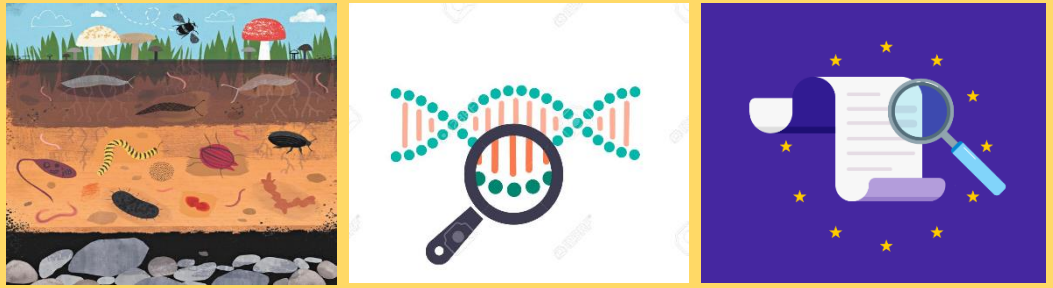

Soil eukaryotes diversity in the EU - environmental drivers in agricultural land, forests and grasslands

EGU 2022
Julia Köninger, Panos Panagos, Maria J. I. Briones, Alberto Orgiazzi

Universidade Vigo

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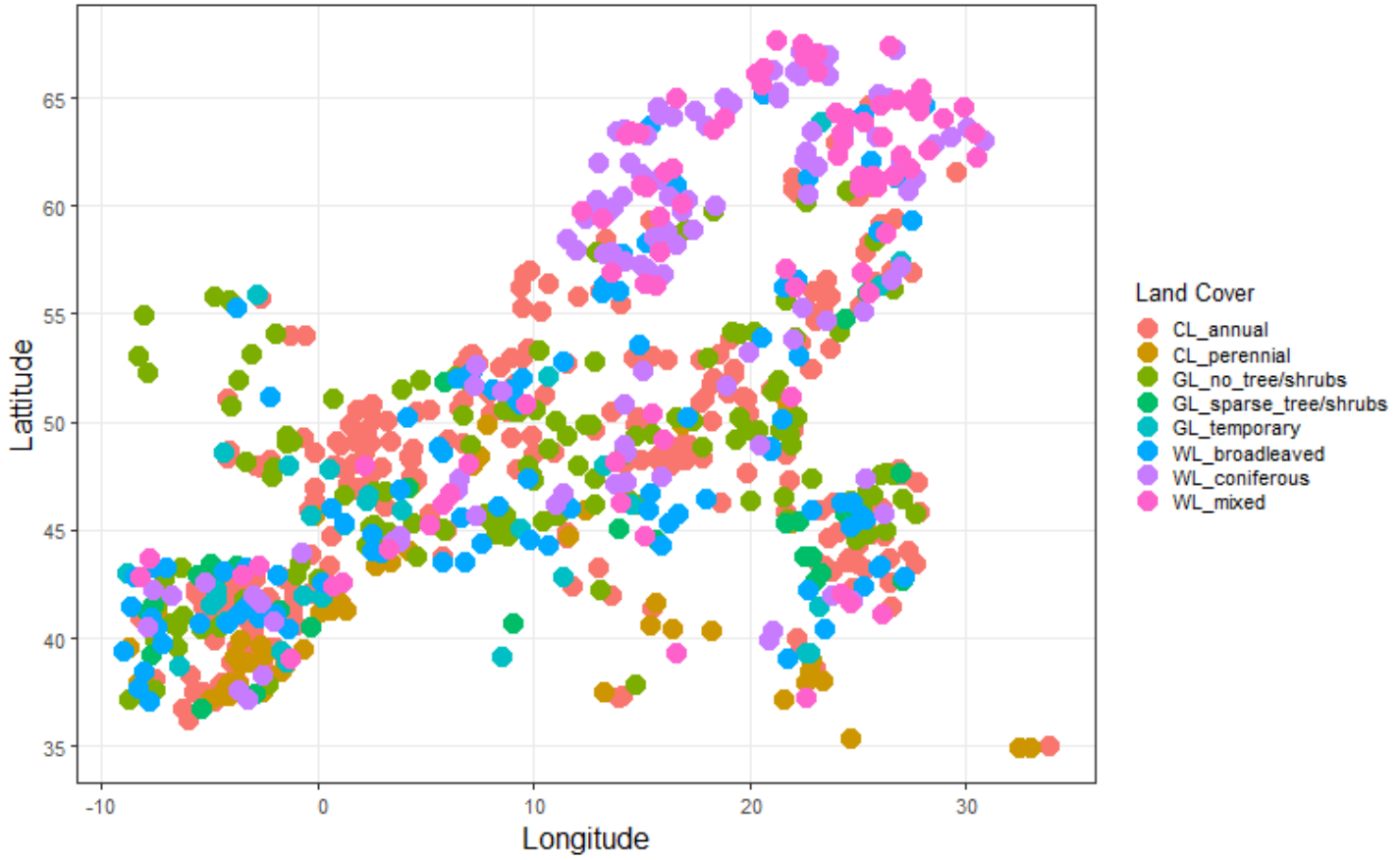
Introduction

