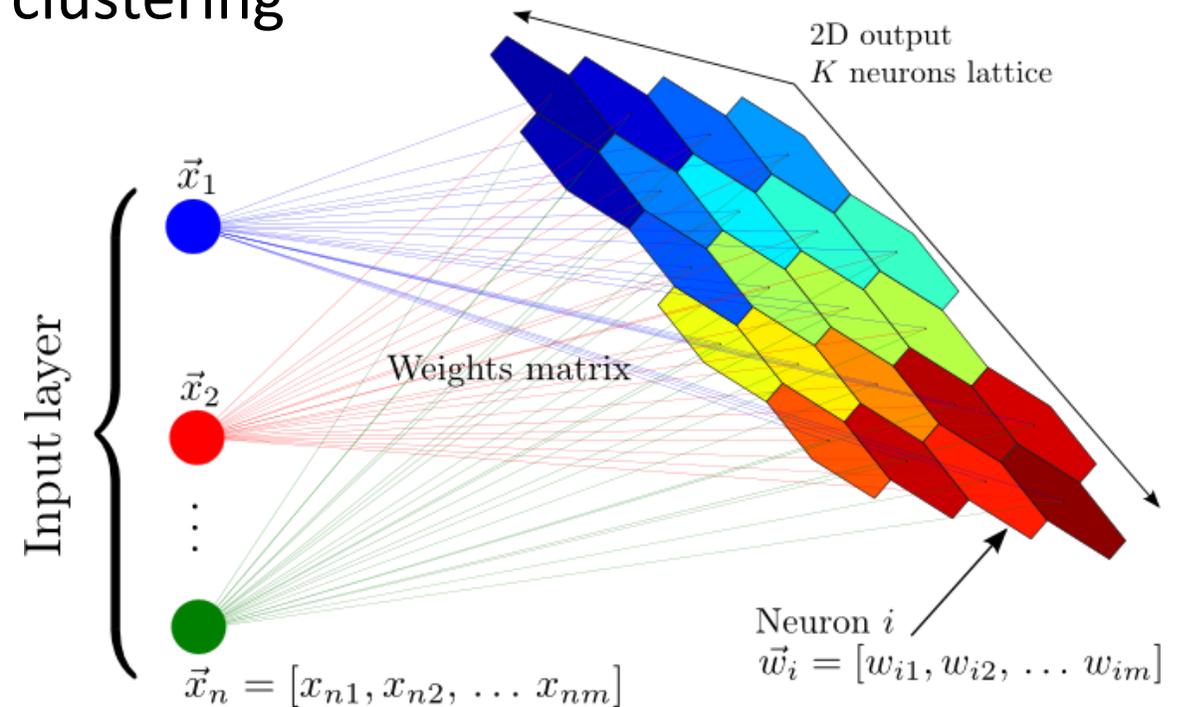
- Fluorescence indices
- Self-organizing map (Vesanto et al. 2000) of fluorescence excitation-emission matrices followed by Ward clustering

- Six Clusters:

for each Cluster PARAFAC models with 2-4 Components were derived

- Tuckers Convergence Criterion: at $TCC > 0.95$ was used to compile out of these six models

