

# Re-colonization of sterile soil samples during long term field exposure – concept and first results

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## 1. INTRODUCTION

- Heterogeneous distribution of SOM in subsoils (Heinze et al. 2018)
- Localized input of fresh substrate and nutrients from rhizodeposition and preferential flow paths forming hotspots of microbial activity (Wang et al. 2013)
- Non-hotspot soil contains substantial amounts of labile substrates that are readily mineralized during lab incubation experiments
- Spatially separation of consumers from these substrates due to the low microbial densities in subsoils

**Goal:** Investigation of the **re-colonization potential** of microorganisms by exposing 24-well microplates containing **sterilized soil samples** in the field at two different depths (topsoil: 10 cm, subsoil 60 cm) at a beech forest site in northern Germany.

### We expect:

- (1) different temporal dynamics of re-colonization between top- and subsoil samples;
- (2) that the re-colonization potential is related to the microbial activity in the soil compartments above the exposed samples and
- (3) that the heterogeneous re-colonization is maintained throughout the field exposure and thus indicates the relevance of preferential flow paths for microbial transport especially in subsoils.

### Materials and methods:

- Sampling: April 2019, November 2019, April 2020, November 2020
- Enzyme activities (hydrolytic exo-enzymes involved in different nutrient cycles using MUF and AMC substrates)
- Microbial activity parameters (soil respiration and SIR using the MicResp® system)
- Spatial distribution of enzyme activities (on selected samples after final sampling)
- Real-time quantitative PCR (16s bacteria taxa, archaea and fungi)

## 2. CONCEPT AND FIRST RESULTS



1a, 1b and 1c Bringing out the containers with sterile soil samples (Nov 2018)

2 First field campaign (April 2019)

3a and 3b Separating the soil samples from each well (May 2019)

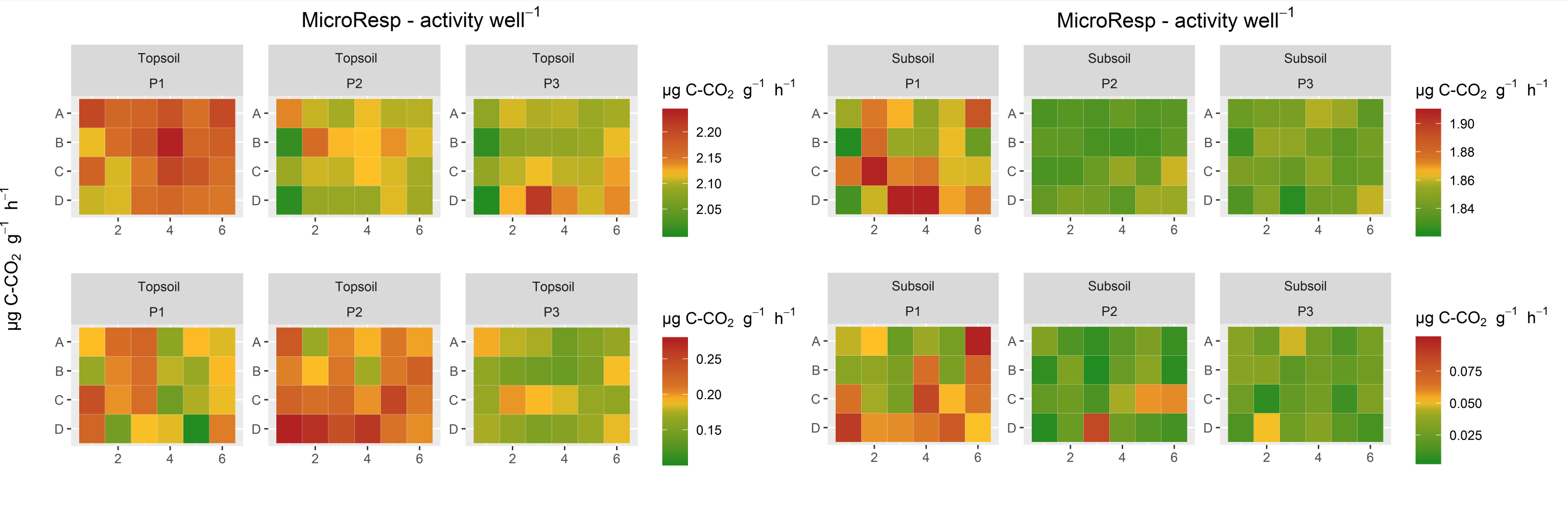


Fig 4a & 4b: SIR assay for undisturbed (top row) and re-colonized samples (bottom row) of topsoil (left) and subsoil (right).