Metabarcoding analysis of eukaryotes in the EU

- First EU-wide eukaryote metabarcoding analysis
- 800 sites considered
- Comparable methodology applied (ASVs)
- Initial study for time series of analysis allowing monitoring and indicator analysis
- Important step towards quantitative conservation targets

LUCAS sample sites eukaryotes

A



Environmental drivers for soil eukaryotes diversity in the EU

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METHODS AND MATERIALS

Environmental data

- Abiotic factors:
  - Soil data: LUCAS Soil Survey 2018
  - Climatic data: WorldClim variables
- Biotic factors:
  - Cmic (Smith et al. 2021)

DNA analysis, next-generation sequencing

- Hypervariable 4 region (V4) of the eukaryotic 18S, amplified with the primers Euk575F and Euk895R
- DNA extraction by Qiagen DNeasy PowerSoil HTP 96 Kit Q12955-4
- Illumina MiSeq of 120,000 raw sequences per sample
- Quality check with Qubit™ 1X dsDNA HS Assay Kit

Bioinformatic pipeline & data processing

- DADA2 pipeline binned reads into ASVs (100% OTU similarity)
- Taxonomic assignment with SILVA132 database
- Data was manually curated and separately analysed for fungi, protists and animals
- Data normalization with scaling with ranked subsampling (SRS) (Beule and Karlovsky 2018)

Statistical analysis

- Kruskal-Wallis test to test alpha diversity
- Canonical multivariate analysis: distance-based redundancy analysis (dbRDA) by forward selecting relevant variables
- Variation partitioning
- Spatial analysis using distance-based Moran's eigenvectors maps (dbMEM analysis)

RESULTS – Alpha diversity

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RESULTS – Alpha diversity

Comparison of alpha diversity by Kruskal-Wallis test.

