



This project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 817819



SoildiverAgro

Soil biodiversity enhancement in European agroecosystems to promote their stability and resilience by external inputs reduction and crop performance increase

D7.1- OPERATIONAL SOIL BIODIVERSITY TARGETS

Universidade de Vigo



D7.1. OPERATIONAL SOIL BIODIVERSITY TARGETS

Summary

One of the main objectives of Work Package 7 (WP7) of the SoildiverAgro project was to define operational biodiversity targets (including functional groups, density, activity, number of groups and species, and the presence of beneficial species or pathogens) for European agricultural soils, based on regional edaphic and climatic characteristics. However, due to the inherent complexity of soil ecosystems associated organisms, it was not possible to fully achieve this highly ambitious goal due to the impossibility of establishment of biodiversity thresholds.

Nonetheless, a key achievement of this work has been the identification of relationships between specific groups of soil organisms and key ecosystem services—such as carbon sequestration and storage, nutrient cycling and fertility, water retention, water infiltration and erosion control, and contamination reduction—using network-based modelling approaches. While the definition of operational biodiversity targets could not yet be realized, the findings presented here offer valuable insights into the links between soil microorganisms and ecosystem services. They also illustrate how these relationships are shaped by pedoclimatic variables and highlight those that are consistent across different European regions.

These results lay a solid foundation that, when combined with outcomes from other complementary research projects, could significantly contribute to the future establishment of operational biodiversity targets for European agricultural soils.

Deliverable Number		Work Package	
D7.1.		WP7_ Development of strategies and tools for sustainable crop management	
Lead Beneficiary		Deliverable Author(s)	
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Versions (updates)		Date	
V1		10.05.2025	
V2		19.05.2025	
Deliverable Quality Check		Date	
David Fernández Calviño [UVIGO]		26.05.2025	
Planned Delivery Date		Final Delivery Date	
31.05.2025		25.05.2025	
Type of deliverable	R	Document, report (excluding periodic and final reports)	X



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	DEC	Websites, patents filing, press & media actions, videos	
	E	Ethycs	
Dissemination Level	PU	Public	X
	CO	Confidential, only for members of the consortium	



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1 INTRODUCTION

An adequate level of soil biodiversity is essential for maintaining the long-term sustainability of agricultural systems. This is key to ensuring a stable food supply for a continuously growing global population, while simultaneously mitigating environmental pollution. It is well established that soil biodiversity plays a crucial role in multiple ecosystem functions and services, including nutrient cycling, organic matter decomposition, biological control of pests and diseases, soil fertility and productivity, improvement of soil structure, regulation of the hydrological cycle, carbon sequestration and climate regulation, and contaminant remediation, among others (Rehman et al., 2022; Köninger et al., 2022; Robinson et al., 2024).

Therefore, achieving optimal levels of biodiversity in agricultural soils is expected to significantly enhance these ecosystem functions and services. This, in turn, would increase soil fertility and, consequently, crop yields and associated economic benefits. Furthermore, it would reduce the need for external inputs such as pesticides, synthetic fertilizers, and irrigation water—thus lowering associated costs and minimizing ecological and human health impacts. Additionally, it would contribute to greater carbon sequestration in agricultural soils, thereby helping to mitigate the effects of climate change, a matter of global concern.

In this context, the original objective of Deliverable 7.1 was to define operational biodiversity targets (e.g., functional groups, density, activity, number of groups and species, presence of beneficial species or pathogens) for European agricultural soils as a function of edaphic and climatic regional characteristics. This objective was to be achieved based on the results obtained throughout the SoildiverAgro Project, which assessed the status of soil biodiversity across agricultural soils in nine distinct pedoclimatic regions in Europe (Boreal, Nemoral, Atlantic North, Continental, Atlantic Central, Pannonian, Lusitanian, Mediterranean North, and Mediterranean South). The project also evaluated the effects of different sustainable agricultural practices—such as crop diversification, reduced tillage, cover cropping, organic amendments, the use of biofertilizers, and integrated pest management—on soil biodiversity.

However, the inherent complexity of soil ecosystems makes it extremely challenging to define the expected composition of soil biota for a given soil type or to draw meaningful comparisons across experimental results. Consequently, the ambitious objective of defining operational biodiversity targets could not be fully achieved. Nevertheless, a major and novel outcome of this research has been the establishment of relationships between soil organisms groups and soil ecosystem services and functions by using network-based models. These findings offer a valuable foundation for the future development of operational biodiversity targets, particularly when integrated with results from other complementary research initiatives.

Importantly, the relationships identified in this research between soil organisms groups and soil ecosystem functions and services are groundbreaking. Despite ongoing efforts to define biological indicators of soil health, there is currently a lack of ecological indicators that directly link soil biodiversity variables to ecosystem functions and services. Most existing studies focus separately on soil communities (mainly fungi and bacteria) and ecosystem services, or address

only a limited subset of these services, without establishing a standardized reference set of biological indicators. Consequently, very few study sites currently contain integrated information on biodiversity and its functional implications for soil performance.

