

Functional and taxonomic microbial diversity profiling across soil depth in a diversified cover cropping system in the Netherlands

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Research question and hypothesis

- Microbial processes play an important role in carbon cycling
 - Crop diversification (cover crops in this case) increase chemical complexity of organic matter input
 - This can affect microbial physiology and its relationship with carbon dynamics
- Main hypothesis: greater crop diversity results in more efficient microbial use of carbon, thus enhancing soil carbon storage
- How does this effect expand into deeper soil layers?
 - SOC and C:N ratio of organic material decrease with depth
 - While a narrower C:N ratio may positively influence CUE, nutrient scarcity may negatively affect CUE
 - Microbial diversity may play an important role in regulating microbial CUE
- Main research question: What is the effect of cover crop diversification and soil depth on functional and taxonomic microbial diversity?

Clever Cover Cropping Experiment

Three cover crop types



Raphanus sativus
(Radish)

- High biomass
- C:N ratio ~ 13
- Taproot
- Allelochemicals

Avena strigosa
(Oat)

- High biomass
- C:N ratio ~16
- Fibrous roots
- Winter hardy

Vicia sativa
(Vetch)

- Low biomass
- C:N ratio ~10
- Fibrous roots
- Biological N fixation

Porre, 2020

and their combinations in mixtures



Figure 2: Drone image of the field including plot borders and treatment description. Letters inside plots indicate all different cover crop treatments with Fal: fallow treatment, V: *V. sativa* (vetch), R: *R. sativus* (radish), A: *A. strigosa* (oat) and combinations of letters are two or three species mixtures. The experimental plots are 6*10 m, space between the plots is 3 m within a block and 9 m between blocks.

Drost, 2022

Crop rotation and sampling moment

2016: winter wheat -> start of the experiment -> cover crops

2017: maize -> cover crops

2018: potato -> cover crops

2019: barley -> cover crops

2020: pea -> cover crops

2021: barley -> cover crops

2022: maize -> cover crops -> sampling in December (0-30 and 30-60 cm depth)

Collected data

- Amplicon-sequestration of prokaryotes (16S) and fungal (ITS) DNS
- Fungal and bacterial growth
- Phospholipid-derived Fatty Acids (PFLA)
- Microbial biomass C and N
- Soil microbial carbon use efficiency (CUE)

