Soil Structure, Texture, and Moisture Affect Soil Bacterial Diversity: A Biophysical Perspective to Understand Soil Inner Space

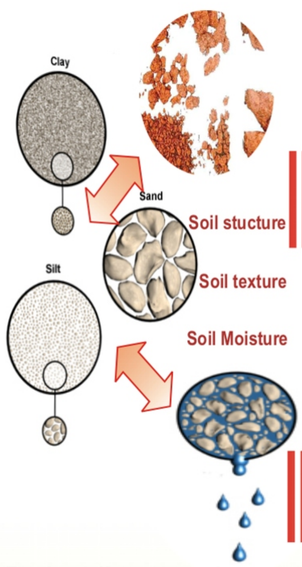
H.M.L.I. Herath[1*], Per Moldrup[2], Mogens Nicolaisen[3], Trine Norgaard[1], Muhammad Naveed[1], and Lis W. de Jonge[1]

[1]Department of Agroecology, Faculty of Science and Technology, Aarhus University, Blichers Allé 20, Postboks 50, 8830 Tjele, Denmark.
[2]Department of Civil Engineering, Aalborg University, Sohngaardsholmsvej 57, DK-9000 Aalborg, Denmark.
[3]Department of Agroecology, Faculty of Science and Technology, Aarhus University, Forsøgsvej 1, 4200 Slagelse, Denmark.

STAiR

"Life and its diversity in earth define life and its diversity on earth"

Structural and textural complexity of the soil determines the biochemical processes that affect life in the soil inner space



Basic soil structural properties such as bulk density, porosity, air permeability and volumetric water content also influence the soil microenvironment

Bacteria are the governing force pumping 'life' into the soil inner space via various biological processes that are unique to prokaryotes. continued functioning of all major biogeochemical cycles depends on the activity of various bacteria

Oxygen and carbon cycle
Various Photoheterotrophs
Various Chemolithoheterotrophs
Various Chemoorganoheterotroph
Various Photoautotrophs
Various Chemoautotrophs

Phosphorous solubilizing bacteria
Pseudomonas spp.
Bacillus spp.
Burkholderia spp.
Achromobacter spp
Agrobacterium spp.
Micrococcus spp.

Nitrogen Fixation
Rhizobium spp.
Bradyrhizobium spp.
Azotobacter spp.
Acetobactor spp.

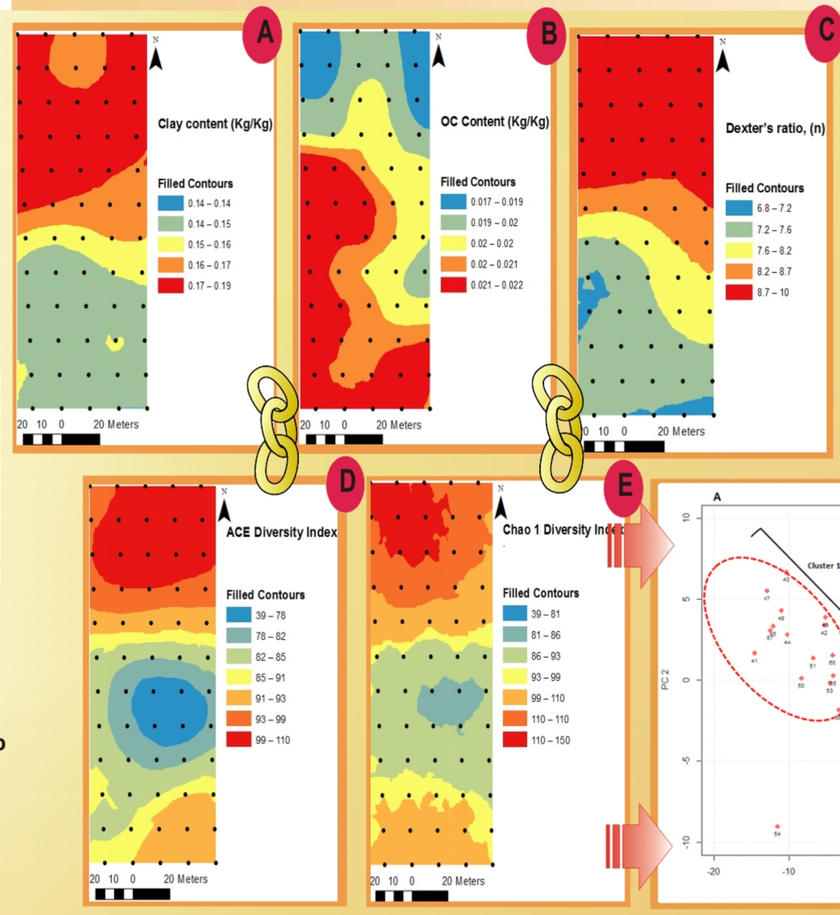
Nitrification
Nitrosomonas spp.
Nitrosospira spp.
Nitrosococcus spp.