Moreover, most previous research has primarily addressed CO₂ emissions and soil pH, leaving a significant knowledge gap regarding the relationships between soil biodiversity and factors such as carbon sequestration and storage, nutrient cycling and fertility, water retention, water infiltration and erosion control, and contamination reduction—all of which were examined in the present study.

Therefore, the findings presented in this deliverable offer valuable insights into the relationship between soil microorganisms and ecosystem services, illustrating how these relationships are conditioned by pedoclimatic variables and identifying those that are consistent across the European context.

These results provide a solid basis for developing biodiversity-based indicators to assess soil health and guide sustainable agricultural practices. They can support future policy initiatives aimed at improving soil resilience and contribute to practical decision-making for farmers, helping to enhance productivity, reduce input dependency, and mitigate climate change effects.

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2 MATERIAL AND METHODS

2.1. Construction of co-abundance networks

The relationship between OTUs was described using co-abundance networks, as already described in Deliverable D3.3. To avoid rare OTUs having a too large influence on the statistical results, the data was pre-filtered by abundance. Namely, only OTUs with a relative abundance of at least 0.03% for prokaryotes, 0.2% for eukaryotes (fungal species) and 0.5% per nematodes was considered in the analysis. Then, only OTUs present on every sample were included in the analysis. The relative abundance threshold were selected to keep approximately 80% of the average relative abundance. These values are relatively high compared to other studies from the scientific literature because further steps down the pipeline (clustering, sPLS, bipartite network) introduce further filters, so it is desirable not to filter out rare OTUs that may have a key ecosystem function.

Co-abundance networks were built using the SPIEC-EASI pipeline (Kurtz et al., 2015) through the implementation included in the SpiecEasi package (version 1.1.3). This algorithm was selected, as it is based on conditional independence, so it is generally more robust against spurious associations than traditional methods based on correlations.

The models were fitted using the *multi.spiec.easi* function, which enables combining multiple compositional datasets. Hence, the non-normalized OUT tables for bacteria, fungi and nematodes were passed as inputs. The model was fitted by covariance selection with LASSO regularization (Kurtz et al., 2015). Model selection was done by pulsar with 20 random subsamples.

The results of the SPIEC-EASI pipeline were represented as a graph, where the vertices are the OTUs measured. The edges describe the relationships between OTUs, with positive values indicating positive associations (e.g., synergism) and negative ones the opposite (e.g., competition). Edges were calculated from the optimal covariance matrix of the fitted model and converted to correlations to facilitate interpretation.

2.2 Clustering of highly related OTUs

Clusters of highly related OTUs were identified using the spin-glass community detection algorithm (Reichardt & Bornholdt, 2006) from the co-abundance networks estimated using SPIEC-EASI with default arguments. This results in the detection of highly connected groups of OTUs (including bacteria, fungi and nematodes).



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For further analysis, the results of the clustering were combined with the raw copies measured for each OTUs. Hence, the total copies of bacterial, fungal and nematode OTUs for each cluster on each sample were calculated.

2.3 Relationships between biodiversity, soil characteristics, climate conditions and agricultural management

Biodiversity was related to meta-information on soil characteristics by sparse Partial Least Squares (sPLS) (Lê Cao et al., 2008) using the functions included in the *mixOmics* package (version 6.23.4). PLS are a type of dimensionality reduction technique, where the data from two separate matrix are projected (in a latent space) in a manner that optimizes their covariance. In this sense, it can be seen as an extension of Principal Component Analysis (PCA) to separate datasets. The sPLS algorithm combines PLS with LASSO regularization to reduce the number of variables included in the model.

The X matrix (matrix of predictors) for the sPLS comprises the soil characteristics, climate conditions and agricultural management variables. Then, the Y matrix (matrix of outcomes) includes the sum of gene copies for every OUT belonging to a given cluster (calculated independently for bacteria, fungi and nematodes). The model was defined as a canonical problem (as opposed to regression), as there is no clear cause-effect between matrices X and Y. Hyperparameter tuning was done by M-fold cross-validation with 10 folds, up to 8 components and 2 independent repeats.

The key associations within the sPLS models were identified using relevance association networks (González et al., 2012) using the implementation included in *mixOmics*. Cut-off values were defined independently for each region to identify the most relevant interactions.

2.4 Linking ecosystem services with clusters of OTUs

The association networks calculated by sPLS were filtered per region based on the strength of the association. As these correlations have no direct statistical/biological interpretation (e.g., they are not comparable to a p-value), each pedoclimatic region was analyzed independently, selecting only the strongest associations based on threshold values. For feasibility regions, the threshold values were defined, so the networks did not include more than two clusters.