Groups	Alpha diversity			chi-squared	df	p-value
Protists	Richness		Land Cover	70.243	2	5.58E-16
			Land Cover_8 gr	87.037	7	5.02E-16
			pH	36.463	2	1.21E-08
			Elevation	1.4674	3	6.90E-01
			Erosion	5.1426	2	7.64E-02
			Soil depth	10.729	2	4.68E-03
			Sample season	15.202	3	1.65E-03
		Shannon	Land Cover	73.969	2	2.20E-16
			Land Cover_8 gr	85.437	7	1.07E-15
			pH	41.328	2	1.06E-09
			Elevation	1.3234	3	7.24E-01
			Erosion	4.9072	2	8.60E-02
			Soil depth	15.643	2	4.01E-04
			Sample season	12.67	3	5.41E-03
Fungi	Richness		Land Cover	125.66	2	2.20E-16
			Land Cover_8 gr	143.96	7	2.20E-16
			pH	91.149	2	2.20E-16
			Elevation	2.804	3	4.23E-01
			Erosion	12.314	2	2.12E-03
			Soil depth	11.335	2	3.46E-03
			Sample season	9.134	3	2.76E-02
		Shannon	Land Cover	70.59	2	4.69E-16
			Land Cover_8 gr	793804	7	1.51E-14
			pH	58.447	2	2.03E-13
			Elevation	5.6167	3	1.32E-01
			Erosion	2.2196	2	3.30E-01
			Soil depth	7.2457	2	2.67E-02
			Sample season	0.94536	3	8.15E-01
Micro-fauna	Richness		Land Cover	49.646	2	1.66E-11
			Land Cover_8 grc	74.056	7	2.23E-13
			pH	30.374	2	2.54E-07
			Elevation	1.7179	3	6.33E-01
			Erosion	3.0653	2	2.16E-01
			Soil depth	10.314	2	5.76E-03
			Sample season	7.7923	3	5.05E-02
		Shannon	Land Cover	18.712	2	8.65E-05
			Land Cover_8 grc	34.815	7	1.21E-02
			pH	12.355	2	2.08E-03
			Elevation	3.2943	3	3.48E-01
			Erosion	3.5448	2	1.70E-01
			Soil depth	4.5875	2	1.01E-01
			Sample season	2.4047	3	4.93E-01
Meso-fauna	Richness		Land Cover	70.198	2	5.71E-16
			Land Cover_8 grc	81.671	7	6.28E-12
			pH	32.67	2	8.05E-05
			Elevation	0.57898	3	9.01E-01
			Erosion	8.927	2	1.15E-02
			Soil depth	11.009	2	4.07E-03
			Sample season	12.887	3	4.89E-03
		Shannon	Land Cover	43.284	2	3.99E-10
			Land Cover_8 grc	53.341	7	3.18E-06
			pH	35.064	2	2.43E-08
			Elevation	1.7514	3	6.26E-01
			Erosion	5.5936	2	6.10E-02
			Soil depth	9.6623	2	7.98E-03
			Sample season	20.821	3	1.15E-04
Macro-fauna	Richness		Land Cover	61.223	2	5.08E-11
			Land Cover_8 grc	75.423	7	1.18E-10
			pH	42.328	2	6.44E-10
			Elevation	0.26904	3	9.66E-01
			Erosion	9.0082	2	1.11E-02
			Soil depth	8.1705	2	1.68E-02
			Sample season	0.6399	3	8.43E-02
		Shannon	Land Cover	46.861	2	6.67E-11
			Land Cover_8 grc	67.675	7	4.35E-12
			pH	30.892	2	1.96E-07
			Elevation	1.0471	3	7.90E-01
			Erosion	7.3672	2	2.51E-02
			Soil depth	12.885	2	1.59E-03
			Sample season	1.2745	3	7.90E-01

RESULTS – Beta diversity

RESULTS – Beta diversity

- Clear differences between land cover types visible
- pH value, carbonates, C:N ratio, precipitation in warmest quarter & latitude shape beta diversity most
- Aridity important driver for meso and macrofauna