Microbial growth and biomass

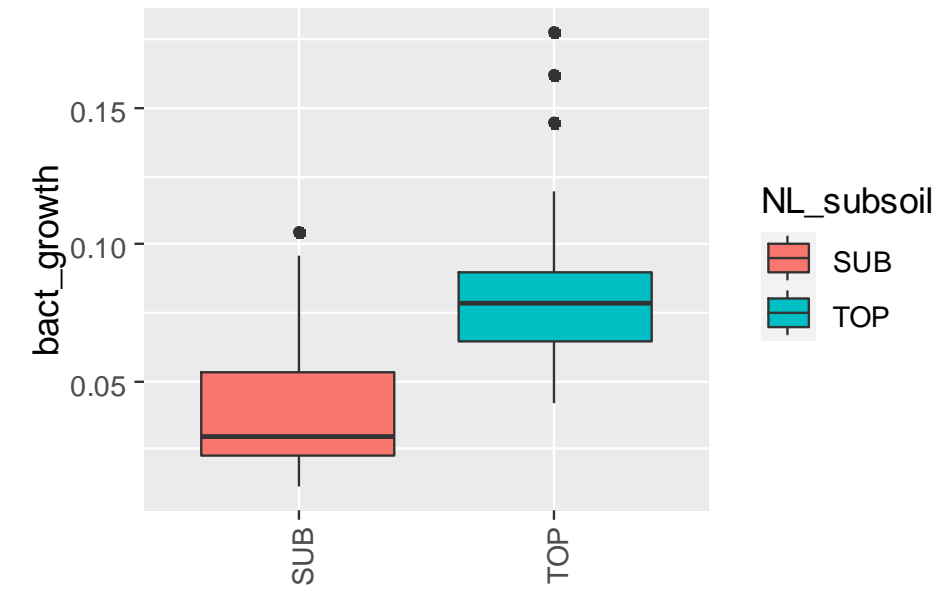
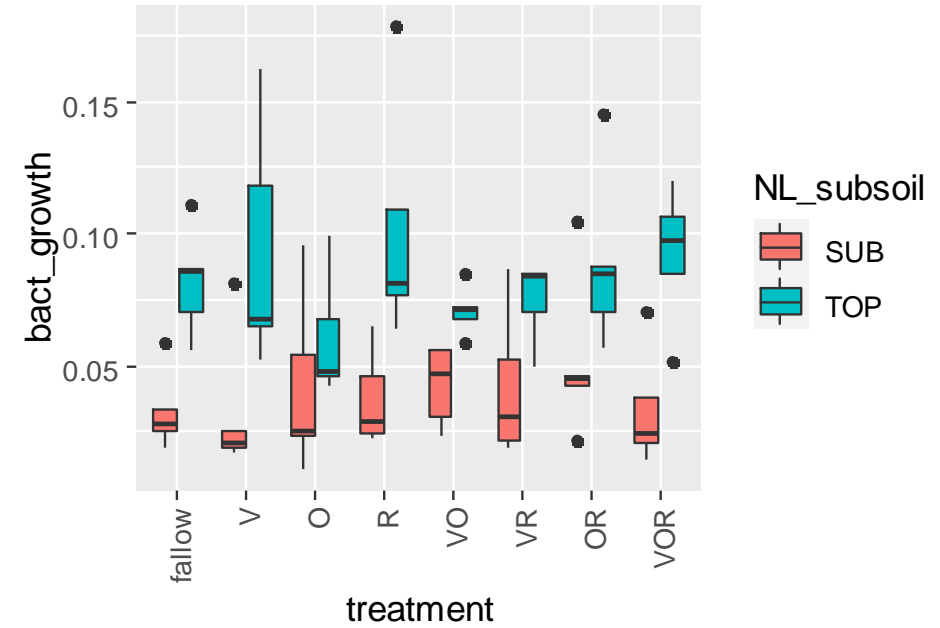
- Univariate statistics on the different variable: bacterial and fungal growth, PLFA, microbial biomass C and N