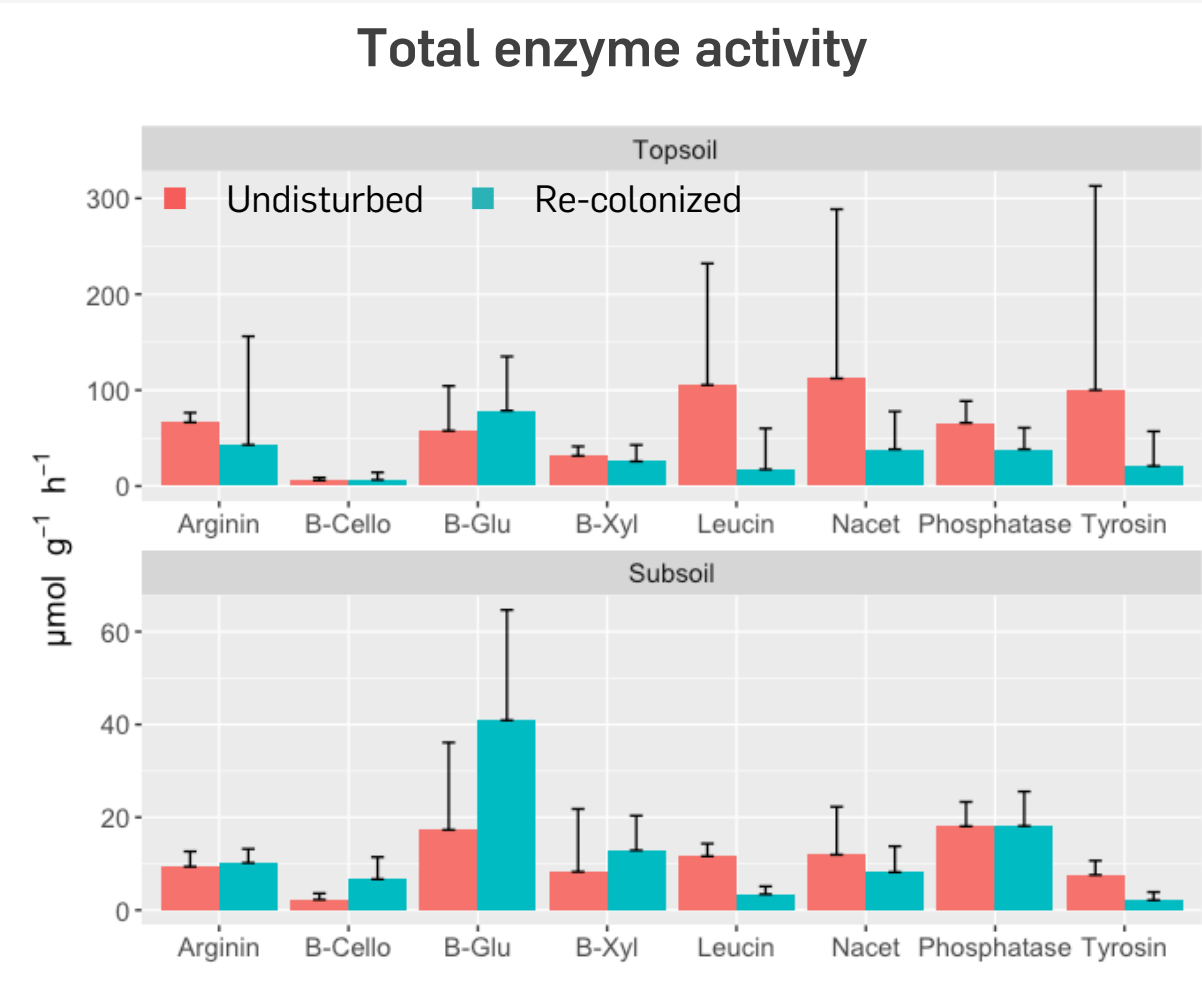


Fig 5: Total enzyme activity for topsoil P1 (top) and subsoil P2 (bottom)