Denitrification
Bacillus spp.
Paracoccus spp.
Pseudomonas spp.

Ammonification
Various prokaryotes

Identification of biophysical phenomena that can influence bacterial diversity can open up new ways to define the most complicated biophysical properties of soils

Soil as a biomaterial; understanding soil inner space links



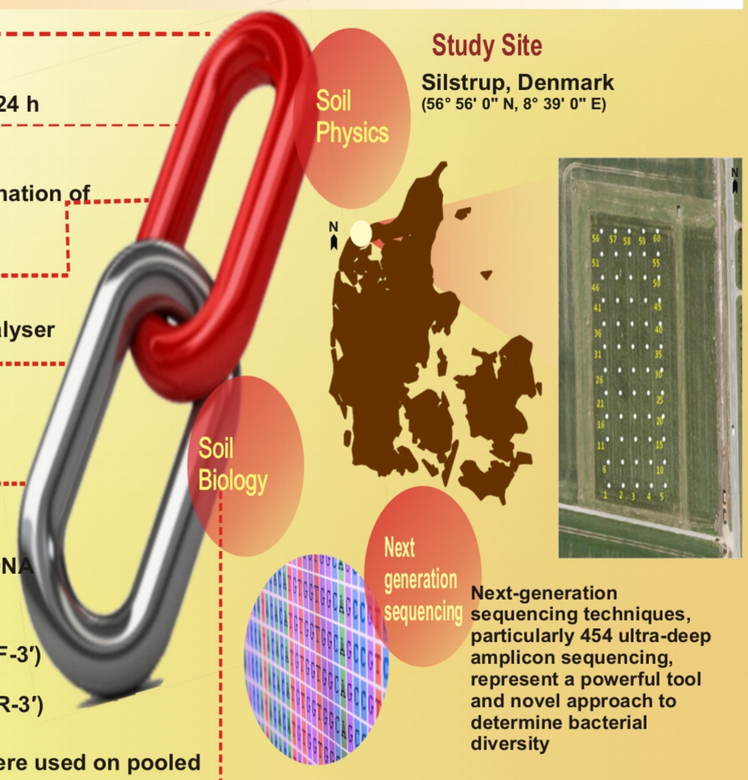
Contour maps of (A) clay content, (B) organic carbon content (OC), (C) Dexter's ratio (D), abundance-based coverage estimation (ACE) diversity index and (E) Chao diversity index across the experimental field at Silstrup

Contour maps of clay and organic carbon (OC) showed heterogeneous distribution across the field, which was the result of naturally occurring; opposing clay and OC gradients

From the Dexter ratio contour map , it could be seen that soil structure and structural properties differed in the northern and southern parts

How bacterial diversity is affected by soil structure, texture and moisture?

Cross-disciplinary approach linking soil physics with soil biology



Next-generation sequencing techniques, particularly 454 ultra-deep amplicon sequencing, represent a powerful tool and novel approach to determine bacterial diversity

1 **Porosity and volumetric water content**
Determined by oven-drying at 105 °C for 24 h

2 **Soil texture**
Determined 2-mm sieved soil by a combination of wet sieving and hydrometer methods

3 **Total organic carbon**
Determined with a CNS-1000 (LECO) analyser

4 **DNA Extraction**
PowerSoil DNA Isolation Kit
(Mo Bio Laboratories, Inc.)

5 **PCR Amplification, Purification, and Pyrosequencing**
Amplify the V4 domain of bacterial 16S rDNA for 454 pyrosequencing

forward primer
(5′-GTGCCAGCMGCCGCGGTAA-MID-515F-3′)
reverse primer
(5′- GGACTACVSGGGTATCTAAT-MID-806R-3′)

Sample identification,
60 MID primer tags with 10 nucleotides were used on pooled samples

(GeneAmp PCR System 9700 thermal cycler)
(PE Applied Biosystems)

6 **Sequence analysis and community structure visualization**

The bacterial V4 region sequences were processed using the visualization and analysis of microbial population structure (VAMPS) (http://vamps.mbl.edu)