one global model with 9 Components

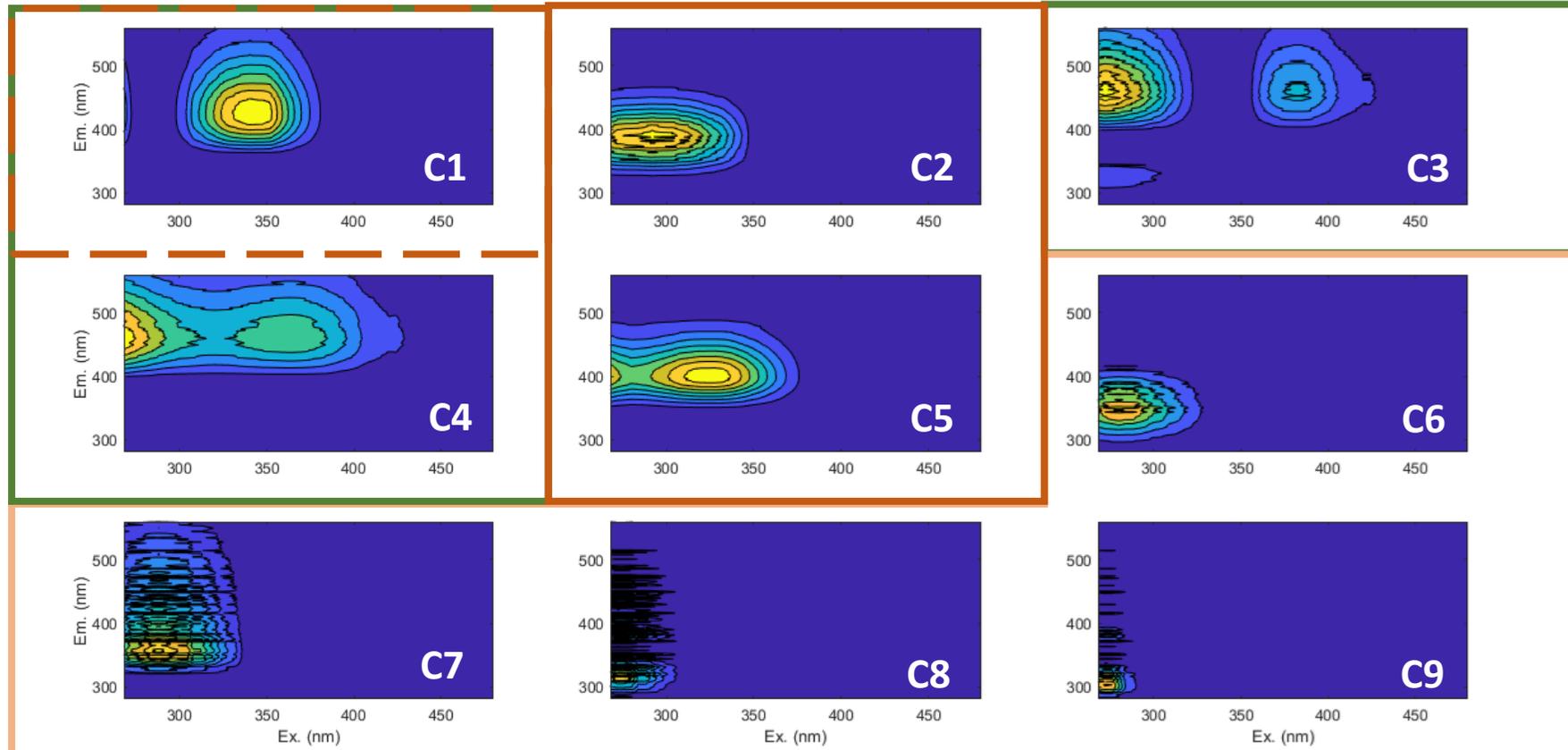


Resulting PARAFAC Model

Terrestrial, high molecular weight, found in soils and rivers, C3 also found in waste water, C4 in ground water, photodegraded rapidly

Humic, highly degraded, found in agricultural catchments, related to microbial degradation, abundant in the oceans (recalcitrance)

Protein-like substances, C6 and C7 found in wide range of surface water and soil, tryptophan-like, C8 and C9 found in the dark ocean