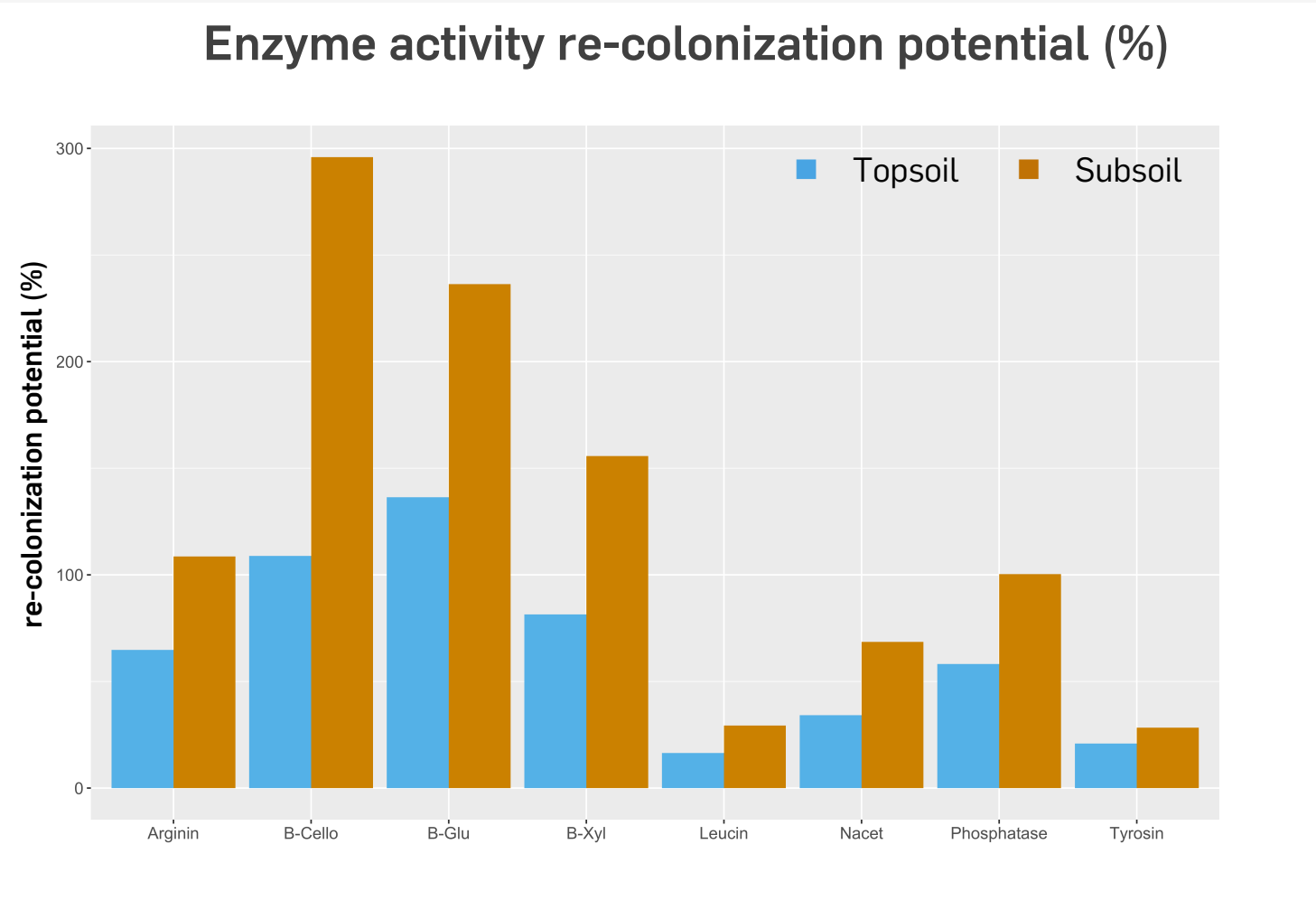


Fig 6: Enzyme activity of the re-colonized samples in relation to the undisturbed samples in %.