Before providing a biological interpretation, physicochemical variables were grouped by ecosystem services based on expert knowledge as exposed below:

- **Carbon sequestration and storage:** Organic Matter (OM), Total Organic Carbon (TOC), Labile Carbon (LC), Particulate Organic Carbon (POC), and Mineral associated Organic Carbon (MOC).
- **Nutrient cycling and fertility:** total Nitrogen (Nt), Carbon Exchange Capacity (CEC), Exchangeable Ca (Caex), Exchangeable Mg (Mgex), Exchangeable K (Kex), ammonium (NH_4^+), Nitrate (NO_3^-), Nitrite (NO_2^-), Available P (Pav), Bioavailable Cu (Cuba), Bioavailable Zn (Znba), Bioavailable Fe (Feba), Bioavailable Mn (Mnba), Bioavailable B (Bb), Mineral N, and Sulfate (SO_4^{2-}).
- **Water retention:** Actual field moisture content (FMa).
- **Water infiltration and erosion reduction:** Bulk density 0-10 cm (BD 0-10), Bulk density 10-25 cm (BD 10-25), Aggregates mean weight diameter (AMWD), and aggregates size distribution, including: large macroaggregates ($> 2000 \mu\text{m}$), small macroaggregates ($250 \mu\text{m} - 2000 \mu\text{m}$), microaggregates ($53 \mu\text{m} - 250 \mu\text{m}$) and silt + clay fractions ($< 53 \mu\text{m}$).
- **Contamination reduction:** pesticides.

3 RESULTS

Positive or negative associations between bacterial, fungal and/or nematode clusters were identified for most Ecosystem Services, as summarized in Table 1.

Table 1: Summary of regions where links between OTU clusters and ecosystem services were identified (marked as "X").

Pedoclimatic region	Carbon Sequestration and Storage	Nutrient Cycling and Fertility	Water Retention	Water Infiltration and Erosion Reduction	Contamination Reduction
Atlantic North		X		X	
Atlantic Central		X			
Boreal	X	X			
Continental	X	X			
Lusitanian	X	X		X	
Mediterranean North		X	X		
Mediterranean South					X
Nemoral	X	X			
Pannonian	X	X			

Appendix I describes in detail the OTUs identified with each ecosystem service. The following taxonomic units were consistently linked with each ecosystem service along every pedoclimatic region (provided a link with the ecosystem service was identified):

- Carbon Sequestration and Storage Ecosystem Service

- **Bacteria:**

Families: Acidobacteriaceae (Subgroup 1), Chitinophagaceae, MNG7, Nitrosomonadaceae, Solibacteraceae (Subgroup 3), Xanthobacteraceae

Orders: Acidobacteriales, Burkholderiales, Chthoniobacterales, Myxococcales, Nitrosomonadales, Rhizobiales, Rhodospirillales, Solibacterales, Sphingobacteriales, Xanthomonadales

- **Fungi:**

Genus *Chaetomium* (family Chaetomiaceae)

Genus *Mortierella* (family Mortierellaceae)

• Nutrient Cycling and Fertility Ecosystem Service

- **Bacteria:**

Families: Chitinophagaceae, Oxalobacteraceae, Phyllobacteriaceae, Pseudonocardiaceae, Solirubrobacteraceae

Orders: Burkholderiales, Gaiellales, Pseudonocardiales, Solirubrobacterales, Sphingobacteriales, Sphingomonadales, Rhizobiales

- **Fungi:**

Genus *Mortierella* (family Mortierellaceae)

- **Nematodes:**

Genus *Acrobeloides* (family Cephalobidae)

• Water Infiltration and Erosion Reduction Ecosystem Service

- **Nematodes:**

Order Rhabditida

4 APPENDIX I: DETAILED DESCRIPTION OF OTUS LINKED TO ECOSYSTEM SERVICES

Appendix I.1 Ecosystem Service: Carbon Sequestration and Storage

- **Continental pedoclimatic area**

- **Cluster 4 bacteria**

Negatively associated with Organic Matter (OM). The number in parenthesis indicates the number of OTUs (Operational Taxonomic Units) assigned to the taxonomic level.

Family: Gemmatimonadaceae (16), Nocardioideaceae (10), Hyphomicrobiaceae (8), Micromonosporaceae (7), Sphingomonadaceae (6), **Nitrosomonadaceae** (6), Comamonadaceae (5), **Xanthobacteraceae** (4), **Solibacteraceae_(Subgroup_3)** (4), Bradyrhizobiaceae (3), Xanthomonadaceae (3), **Chitinophagaceae** (3), DA-101_soil_group (3), Blastocatellaceae_(Subgroup_4) (3), Erythrobacteraceae (3), Methylobacteriaceae (3), Roseiflexaceae (3), Micrococcaceae (2), Bacillaceae (2), Mycobacteriaceae (2), Pseudonocardiaceae (2), Streptomycetaceae (2), Elev-16S-1332 (2), Intrasporangiaceae (2), Pseudomonadaceae (2), Nannocystaceae (2), Planococcaceae (1), Gaiellaceae (1), Phyllobacteriaceae (1), JG34-KF-161 (1), Rhizobiaceae (1), **MNG7** (1), Myxococcaceae (1), Planctomycetaceae (1), Caulobacteraceae (1), Microbacteriaceae (1), Solirubrobacteraceae (1), Longimicrobiaceae (1), Nocardiaceae (1), FFCH11085 (1), Acetobacteraceae (1), Burkholderiaceae (1), Acidimicrobiaceae (1), Oxalobacteraceae (1), Patulibacteraceae (1), KD3-93 (1), JG34-KF-361 (1), Geobacteraceae (1), env.OPS_17 (1), **Acidobacteriaceae (subgroup 1)**, Desulfurellaceae (1), Verrucomicrobiaceae (1), Haliangiaceae (1), Rhodospirillaceae (1)