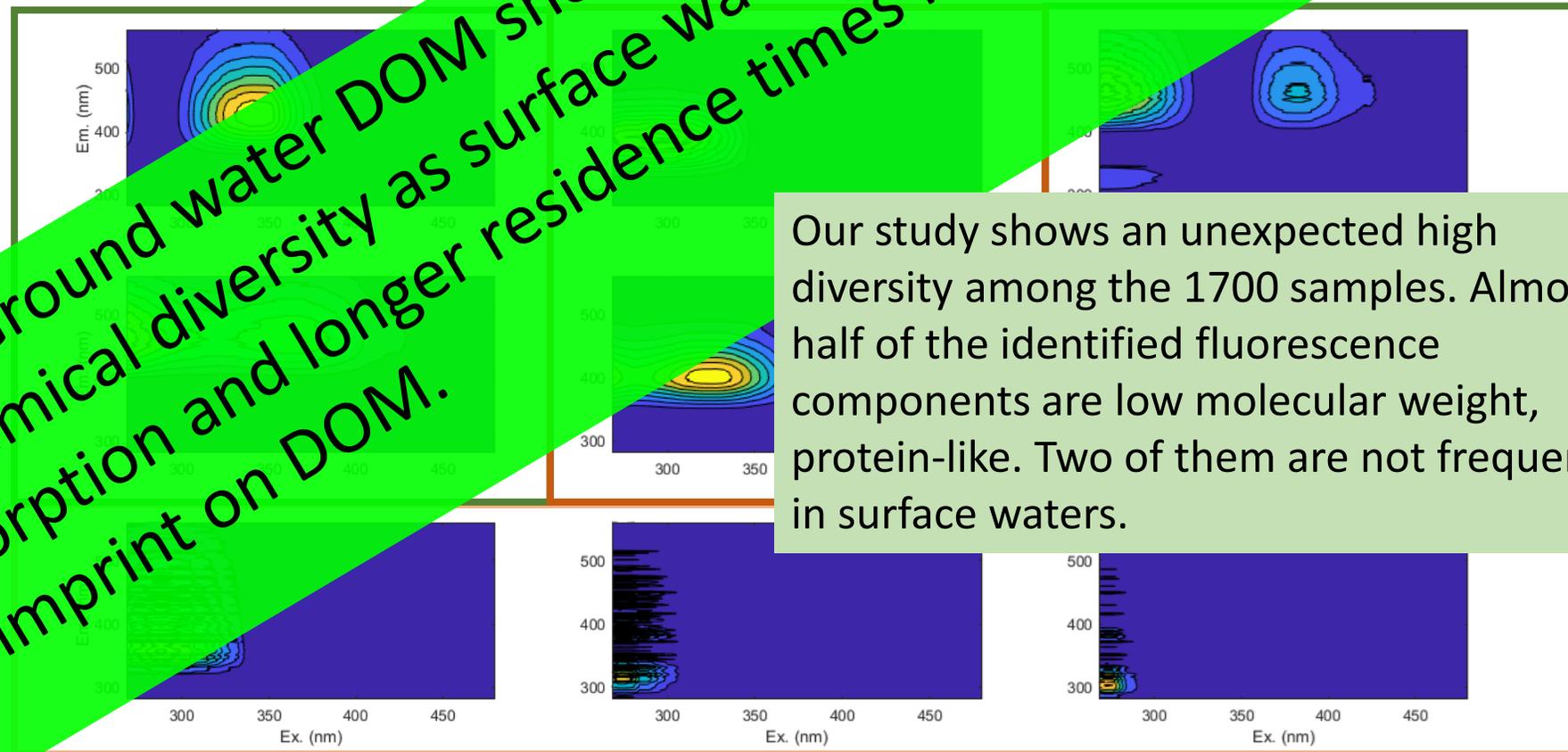


Resulting PARAFAC Model

Terrestrial, high molecular weight, found in soils and rivers, C3 also found in waste water, C4 in ground water, photodegraded rapidly

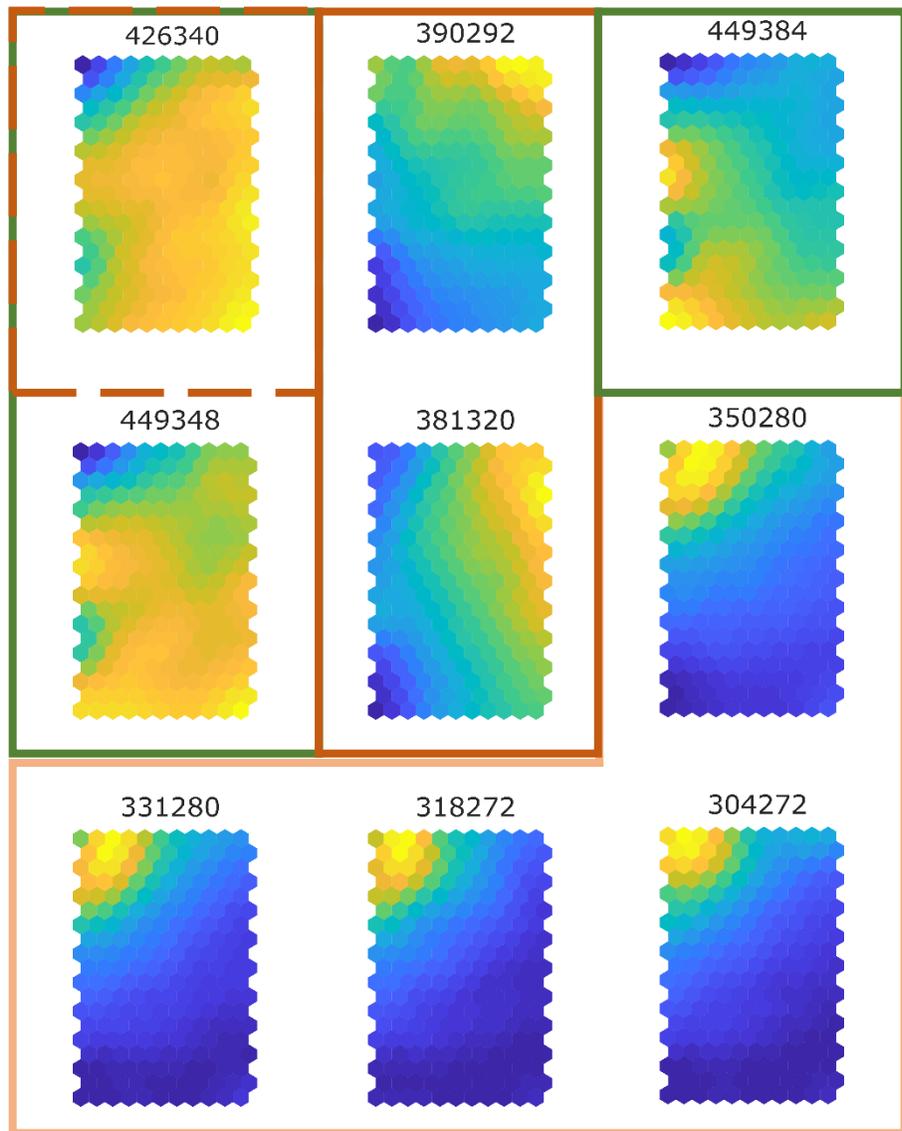
Humic, highly degraded, found in agricultural catchments, related to microbial degradation, abundant in the ocean (calcification)
Protein-like substances, C6 and C7 found in wide range of surface water and soil, tryptophan-like, C8 and C9 found in the dark ocean

1. H1: Ground water DOM shows an equally high chemical diversity as surface waters, however, due to sorption and longer residence times higher microbial imprint on DOM.