No significant effect of treatment

Same results found for the other variable

Bacterial growth significantly higher in topsoil

Example of bacterial growth



Alpha diversity

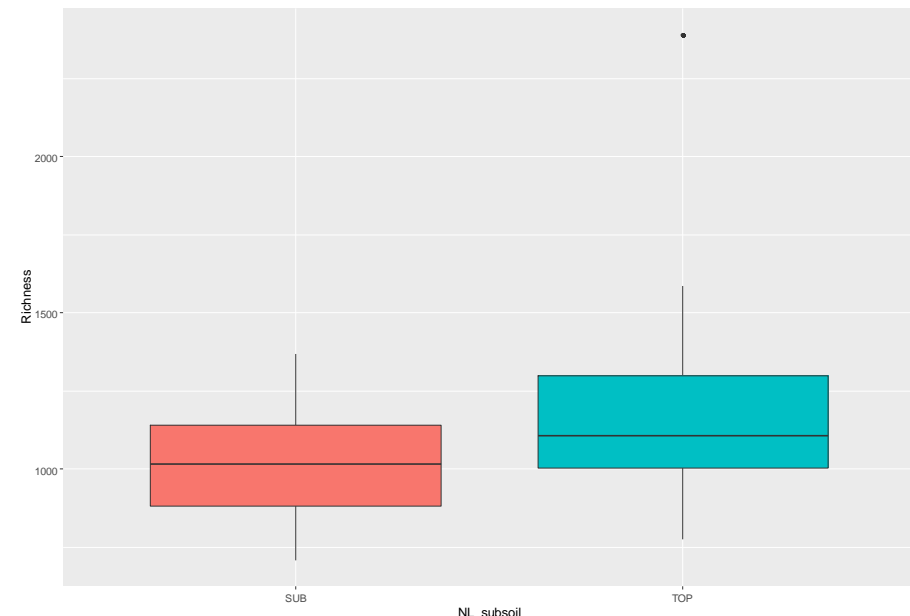
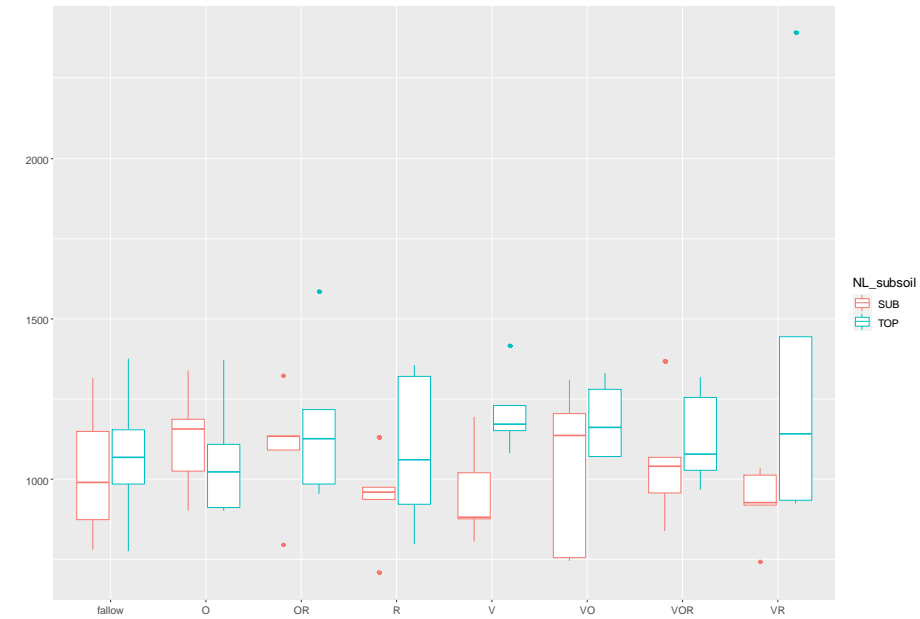
- Univariate statistics on the different variable: bacterial and fungal growth, PLFA, microbial biomass C and N
- Univariate statistics on 16S and ITS DNA sequestration for richness and Shannon diversity

Same results found for 16S Shannon diversity,
➤ But not significant for ITS richness and Shannon diversity

No significant effect of treatment

Richness is significantly higher in topsoil

Example of 16S Richness



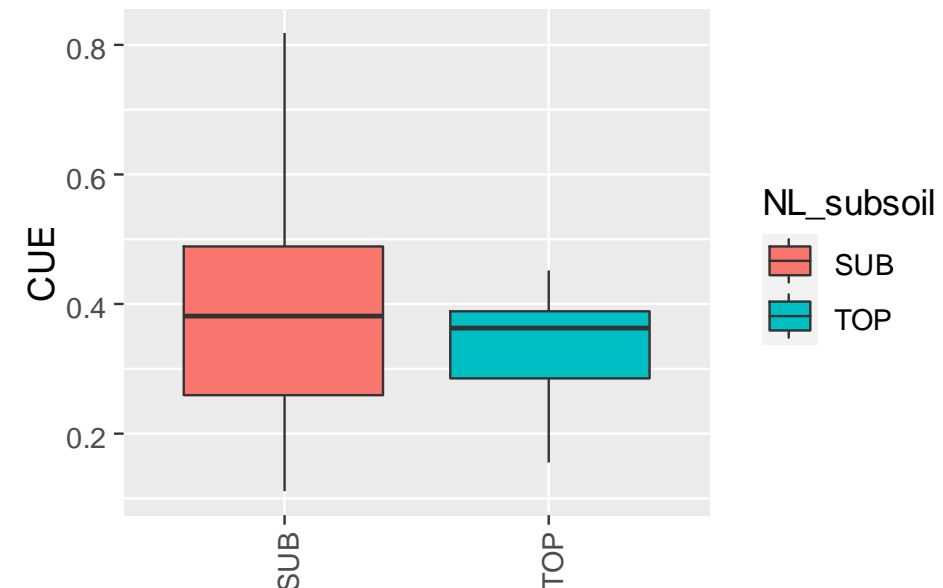
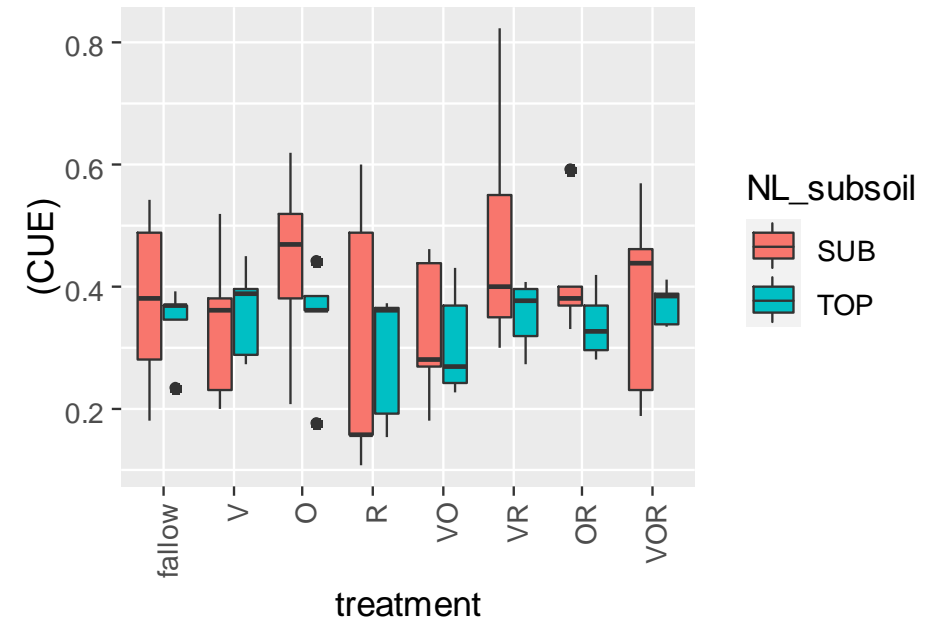
Carbon use efficiency