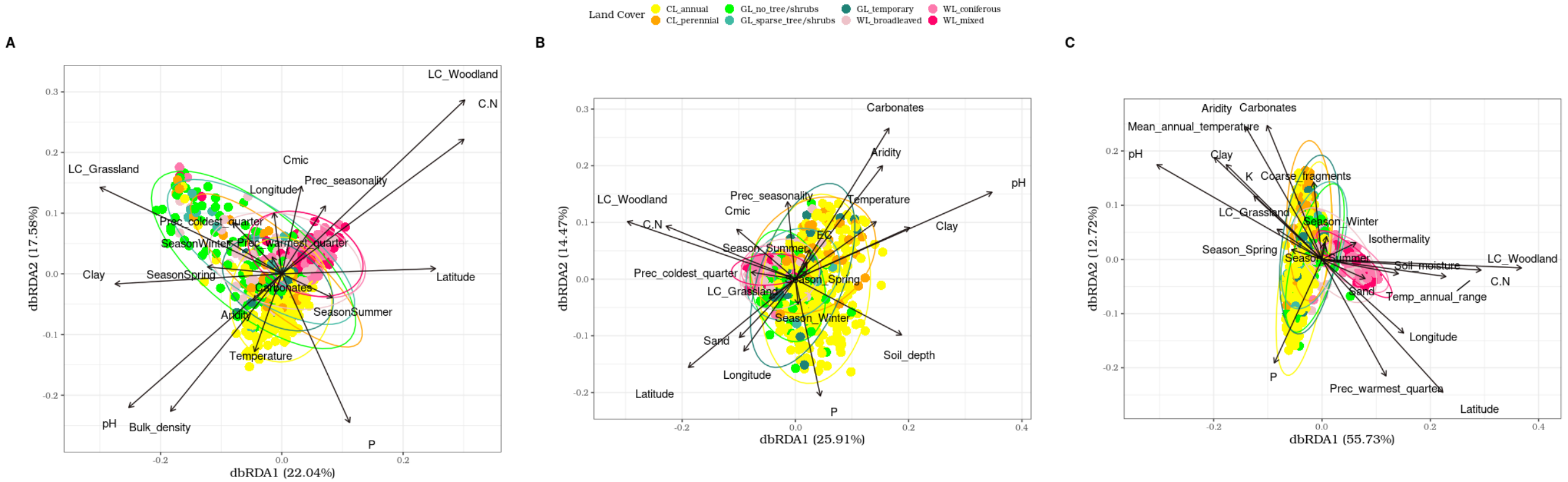

Figure 3. dbRDA plots for **A** microfauna **B** mesofauna **C** macrofauna

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RESULTS – Beta diversity

- Woodlands shaped by C:N ratio
- Croplands: most important drivers are pH, aridity & carbonates
- Grasslands shaped by phosphorus