Our study shows an unexpected high diversity among the 1700 samples. Almost half of the identified fluorescence components are low molecular weight, protein-like. Two of them are not frequent in surface waters.

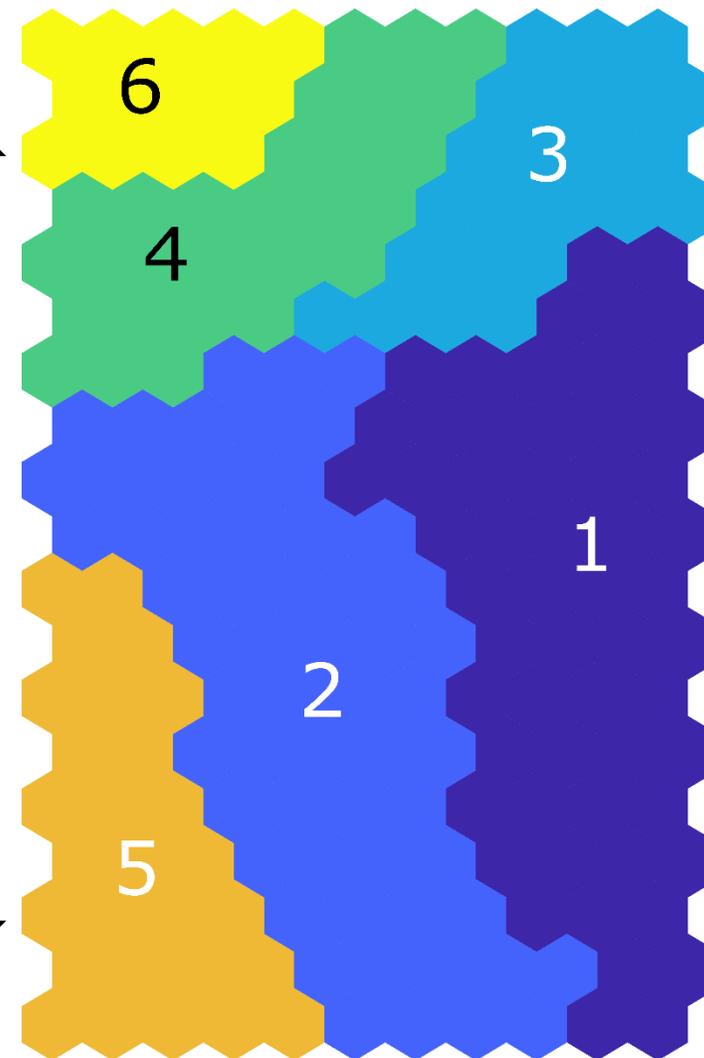
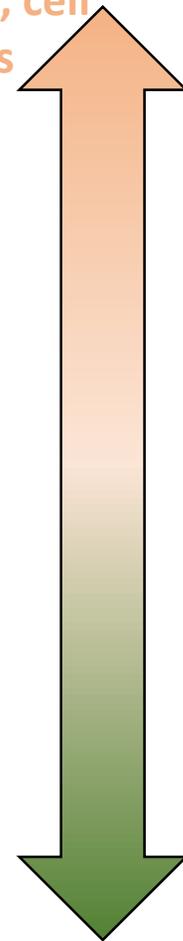
EmissionExcitation



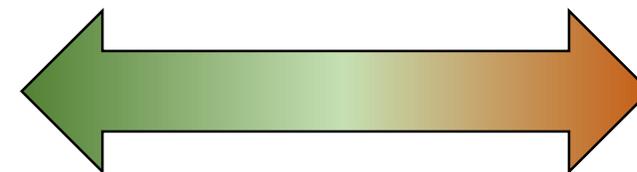
Self organizing map (SOM) component planes for all 9 components. Yellow areas show higher intensity of the respective component, dark blue lower.