Order: Rhizobiales (24), Gemmatimonadales (16), Sphingomonadales (12), Propionibacteriales (10), SC-I-84 (7), **Burkholderiales** (7), Micromonosporales (7), Gaiellales (6), Solirubrobacterales (6), **Nitrosomonadales** (6), Micrococcales (5), **Xanthomonadales** (5), **Sphingobacteriales** (5), **Myxococcales** (4), **Solibacterales** (4), Bacillales (3), Corynebacteriales (3), **Chthoniobacterales** (3), Chloroflexales (3), TRA3-20 (2), Pseudonocardiales (2), Streptomycetales (2), **Rhodospirillales** (2), Pseudomonadales (2), Planctomycetales (1), Caulobacterales (1), Longimicrobiales (1), Acidimicrobiales (1),

Desulfuromonadales (1), **Acidobacteriales** (1), Desulfurellales (1),
Verrucomicrobiales (1)

- **Boreal pedoclimatic area**

- **Cluster 5 fungi.**

Positively associated with Organic Matter (OM), Total Organic Carbon (TOC) and Labile Carbon (LC).

Family: **Mortierellaceae** (2), **Chaetomiaceae** (1), Nectriaceae (1)

Genus: **Mortierella** (2), **Chaetomium** (1), **Chrysosporium** (1)

- **Cluster 24 nematodes**

Positively associated with Organic Matter (OM), Total Organic Carbon (TOC) and Labile Carbon (LC).

Order: Dorylaimida (1)

- **Lusitanian pedoclimatic area**

- **Cluster 8 bacteria**

Positively associated with Total Organic Carbon (TOC), Labile Carbon (LC) and Mineral associated Organic Carbon (MOC).

Family: **Acidobacteriaceae (subgroup 1)** (4), **Chitinophagaceae** (2), HSB_OF53-F07 (2), DA111 (2), **MNG7** (1), **Xanthobacteraceae** (1), Alcaligenaceae (1), **Nitrosomonadaceae** (1), Cytophagaceae (1), Xiphinematobacteraceae (1), Polyangiaceae (1), **Solibacteraceae (Subgroup_3)** (1)

Order: **Acidobacteriales** (4), **Xanthomonadales** (4), **Rhizobiales** (2), **Sphingobacteriales** (2), Ktedonobacterales (2), **Rhodospirillales** (2), **Burkholderiales** (1), **Nitrosomonadales** (1), Cytophagales (1), **Chthoniobacterales** (1), **Myxococcales** (1), **Solibacterales** (1)

- **Cluster 8 fungi**

Positively associated with Total Organic Carbon (TOC).

Family: Bulleribasidiaceae (1), **Chaetomiaceae** (1), **Mortierellaceae** (1), Pseudeurotiaceae (1), Chaetosphaeriaceae (1)

Genus: *Vishniacozyma* (1), ***Chaetomium*** (1), ***Mortierella*** (1), *Pseudogymnoascus* (1)

o **Nemoral pedoclimatic area**

- **Cluster 3 fungi**

Negatively associated with Organic Matter (OM) and Total Organic Carbon (TOC).

Family: Nectriaceae (5), **Mortierellaceae** (5), Pleosporaceae (2), **Chaetomiaceae** (2), Cladosporiaceae (1), Bionectriaceae (1), Plectosphaerellaceae (1), Pseudeurotiaceae (1), Mrakiaceae (1), Lasiosphaeriaceae (1), Piskurozymaceae (1), Aspergillaceae (1), Clavicipitaceae (1), Herpotrichiellaceae (1)

Order: Hypocreales (10), Mortierellales (5), Sordariales (4), Pleosporales (2), Capnodiales (1), Helotiales (1), Glomerellales (1), Thelebolales (1), Cystofilobasidiales (1), Filobasidiales (1), Eurotiales (1), Chaetothyriales (1)

Genus: ***Mortierella*** (5), *Alternaria* (2), *Cladosporium* (1), *Dactylonectria* (1), *Clonostachys* (1), *Nectria* (1), *Plectosphaerella* (1), *Pseudeurotium* (1), ***Chaetomium*** (1), *Mrakia* (1), *Podospora* (1), *Trichocladium* (1), *Sarocladium* (1), *Solicoccozyma* (1), *Penicillium* (1), *Metarhizium* (1), *Phialophora* (1)

- **Pannonian pedoclimatic area**

- **Cluster 24 nematodes**

- Positively associated with Particulate Organic Carbon (POC).