- Univariate statistics on the different variable: bacterial and fungal growth, PLFA, microbial biomass C and N
- Univariate statistics on 16S and ITS DNA sequestration for richness and Shannon diversity
- Univariate statistics on CUE

No significant effect of treatment

For CUE subsoil is higher, but not significant

Microbial carbon use efficiency



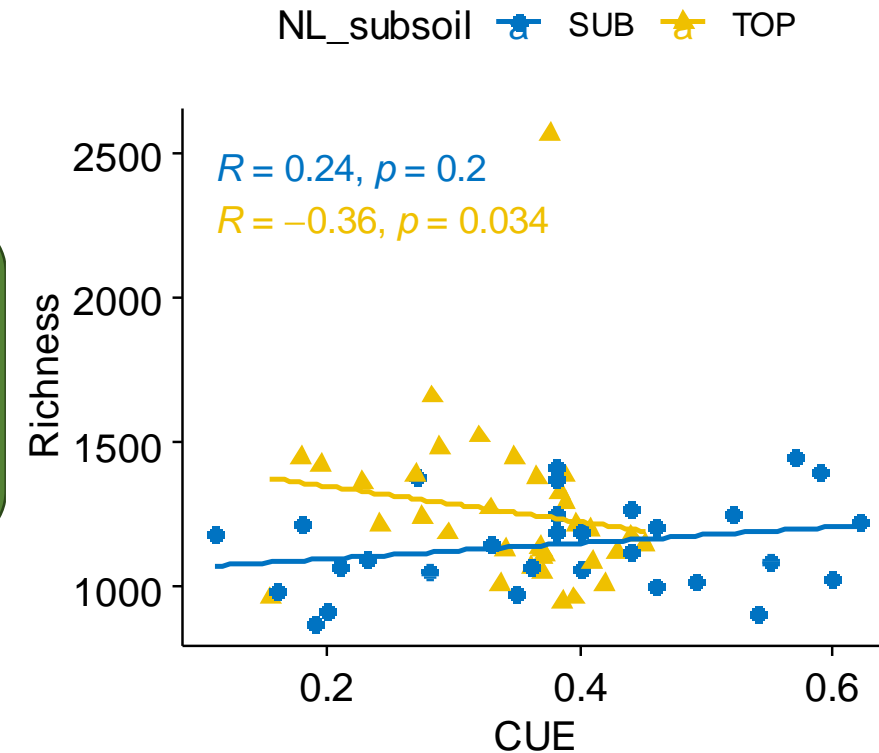
Alpha diversity and CUE

- Univariate statistics on the different variable: bacterial and fungal growth, PLFA, microbial biomass C and N
- Univariate statistics on 16S and ITS DNA sequestration for richness and Shannon diversity
- Univariate statistics on CUE
- Spearman correlation between CUE and 16S and ITS richness and Shannon diversity