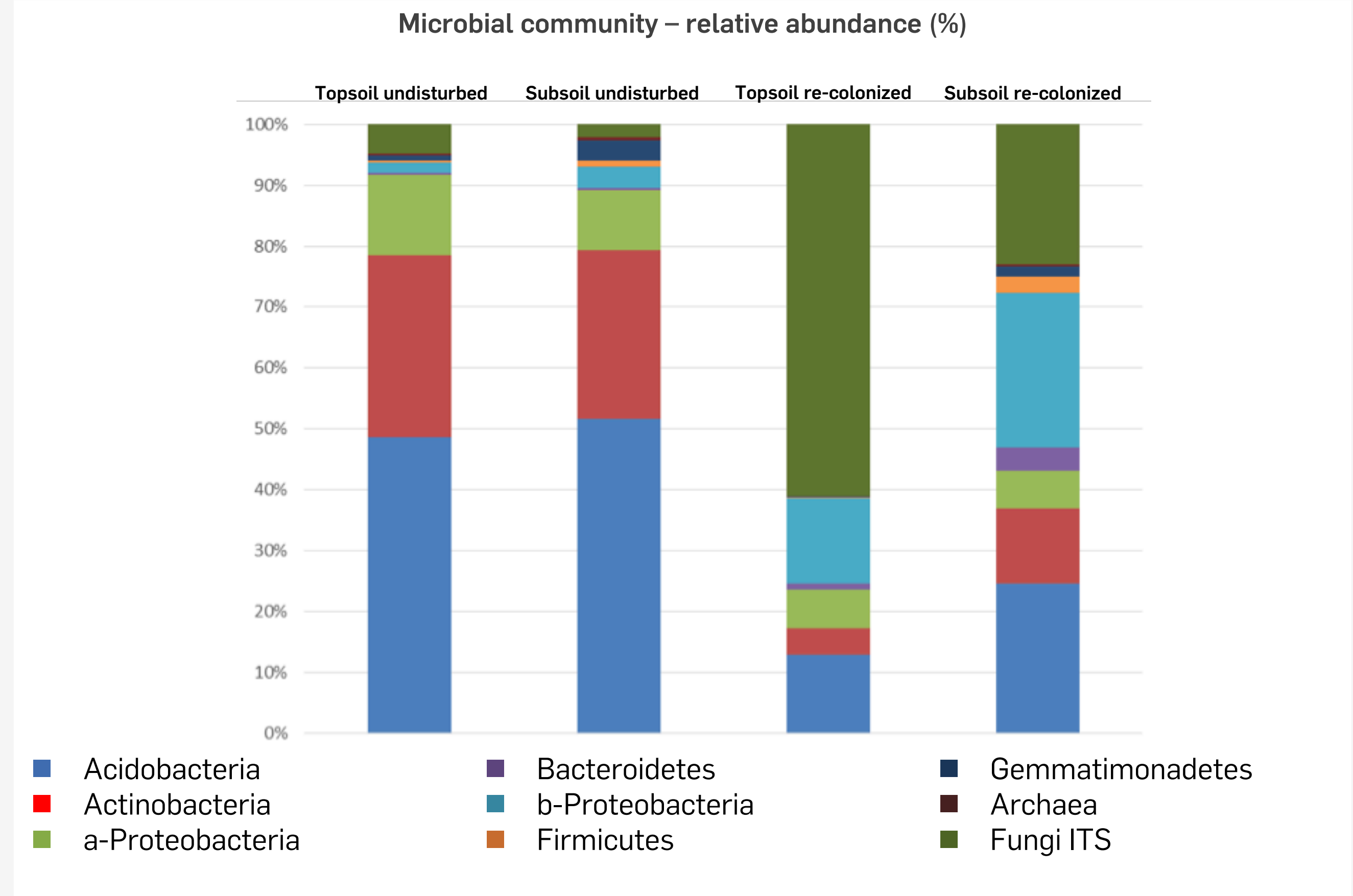


Fig 7: Relative abundance (%) of the microbial community in topsoil and subsoil.