Six Ward Clusters

Protein-like, cell lysis



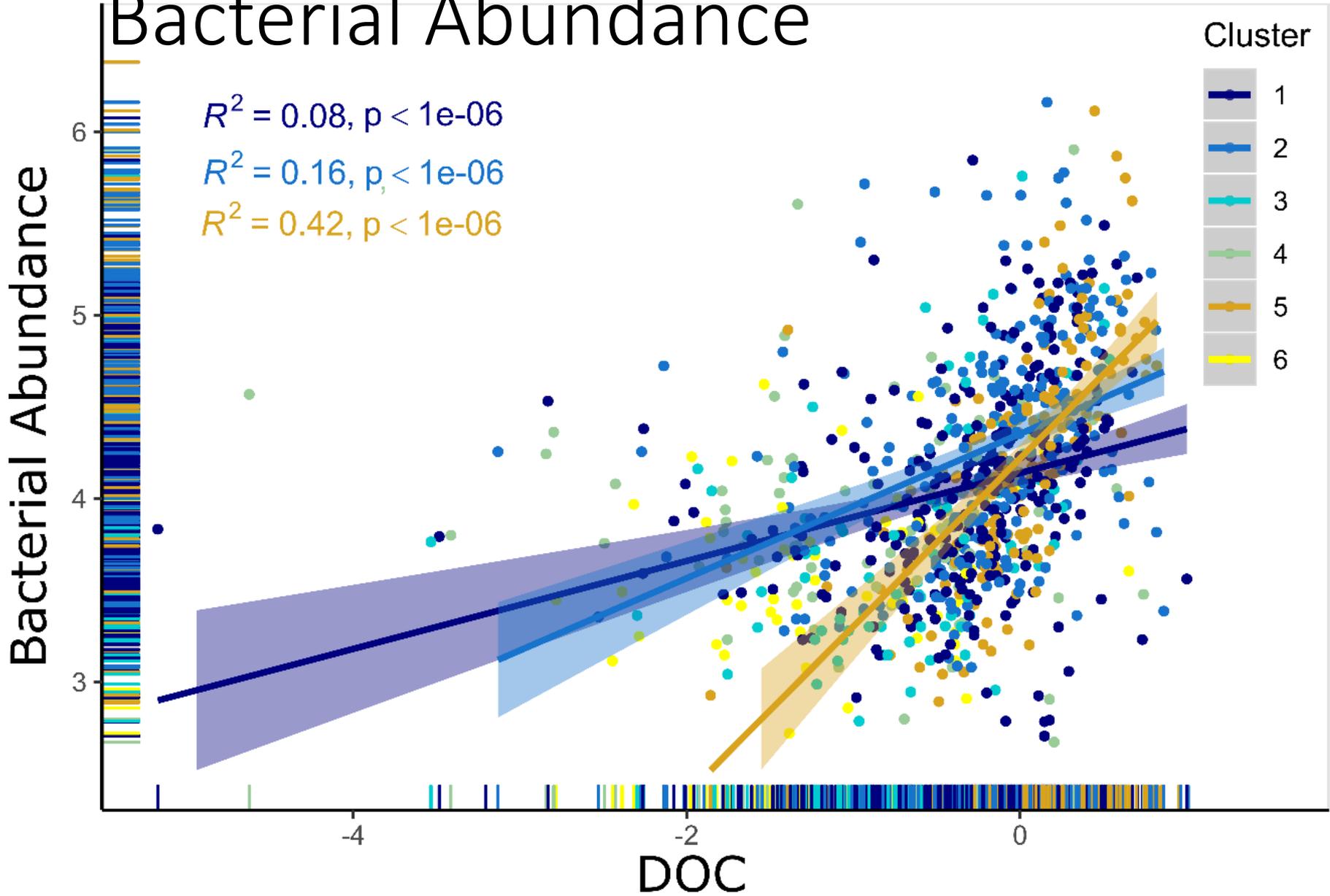
Terrestrial humic



Degraded humic

Bacterial Abundance

$R^2 = 0.08, p < 1e-06$
 $R^2 = 0.16, p < 1e-06$
 $R^2 = 0.42, p < 1e-06$



Results of linear regressions with log transformed bacterial abundance and DOC concentrations for each DOM cluster. Only clusters with a high portion of terrestrial DOM showed a significant correlation. In particular cluster 5 (brown) did not show any protein-like fluorophore in the individual PARAFAC model.

