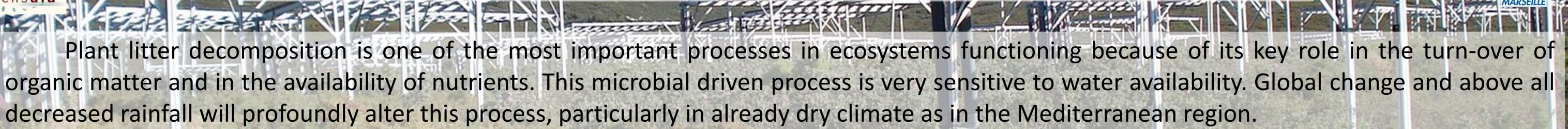


Effects of seasonality, plant identity and leaf-litter diversity on microbial communities in Mediterranean shrubland

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The CLIMED project (CLImate change effects on MEDiterranean biodiversity and consequences for ecosystem functioning) aims to assess the effects of reduced precipitation and plant species diversity on soil microbial and animal diversity and processes they drive in a shrub-dominated typical Mediterranean ecosystem.

This study precedes an in situ rain exclusion experiment and aims to better understand and interpret future results. We compared the effects of seasonality and plant species identity and mixture on the structure and diversity of bacterial and fungal communities in leaf-litter.

MATERIALS AND METHODS

The study site is a Mediterranean shrubland located near Marseille (France):





We considered all possible mixtures of leaf-litter of four typical woody shrub species collected on the ground:









Leaf-litter was collected at the end of each season in December 2010 (autumn), February (winter), June (spring) and September (summer) 2011.

Total DNA extraction from dry litter finely crushed (100 mg)

Molecular fingerprinting method: PCR amplification of bacterial and fungal ITS rDNA spacer

Separation of Operative Taxonomic Units (OTUs) by capillary electrophoresis

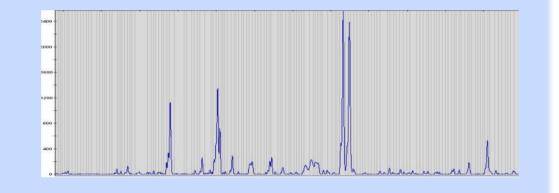
(ABI 3730, Plateforme Génomique du Genopôle de *Toulouse,* France)

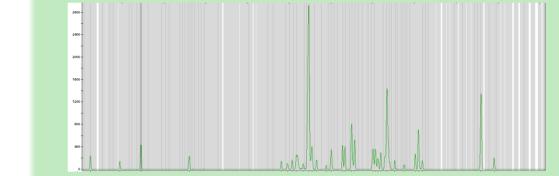
Statistical analysis were realized using Primer v6 and Permanova v1 (Primer E LTD).

RESULTS AND DISCUSSION

Bacterial community: ARISA PCR

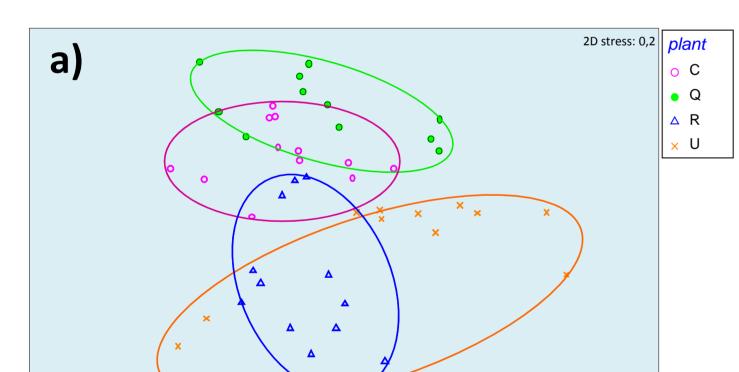
[FAM] S-D-Bact-1522-b-S-20 & L-D-Bact-132-a-A-18