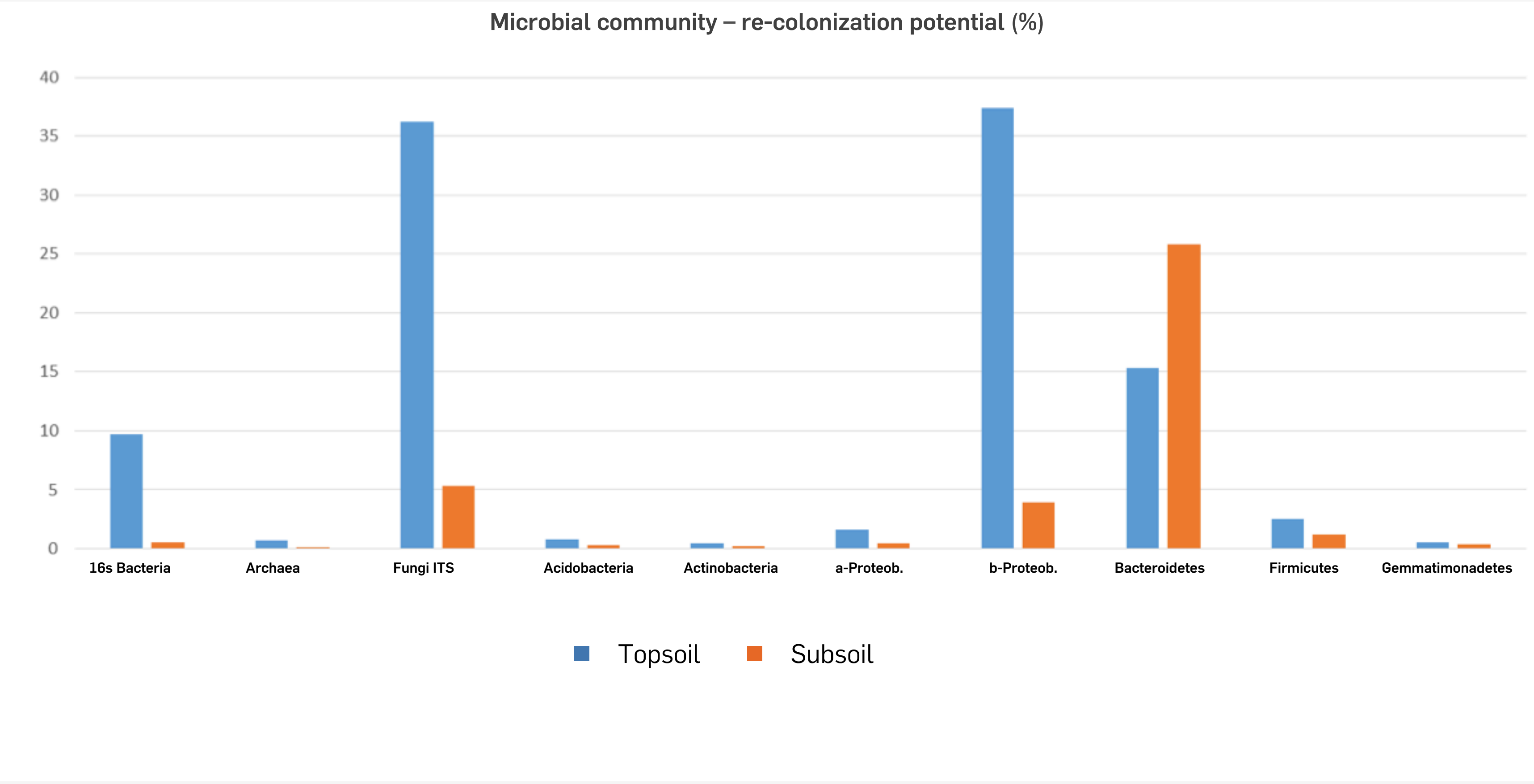


Fig 8: Re-colonization potential of the microbial community (%).

## 3. SUMMARY AND OUTLOOK

### Microbial community

- Significant differences in the microbial community composition between top- and subsoil
- Higher re-colonization in all topsoil samples → No C- and O<sub>2</sub> limitation
- High relative abundance of **fungi** in re-colonized soil samples in both depth – pioneer organism
- Higher relative abundance of **fungi** in the topsoil compared to the subsoil → O<sub>2</sub> and nutrient limitation in deeper soil layers
- 16s bacteria show a higher abundance in the subsoil samples, where fungi is no competitor
- **Acidobacteria**, **Actinobacteria** and **a-Proteobacteria** are the dominant group in the natural top- and subsoilsamples (Fierer et al. 2005 & 2007)
- Re-colonized soil samples show a higher abundance of **b-Proteobakterien** → r-strategist
- Higher relative and absolute abundance of Gemmatimonadetes in subsoil samples (natural and re-col.) → Gemmatimonadetes are oligotrophic

### Microbial activity

- Topsoil: Results after 6 months of field exposure show that microbial activity has been re-established in all of the wells, but is still below the mean activity in the undisturbed soil above the sterilized samples
- Subsoil: the re-established microbial activity was much lower and even below detection limit in some of the wells
- In both depths, the SIR assays show a very patchy distribution of wells with higher microbial activities indicating that the influx of organisms is limited to small areas from the soil above the exposed containers
- The enzyme activity assay shows a high activity of c-cycling ( $\beta$ -cellobiosidase,  $\beta$ -glucosidase and  $\beta$ -xylosidase) enzymes in the re-colonized samples indicating a demand of pioneer organisms which is more pronounced in the subsoil samples
- The qPCR results reveal a significant different re-colonization potential between the two depths with fungi as a pioneer organism in both depth and oligotrophic Gemmatimonadetes in the subsoil.

### Outlook:

- Samples from the second (November 2019) and third (April 2020) field campaign will be analyzed in May/June '20
- Expected results: Increasing microbial activity and higher abundance of micro-organisms in the re-colonized samples

REFERENCES:  
Fierer et al. (2005): Assessment of soil microbial community structure by use of taxon-specific quantitative PCR assays. Applied and Environmental Microbiology 71, 4117-4120.  
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