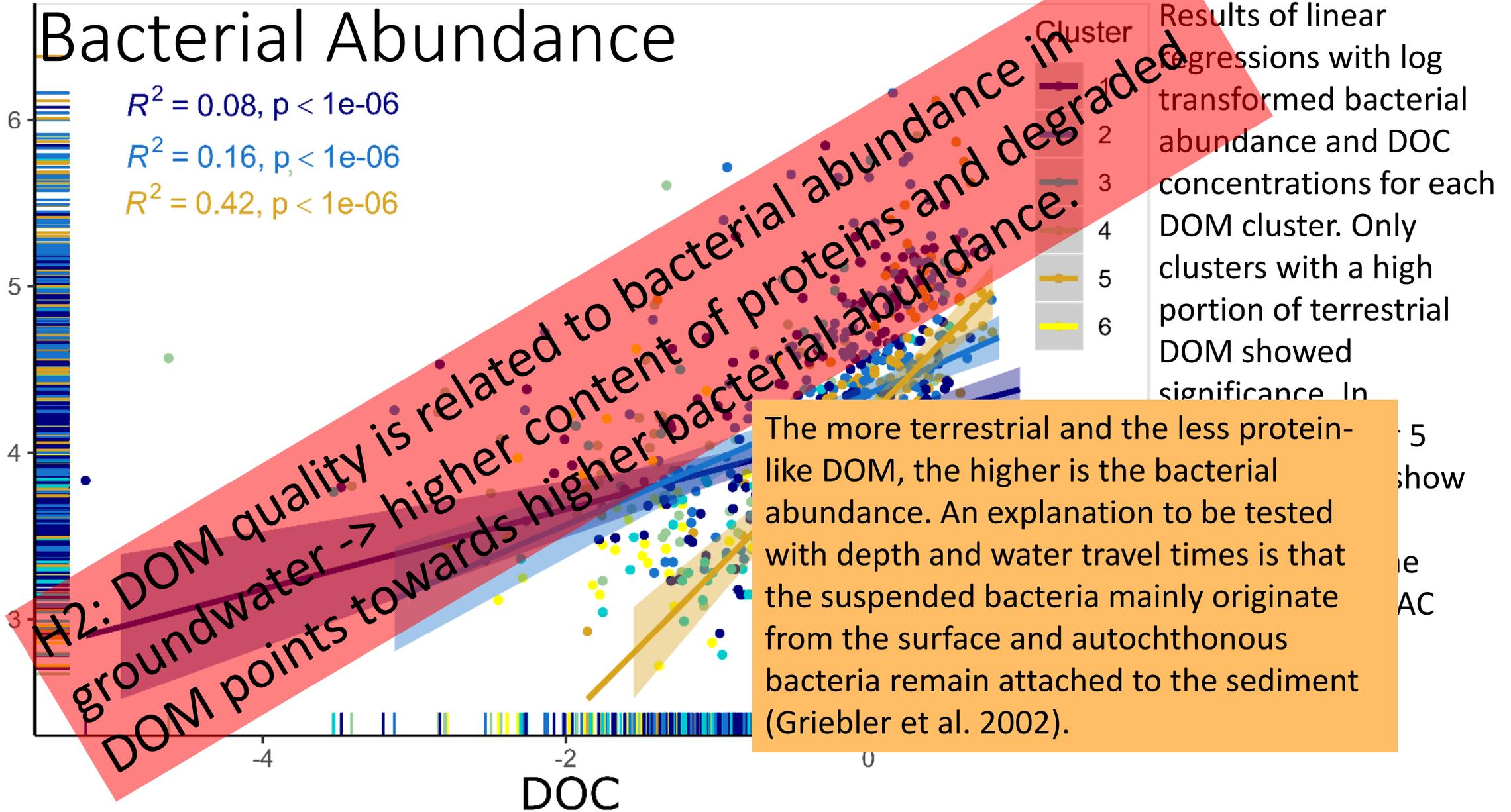
Bacterial Abundance

$R^2 = 0.08, p < 1e-06$

$R^2 = 0.16, p < 1e-06$

$R^2 = 0.42, p < 1e-06$

Bacterial Abundance



H2: DOM quality is related to bacterial abundance in groundwater -> higher content of proteins and degraded DOM points towards higher bacterial abundance.

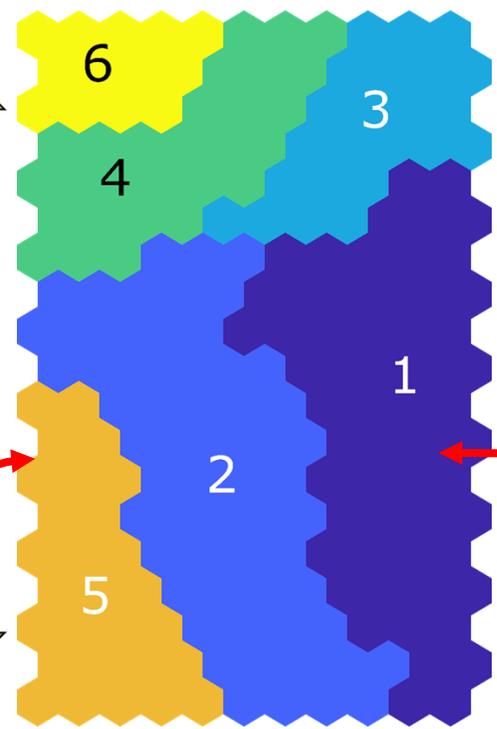
The more terrestrial and the less protein-like DOM, the higher is the bacterial abundance. An explanation to be tested with depth and water travel times is that the suspended bacteria mainly originate from the surface and autochthonous bacteria remain attached to the sediment (Griebler et al. 2002).