Family: Aphelenchoididae (1)

Genus: *Aphelenchoides* (1)

Feeding type: fungivores

Appendix I.2 Ecosystem Service: Nutrient Cycling and Fertility

- **Continental pedoclimatic area**

- **Cluster 4 bacteria**

Negatively associated with total Nitrogen (Nt).

Family: Gemmatimonadaceae (16), Nocardioideaceae (10), Hyphomicrobiaceae (8), Micromonosporaceae (7), Sphingomonadaceae (6), Nitrosomonadaceae (6), Comamonadaceae (5), Xanthobacteraceae (4), Solibacteraceae_(Subgroup_3) (4), Bradyrhizobiaceae (3), Xanthomonadaceae (3), **Chitinophagaceae** (3), DA-101_soil_group (3), Blastocatellaceae_(Subgroup_4) (3), Erythrobacteraceae (3), Methylobacteriaceae (3), Roseiflexaceae (3), Micrococcaceae (2), Bacillaceae (2), Mycobacteriaceae (2), **Pseudonocardiaceae** (2), Streptomycetaceae (2), Elev-16S-1332 (2), Intrasporangiaceae (2), Pseudomonadaceae (2), Nannocystaceae (2), Planococcaceae (1), Gaiellaceae (1), **Phyllobacteriaceae** (1), JG34-KF-161 (1), Rhizobiaceae (1), MNG7 (1), Myxococcaceae (1), Planctomycetaceae (1), Caulobacteraceae (1), Microbacteriaceae (1), **Solirubrobacteraceae** (1), Longimicrobiaceae (1), Nocardiaceae (1), FFCH11085 (1), Acetobacteraceae (1), Burkholderiaceae (1), Acidimicrobiaceae (1), **Oxalobacteraceae** (1), Patulibacteraceae (1), KD3-93 (1), JG34-KF-361 (1), Geobacteraceae (1), env.OPS_17 (1), Acidobacteriaceae (subgroup 1), Desulfurellaceae (1), Verrucomicrobiaceae (1), Haliangiaceae (1), Rhodospirillaceae (1)

Order: **Rhizobiales** (24), Gemmatimonadales (16), **Sphingomonadales** (12), Propionibacteriales (10), SC-I-84 (7), **Burkholderiales** (7), Micromonosporales (7), **Gaiellales** (6), **Solirubrobacteriales** (6), Nitrosomonadales (6), Micrococcales (5), Xanthomonadales (5), **Sphingobacteriales** (5), Myxococcales (4), Solibacteriales (4), Bacillales (3), Corynebacteriales (3), Chthoniobacteriales (3), Chloroflexales (3), TRA3-20 (2), **Pseudonocardiales** (2), Streptomycetales (2), Rhodospirillales (2), Pseudomonadales (2), Planctomycetales (1), Caulobacterales (1), Longimicrobiales (1), Acidimicrobiales (1), Desulfuromonadales (1), Acidobacteriales (1), Desulfurellales (1), Verrucomicrobiales (1)

- **Atlantic Central pedoclimatic area**

- Cluster 2 bacteria

Negatively associated with exchangeable Ca (Caex).

Family: Gemmatimonadaceae (16), Acidobacteriaceae_(Subgroup_1) (15), Bacillaceae (14), Xanthobacteraceae (13), Solibacteraceae (Subgroup_3) (12), Planococcaceae (10), Nocardiodaceae (10), **Chitinophagaceae** (9), **Oxalobacteraceae** (8), Elev-16S-1332 (8), Xanthomonadaceae (8), Comamonadaceae (7), DA101_soil_group (6), Hyphomicrobiaceae (6), Micromonosporaceae (6), Intrasporangiaceae (5), Gaiellaceae (5), Streptomycetaceae (5), Nitrosomonadaceae (5), Mycobacteriaceae (4), Micrococcaceae (4), Paenibacillaceae (4), Blastocatellaceae_(Subgroup_4) (4), Bradyrhizobiaceae (3), Sphingomonadaceae (3), Acidothermaceae (3), Clostridiaceae_1 (3), Alicyclobacillaceae (3), Frankiaceae (3), Kineosporiaceae (3), Nakamurellaceae (2), **Pseudonocardiaceae** (2), Nitrospiraceae (2), Acetobacteraceae (2), Streptosporangiaceae (2), TM146 (2), Peptostreptococcaceae (2), **Solirubrobacteraceae** (2), Acidimicrobiaceae (2), Patulibacteraceae (2), Geodermatophilaceae (2), Hyphomonadaceae (2), Enterobacteriaceae (1), Gsoil-1167 (1), 0319-6A21 (1), **Phyllobacteriaceae** (1), Erysipelotrichaceae (1), Myxococcaceae (1), Geobacteraceae (1), Roseiarcaceae (1), JG37-AG-20 (1), Burkholderiaceae (1), Roseiflexaceae (1), Haliangiaceae (1), Sphingobacteriaceae (1), Erythrobacteraceae (1), YNPFFP1 (1), DA111 (1), Ruminococcaceae (1), Caulobacteraceae (1), Nannocystaceae (1), Beijerinckiaceae (1), Nocardiosporeaceae (1), A0839 (1), 0319-6M6 (1), Rhizobiaceae (1), Nocardiaceae (1), Polyangiaceae (1), KF-JG30-B3 (1), Cellvibrionaceae (1), Flavobacteriaceae (1)