Fungal community: T-RFLP PCR

[HEX] ITS1-F & ITS4 + HinF1 restriction of the PCR products

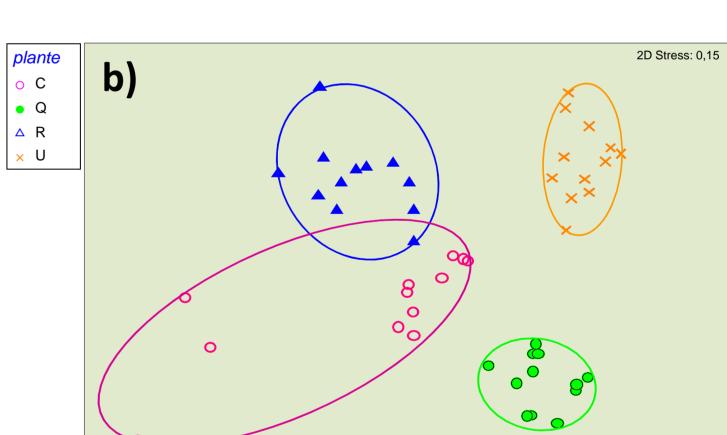


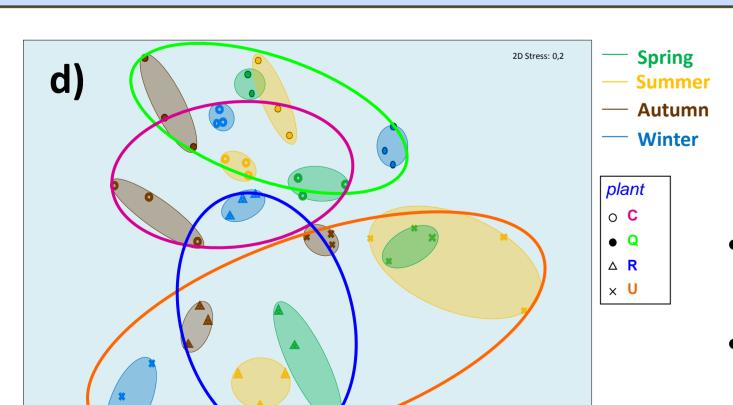
1. The effects of plant species

NMDS plots based on the Bray-Curtis similarity index (fig. a and b) for microbial community in monospecific litter and values of microbial diversity index (fig. c)

- Strong impact of plant identity on the structure of microbial community (p = 0.001).
 - → Plant identity drives the structure of microbial decomposer community.
- Richness and diversity of bacterial community are higher than those of fungi.
- Cistus albidus holds more bacterial diversity than other plant species.
- Quercus coccifera holds more fungal diversity than other plant species.
 - → Microhabitat and resources availability differ according to the plant species?

	Bacterial diversity index		Fungal diversity index		
c)	Species	Shannon	Species	Shannon	
	Richness (S)	index (H')	Richness (S)	index (H')	
Cistus albidus	150	6,5	29	0,93	Cistus albidus
Quercus coccifera	125	5,79	54	1,07	Quercus coccifera
Rosmarinus officinalis	135	6,03	24	1,074	Rosmarinus officinalis
Ulex parviflorus	118	5,12	33	0,896	Ulex parviflorus
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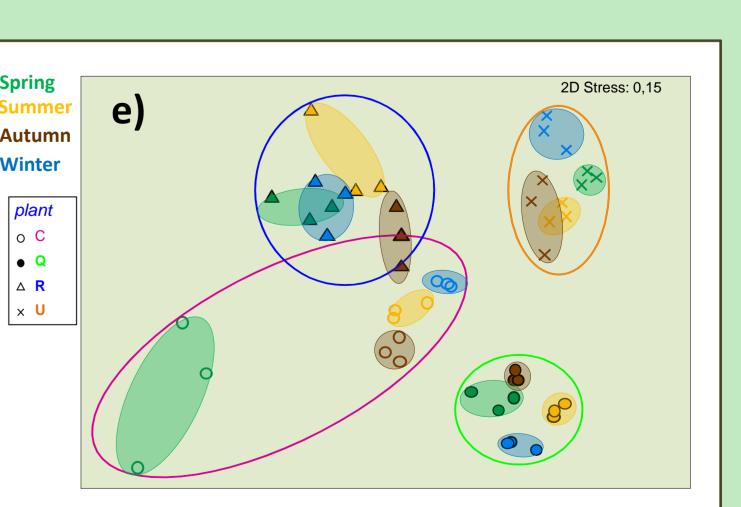


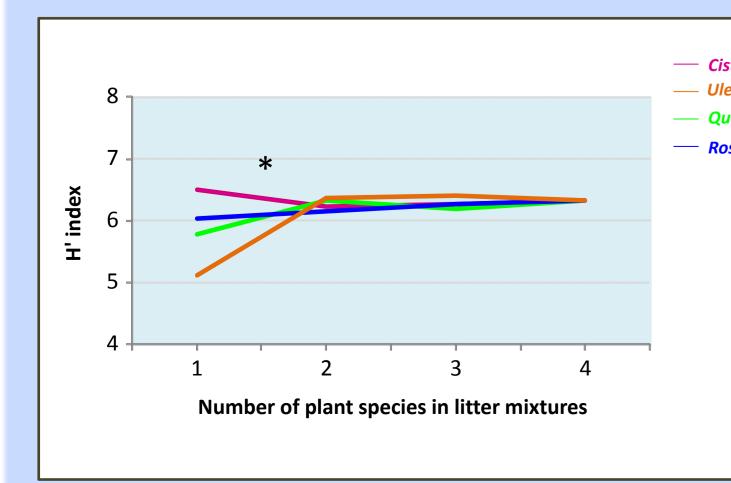


2. The effect of seasonality

NMDS plots based on the Bray-Curtis similarity index (fig. d and e) for microbial community in monospecific litter

- Strong impact of season changes on microbial community (p = 0.001).
 - > Changes in temperature and rainfall alter the microbial community structure.
- The seasonality effect is included in the plant identity effect.
 - → Does microbial community structure respond to seasonality as a natural hydric stress?
 - → Will microbial community respond the same way to experimental rain exclusion?





3. The effect of litter mixture

Microbial Shannon index $H' = -\sum [Pi \times log_2(Pi)]$

- Microbial community are strongly disturbed when the number of plant species increases from one to two species in the litter mixture.
- Then, microbial diversity holds steady (plateau) beyond two plant species in the mixtures. → Homogenization of the bacterial and fungal communities in litter mixtures with
- increasing number of plant species.

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