Results of linear regressions with log transformed bacterial abundance and DOC concentrations for each DOM cluster. Only clusters with a high portion of terrestrial DOM showed significance. In

Land use and DOM

Six Ward Clusters

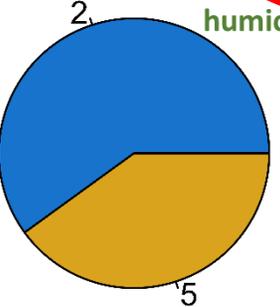
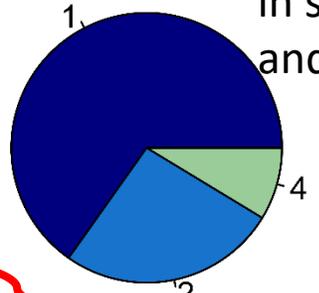
Protein-like, cell lysis



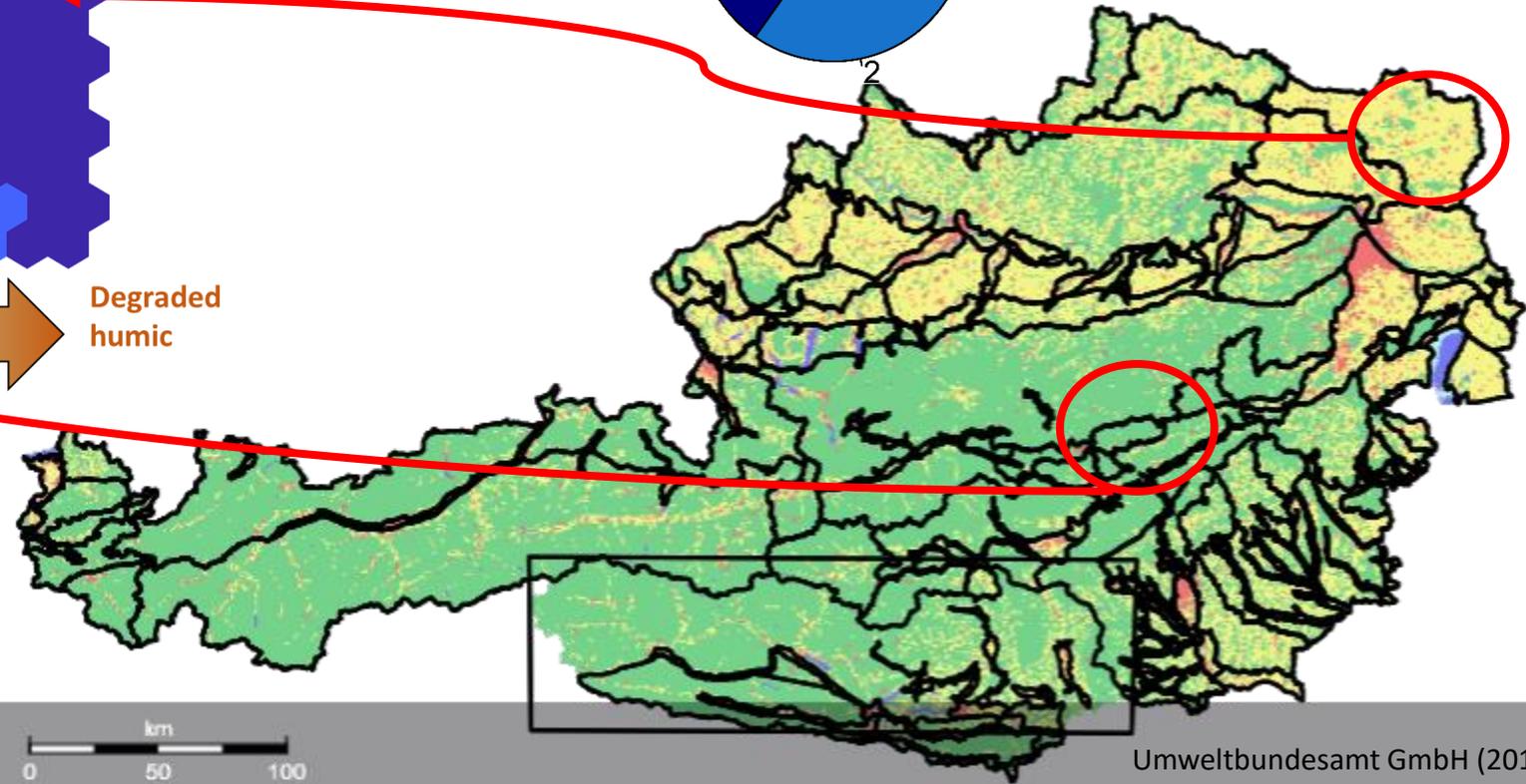
Terrestrial humic

Degraded humic

Dominated by highly degraded humics, in surface waters found in agricultural and wastewater impacted catchments