Order: Rhizobiales (39), Bacillales (33), Gaiellales (33), Solirubrobacterales (24), Acidimicrobiales (17), Gemmatimonadales (16), Burkholderiales (16), Acidobacteriales (15), Frankiales (13), Solibacterales (12), Xanthomonadales (11), Propionibacteriales (10), Sphingobacteriales (10), SC-I-84 (9), Micrococcales (9), Chthoniobacteriales (6), Clostridiales (6), Micromonosporales (6), Sphingomonadales (5), Corynebacteriales (5), Rhodospirillales (5), Streptomycetales (5), Nitrosomonadales (5), Myxococcales (4), Nitrospirales (3), Streptosporangiales (3), Subgroup_7 (3), Kineosporiales (3), JG30-KF-CM45 (3), Caulobacterales (3), Pseudonocardiales (2), JG30-KF-AS9 (2), Enterobacteriales (1), Erysipelotrichales (1), Desulfuromonadales (1), Chloroflexales (1), Cellvibrionales (1), Flavobacteriales (1)

o Atlantic North pedoclimatic area

- **Cluster 11 fungi**

Positively associated with sulfate (SO₄⁻²).

Family: **Mortierellaceae** (2)

Genus: **Mortierella** (2)

Species hypothesis: *Mortierella gamsii* (1), *Mortierella elongata* (1)

o **Boreal pedoclimatic area**

- **Cluster 4 bacteria**

Positively associated with bioavailable Manganese (Mnba).

Family: **Chitinophagaceae** (5), Hyphomicrobiaceae (4), Nitrosomonadaceae (4), 0319-6A21 (4), Blastocatellaceae_(Subgroup_4) (4), Elev-16S-1332 (3), Xanthobacteraceae (3), Comamonadaceae (3), Nitrospiraceae (3), Acidimicrobiaceae (3), Cytophagaceae (3), Gaiellaceae (3), Planctomycetaceae (3), Acidobacteriaceae_(Subgroup_1) (3), Micrococcaceae (2), **Phyllobacteriaceae** (2), Nakamurellaceae (2), **Oxalobacteraceae** (2), Bradyrhizobiaceae (2), Gemmatimonadaceae (2), Solibacteraceae_(Subgroup_3) (2), FFCH13075 (1), Intrasporangiaceae (1), JG34-KF-161 (1), Nocardiodaceae (1), Rhodospirillaceae (1), Desulfurellaceae (1), KF-JG30-B3 (1), Catenulisporaceae (1), Xanthomonadaceae (1), Chthoniobacteraceae (1), Blii41 (1), Microbacteriaceae (1), Geodermatophilaceae (1), Hyphomonadaceae (1), BCf3-20 (1), **Pseudonocardiaceae** (1), Methylophilaceae (1), Streptomycetaceae (1), A0839 (1), Thermomonosporaceae (1), **Solirubrobacteraceae** (1), JG34-KF-361 (1), Bacillaceae (1)

Order: **Rhizobiales** (20), Acidimicrobiales (9), **Gaiellales** (7), Nitrospirales (7), Xanthomonadales (5), **Solirubrobacteriales** (5), **Burkholderiales** (5), **Sphingobacteriales** (5), Micrococcales (4), Frankiales (4), Nitrosomonadales (4), Cytophagales (3), Planctomycetales (3), Acidobacteriales (3), Gemmatimonadales (2), Solibacterales (2), **Sphingomonadales** (1), Propionibacteriales (1), Rhodospirillales (1),

Desulfurellales (1), Catenulesporales (1), Chthoniobacterales (1), Myxococcales (1), Caulobacterales (1), **Pseudonocardiales** (1), Methylophilales (1), Streptomycetales (1), Streptosporangiales (1), Lineage_Ila (1), Bacillales (1), SC-I-84 (1)

- **Lusitanian pedoclimatic area**

- **Cluster 6 fungi**

Positively associated with total Nitrogen (Nt).

Family: Didymellaceae (1), Cladosporiaceae (1), **Mortierellaceae** (1)

Genus: *Epicoccum* (1), *Cladosporium* (1), ***Mortierella*** (1)

Species hypothesis: *Epicoccum nigrum* (1), *Cladosporium delicatulum* (1), *Mortierella sarnyensis* (1)

- **Cluster 5 nema**

Positively associated with bioavailable Zinc (Znba).