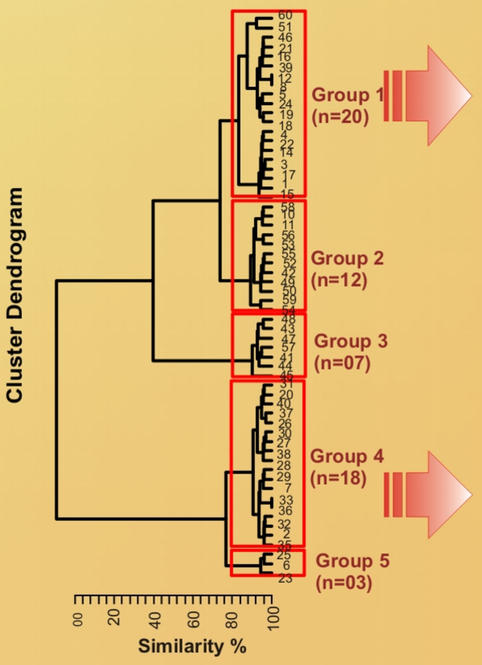
7 **Statistical Analysis**
Principal component analysis (PCA), considering availability and abundance of bacterial species

Cluster dendrogram,
Ward's method was applied in hierarchical clustering

*Correspondence: Lasantha.Herath@agrsci.dk

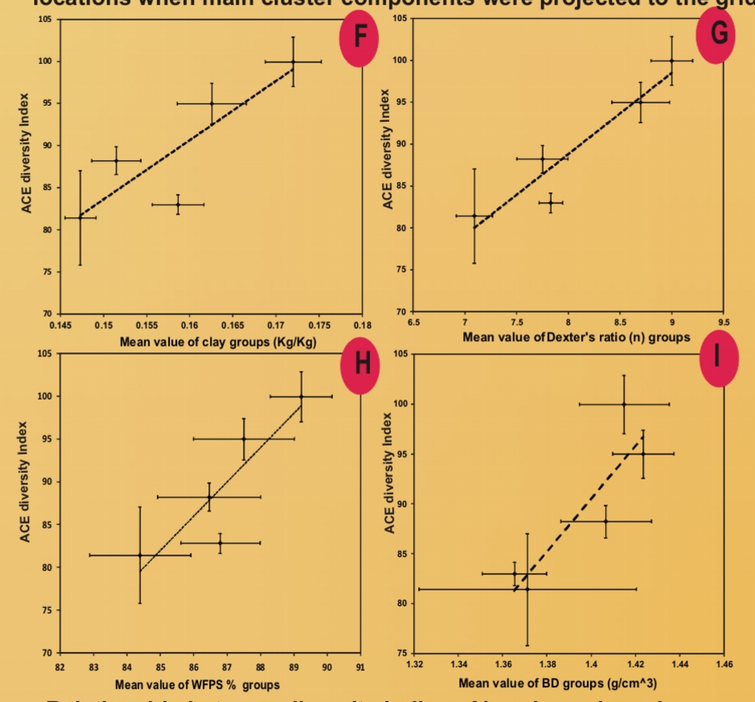
Contour maps of ACE and Chao 1 bacterial richness indicators depict high bacterial diversity in the northern part of the field.

Similar distribution pattern can be also observed between diversity indices and Dexter ratio contour map



Bacterial species and their abundance observed at each sampling points clustered in cluster tree. Groups were identified considering 75% similarity level

(A) Principal component analysis (PCA) plot of bacterial species and their abundance observed at each sampling points. Cluster 1 (red) represent sampling points at northern part of the field. Cluster 2 (blue) represent sampling points at southern part of the field. Each number represents a sampling point and coloured circles (red and blue) represent the two major clusters. (B) Sampling grid locations when main cluster components were projected to the grid



Relationship between diversity indices Abundance-based Coverage Estimation (ACE) and (F) clay content, (G) Dexter's ratio, (H) water-field pore space (WFPS), and (I) bulk density (BD) groups

The physical complexity of the soil matrix created by soil texture, structure, and moisture controls the microenvironment for soil bacteria. Soil bacterial diversity also changes according to soil matrix variations

Acknowledgements
The technical assistance of Stig T. Rasmussen, Bodil B. Christensen, Jørgen M. Nielsen, and Michael Koppelgaard is gratefully acknowledged. This study formed part of the Soil Infrastructure, Interfaces, and Translocation Processes in Inner Space (Soil-it-is) project, which is funded by the Danish Research Council for Technology and Production Sciences