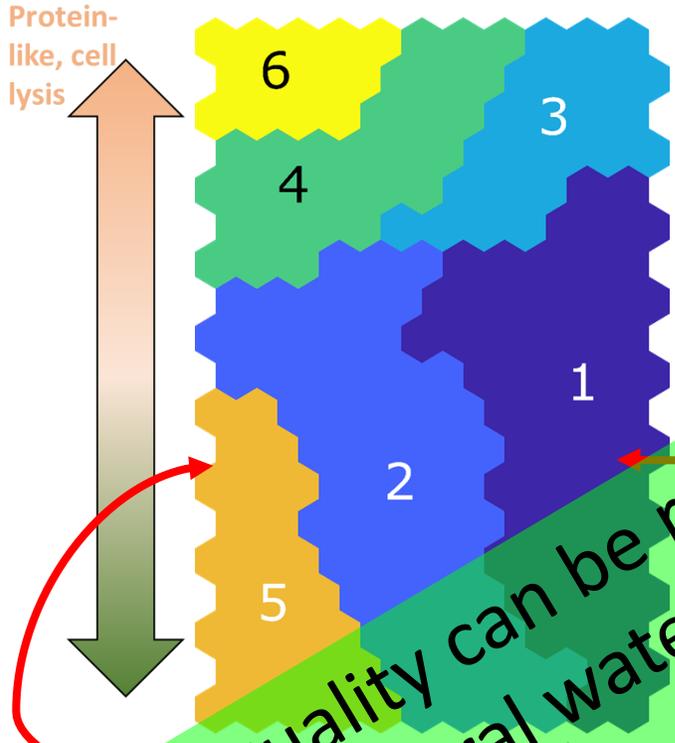


Dominated by terrestrial, aromatic, high molecular weight compounds found in forested catchments

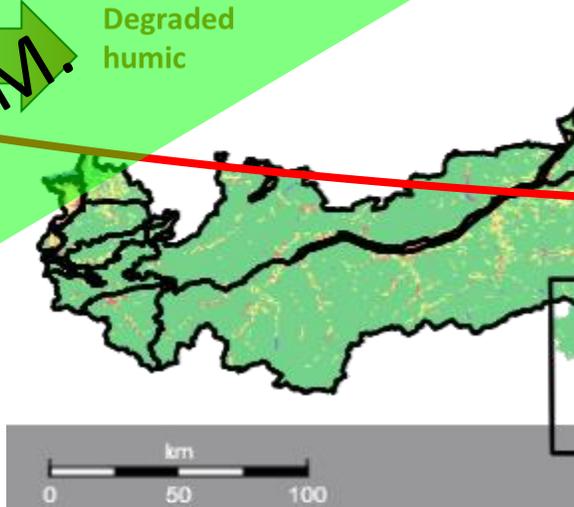


Land use and topography

Six Ward Clusters



H3: DOM quality can be related to land use and topography
-> e.g. Agricultural watersheds have imprint on groundwater DOM.



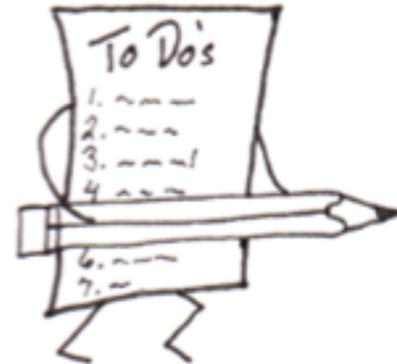
First tests on groundwater bodies showed that >70% of the samples of each groundwater body fell in one to two clusters. Here are shown two examples one from an agricultural area in the low lands in Cluster 1. This Cluster is characterized by DOM components found in agricultural catchments and waste water. Interestingly, this groundwater body has particularly old groundwater >50y, suggesting that the DOM is recalcitrant to microbial metabolism. The other one is located in an forested, karstic area. Cluster 5 shows components found in forested catchments.

Outlook

We found clear indications that DOM processing takes place in the subsurface, however **suspended bacteria** cells seem to have been flushed together with unprocessed **surface** DOM into the groundwater. Highly degraded DOM from **agricultural** areas seem to be **recalcitrant** in the aquifer.

To further evaluate and refine this findings we will examine individual groundwater bodies and catchments. We will relate the DOM quality and bacterial abundance to

- depth
- water age
- water chemistry
- land use in the recharge area
- many more... ideas?



References

- Griebler, C., Mindl, B., Slezak, D., & Geiger-Kaiser, M. (2002). Distribution patterns of attached and suspended bacteria in pristine and contaminated shallow aquifers studied with an in situ sediment exposure microcosm. *Aquatic microbial ecology*, 28(2), 117-129.
- McDonough, L. K., Santos, I. R., Andersen, M. S., O'Carroll, D. M., Rutledge, H., Meredith, K., ... & Lapworth, D. J. (2020). Changes in global groundwater organic carbon driven by climate change and urbanization. *Nature communications*, 11(1), 1-10.
- Shen, Y., Chapelle, F.H., Strom, E.W. et al. Origins and bioavailability of dissolved organic matter in groundwater. *Biogeochemistry* 122, 61–78 (2015).
- Umweltbundesamt GmbH 2012. Corine land cover. & H2O Fachdatenbank.
- Vesanto, J., Himberg, J., Alhoniemi, E., & Parhankangas, J. (2000). SOM toolbox for Matlab 5. *Helsinki University of Technology*, 216.
<http://www.cis.hut.fi/projects/somtoolbox/>

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und Tourismus

umweltbundesamt^u
PERSPEKTIVEN FÜR UMWELT & GESELLSCHAFT