Genus: *Acrobelloides* (1), *Aphelenchoides* (1), *Aphelenchus* (1)

Feeding type: Bacterivores (1), Fungivores (2)

- **Mediterranean North pedoclimatic area**

- **Cluster 13 bacteria**

Positively associated with Mineral Nitrogen (Mineral N) and nitrate (NO₃⁻).

Family: **Pseudonocardiaceae** (3), **Oxalobacteraceae** (1), Cytophagaceae (1), Methylobacteriaceae (1), Sphingomonadaceae (1), **Phyllobacteriaceae** (1), **Solirubrobacteraceae** (1), **Chitinophagaceae** (1), Rubrobacteriaceae (1), Mitochondria (1)

Order: **Pseudonocardiales** (3), **Solirubrobacterales** (3), **Rhizobiales** (2), **Gaiellales** (2), **Burkholderiales** (1), Cytophagales (1), **Sphingomonadales** (1), **Sphingobacteriales** (1), Rubrobacterales (1), Rickettsiales (1)

- **Cluster 7 fungi**

Positively associated with exchangeable Calcium (Caex).

Family: **Nectriaceae** (1), Magnaporthaceae (1), Ophiocordycipitaceae (1), Tricholomataceae (1)

Order: Hypocreales (2), Magnaporthales (1), Onygenales (1), Agaricales (1)

Genus: *Nectria* (1), *Purpureocillium* (1), *Lachnella* (1)

o **Nemoral pedoclimatic area**

- **Cluster 3 fungi**

Negatively associated with total Nitrogen (Nt).

Family: Nectriaceae (5), **Mortierellaceae** (5), Pleosporaceae (2), Chaetomiaceae (2), Cladosporiaceae (1), Bionectriaceae (1), Plectosphaerellaceae (1), Pseudeurotiaceae (1), Mrakiaceae (1), Lasiosphaeriaceae (1), Piskurozymaceae (1), Aspergillaceae (1), Clavicipitaceae (1), Herpotrichiellaceae (1)

Order: Hypocreales (10), Mortierellales (5), Sordariales (4), Pleosporales (2), Capnodiales (1), Helotiales (1), Glomerellales (1), Thelebolales (1), Cystofilobasidiales (1), Filobasidiales (1), Eurotiales (1), Chaetothyriales (1)

Genus: **Mortierella** (5), *Alternaria* (2), *Cladosporium* (1), *Dactylonectria* (1), *Clonostachys* (1), *Nectria* (1), *Plectosphaerella* (1), *Pseudeurotium* (1), *Chaetomium* (1), *Mrakia* (1), *Podospora* (1), *Trichocladium* (1), *Sarocladium* (1), *Solicoccozyma* (1), *Penicillium* (1), *Metarhizium* (1), *Phialophora* (1)

- **Pannonian pedoclimatic area**

- **Cluster 5 fungi**

- Positively associated with bioavailable Manganese (Mnba).

- Family: Nectriaceae (3), **Mortierellaceae** (2), Chaetosphaeriaceae (1), Coniochaetaceae (1), Lasiosphaeriaceae (1), Ophiocordycipitaceae (1), Stachybotryaceae (1), Aspergillaceae (1), Trimorphomycetaceae (1), Bionectriaceae (1), Chaetomiaceae (1), Piskurozymaceae (1)

- Genus: *Fusarium* (3), ***Mortierella*** (2), *Coniochaeta* (1), *Podospora* (1), *Purpureocillium* (1), *Stachybotrys* (1), *Penicillium* (1), *Saitozyma* (1), *Clonostachys* (1), *Dichotomopilus* (1), *Solicoccozyma* (1)

- **Cluster 5 nematodes**

- Positively associated with bioavailable Manganese (Mnba).

- Family: Cephalobidae (3), Plectidae (2), Rhabditidae (1), Tylenchidae (1)

- Order: Rhabditida (6), Plectida (2)

- Genus: *Acrobelloides* (3), *Oscheius* (1), *Plectus* (1), *Tylocephalus* (1)

- Feeding type: Bacterivores (6)

Appendix I.3 Ecosystem Service: Water Retention

- **Mediterranean North pedoclimatic area**

- **Cluster 2 bacteria**

- Positively associated with actual field moisture content (FMa).

- Family: Nocardioideae (18), Gemmatimonadaceae (16), Elev-16S-1332 (15), Solirubrobacteraceae (13), Gaiellaceae (9), Tepidisphaeraceae (9),