Same correlations found for bacterial and fungal diversity, but only significant for bacterial diversity in topsoil

The correlations has different directions in the top- and subsoil

Example of CUE and 16S richness



Beta diversity

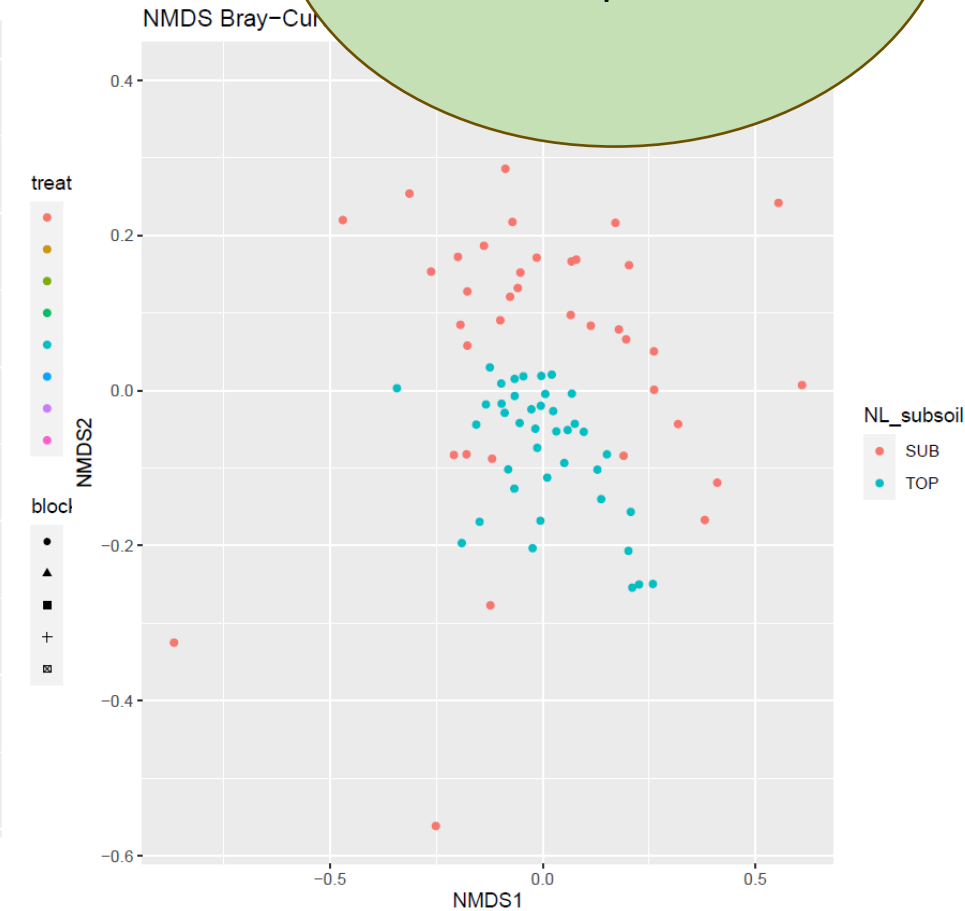
➤ PERMANOVA on 16S and ITS DNA sequestration

8% of fungal community differs between treatments
➤ For bacterial community this is only 2% (not significant)

4% of fungal community differs between top- and subsoil
➤ For bacterial community this is 9%

Example of ITS

Block effect must be further analysed and the correlation with other soil parameters



Discussion and conclusion

- Increased cover crop diversification did not affect functional and taxonomic microbial diversity
- Alpha diversity is significantly higher in topsoil, compared to the subsoil
- In the topsoil higher bacterial diversity is correlated with lower CUE
- Both cover crop diversification and soil layer significantly affect beta diversity

Thank you for your attention, any question?