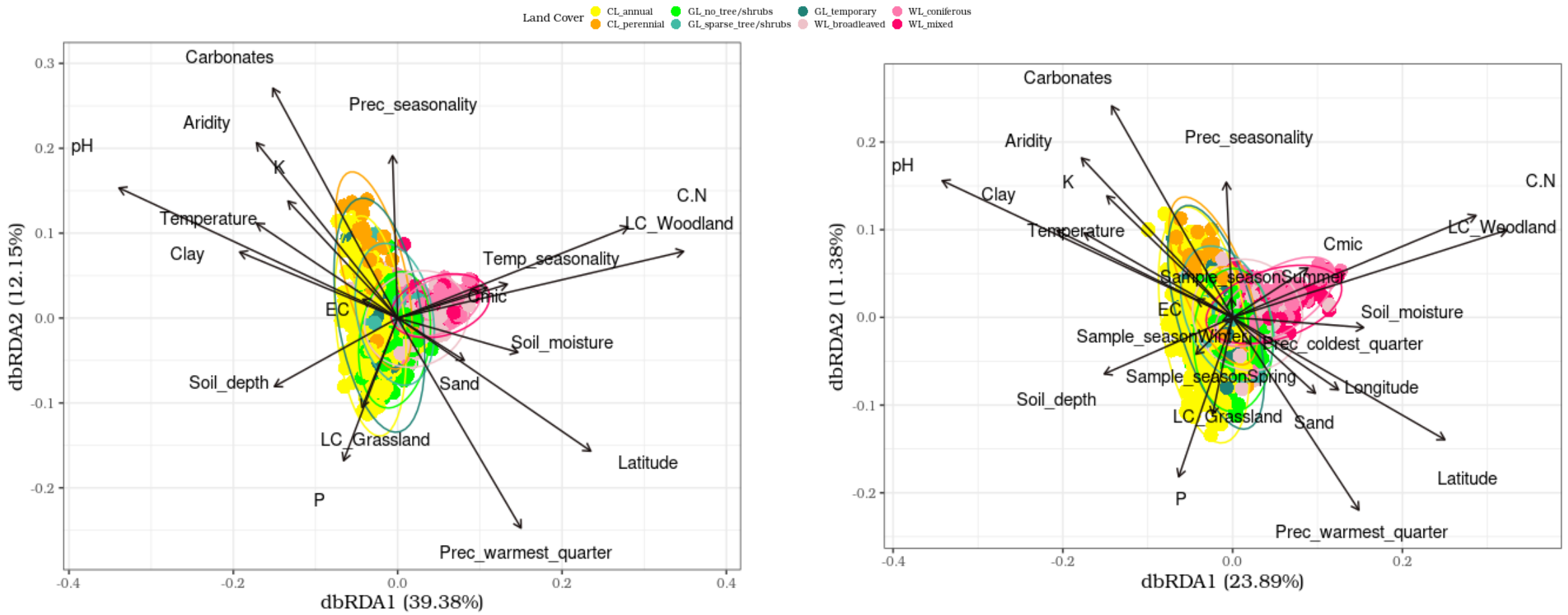
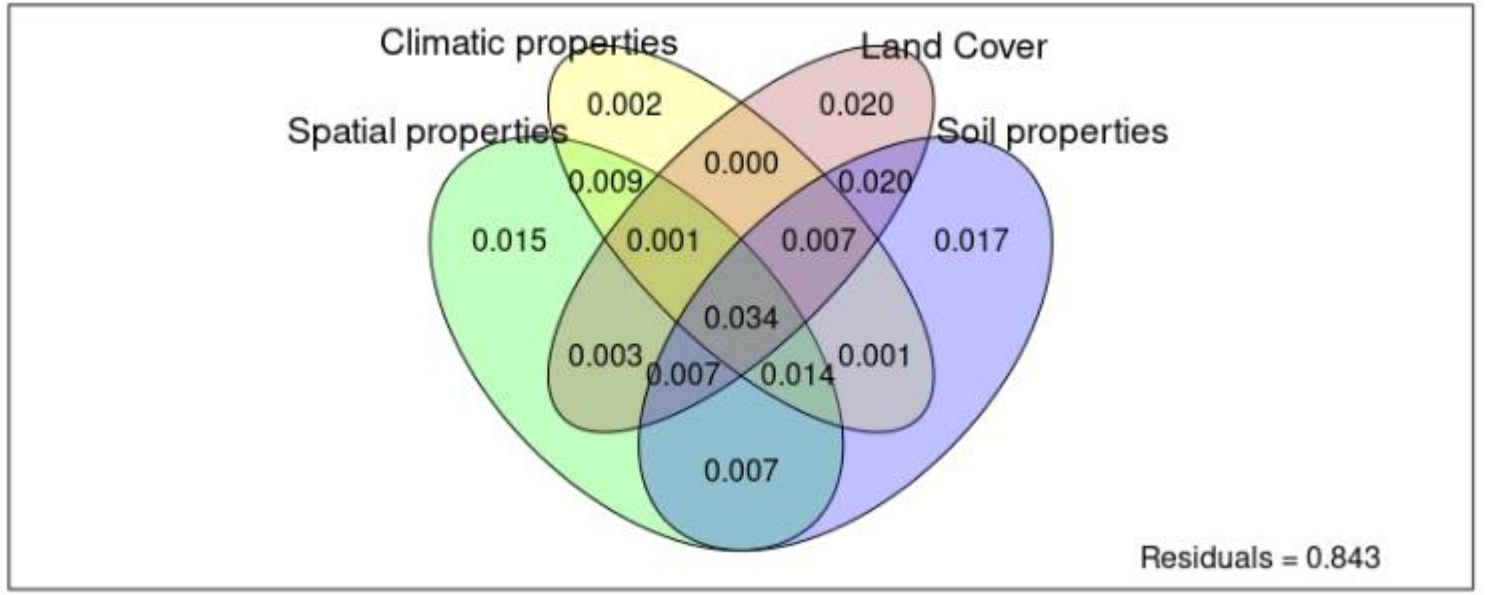