Geodermatophilaceae (8), Nitrosomonadaceae (7), Pseudonocardiaceae (7), 0319-6M6 (7), Micrococcaceae (6), Oxalobacteraceae (5), Rubrobacteriaceae (5), Micromonosporaceae (4), Q3-6C1 (3), Parviterribacteraceae (3), Streptomycetaceae (3), Sphingomonadaceae (3), Thermomonosporaceae (3), Planctomycetaceae (3), Cytophagaceae (3), Bradyrhizobiaceae (3), Cellulomonadaceae (3), Methylobacteriaceae (2), Phyllobacteriaceae (2), Chitinophagaceae (2), Blastocatellaceae_(Subgroup_4) (2), Rhodobacteraceae (2), FFCH13075 (1), Intrasporangiaceae (1), JG34-KF-161 (1), DA101_soil_group (1), FFCH11085 (1), Mycobacteriaceae (1), JG34-KF-361 (1), Acetobacteraceae (1), Propionibacteriaceae (1), FCPU744 (1), Longimicrobiaceae (1), Comamonadaceae (1), Roseiflexaceae (1), Promicromonosporaceae (1), Microbacteriaceae (1), Frankiaceae (1), WW2-159 (1), Erythrobacteraceae (1), Rhizobiaceae (1), 288-2 (1), Sandaracinaceae (1), Xanthomonadaceae (1), Hyphomicrobiaceae (1)

Order: Solirubrobacterales (61), Gaiellales (34), Propionibacteriales (20), Gemmatimonadales (16), Rhizobiales (14), Micrococcales (12), Frankiales (11), Sphingomonadales (9), Tepidisphaerales (9), Acidimicrobiales (8), Burkholderiales (7), Nitrosomonadales (7), Pseudonocardiales (7), Rubrobacterales (5), Subgroup_7 (5), Micromonosporales (4), Streptomycetales (3), Streptosporangiales (3), Planctomycetales (3), Cytophagales (3), TRA3-20 (3), Xanthomonadales (3), Rhodospirillales (2), Sphingobacteriales (2), JG30-KF-CM45 (2), Rhodobacterales (2), Myxococcales (2), Chthoniobacterales (1), Corynebacteriales (1), Longimicrobiales (1), Chloroflexales (1)

Appendix I.4 Ecosystem Service: Water Infiltration and Erosion Reduction

- **Atlantic North pedoclimatic area**

- **Cluster 4 bacteria**

Negatively associated with small macroaggregates (250 µm – 2000 µm).

Family: Chitinophagaceae (3), Gemmatimonadaceae (3), Acidobacteriaceae (Subgroup_1) (2), Caulobacteraceae (2), ABS-19 (2), Mitochondria (1), Micromonosporaceae (1), Mycobacteriaceae (1), Xanthobacteraceae (1), Bacillaceae (1), Patulibacteraceae (1), Xanthomonadaceae (1), KF-JG30-B3 (1), Erythrobacteraceae (1), Peptostreptococcaceae (1), Haliangiaceae (1), MSB-4B10 (1), Oxalobacteraceae (1), Rhizobiaceae (1)

Order: Sphingobacteriales (3), Gemmatimonadales (3), Rhizobiales (3), Acidobacteriales (2), Bacillales (2), Solirubrobacterales (2), Caulobacterales (2), Myxococcales (2), Subgroup_10 (2), Rickettsiales (1), Micromonosporales (1), Corynebacteriales (1), Xanthomonadales (1), Sphingomonadales (1), Clostridiales (1), Burkholderiales (1)

- **Lusitanian pedoclimatic area (zone 5)**

- **Cluster 4 nematodes**

Positively associated with large macroaggregates (> 2000 µm)

Order: Rhabditida (2)

- **Cluster 21 nematodes**

Positively associated with large macroaggregates (> 2000 µm)

Family: Dolichodoridae (1), Pratylenchidae (1)

Order: Rhabditida (2)

Genus: *Geocenamus* (1), *Pratylenchus* (1)

Feeding type: Herbivores – migratory endoparasites (1), Herbivores – ectoparasites (1),

Appendix I.5 Ecosystem Service: Contamination Reduction

- **Mediterranean South pedoclimatic area**

- **Cluster 13 bacteria**

Positively associated with total glyphosate compounds.

Family: Gaiellaceae (3), Gemmatimonadaceae (3), Elev-16S-1332 (2), Oxalobacteraceae (2), 0319-6M6 (2), Nocardiodaceae (2), Blastocatellaceae_(Subgroup_4) (1), Bacillaceae (1), DUNssu044 (1), Planctomycetaceae (1), Rhodobacteraceae (1), Sva0725 (1), Opitutaceae (1), Tepidisphaeraceae (1), Cellulomonadaceae (1), Thermomonosporaceae (1), 288-2 (1), Longimicrobiaceae (1), Cytophagaceae (1), Micrococcaceae (1), Cellvibrionaceae (1), Micromonosporaceae (1), Rubrobacteriaceae (1)

Order: Gaiellales (15), Thermoplasmatales (6), Solirubrobacterales (5), Propionibacteriales (3), Gemmatimonadales (3), Micrococcales (2), Burkholderiales (2), Bacillales (1), Rhizobiales (1), Planctomycetales (1), Acidimicrobiales (1), Rhodobacterales (1), Subgroup_10 (1), Opitutaes (1), Tepidisphaerales (1), Streptosporangiales (1), Rhodospirillales (1), Longimicrobiales (1), Cytophagales (1), Cellvibrionales (1), JG30-KF-CM66 (1), Micromonosporales (1), Rubrobacterales (1)