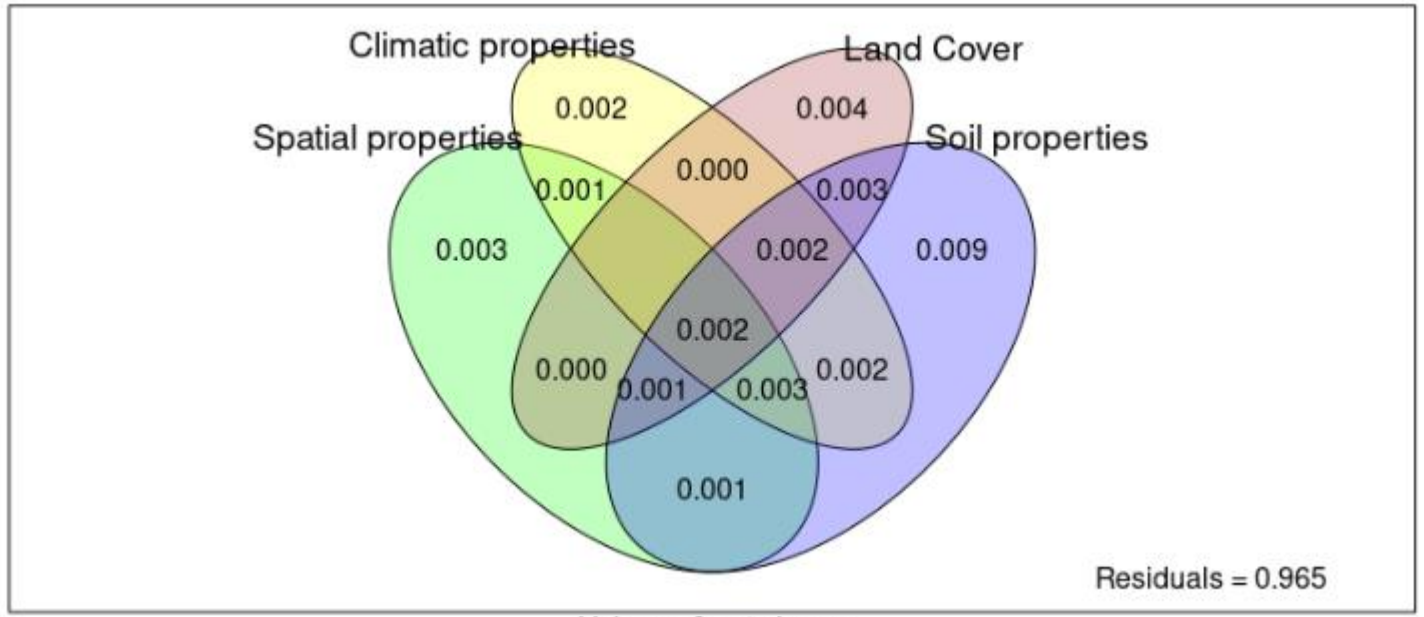
Figure 3. dbRDA plots for **D** fungi **E** protists

RESULTS – Beta diversity

A. Macro-fauna



B. Meso-fauna



Values <0 not shown

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CONCLUSIONS

- Environmental drivers differ for different groups of eukaryotes
- Big share of variation is still unexplained
- Potential/outlook: consider management practices

Many thanks!

Julia Köninger

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