Effects of soil water repellency on microbial community structure and functions in Mediterranean pine forests

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Introduction

Soil water repellency (SWR) is a property commonly observed in forest areas showing wettable and water repellent patches with high spatial variability. SWR can greatly influence the hydrology and the ecology of forest soils. The capacity of soil microorganisms to degrade different organic compounds depends upon species composition, so this may affect changes in SWR at microsite scale (such as the presence of soil water repellent patches; Mülleret al., 2010). In the Mediterranean forest context, SWR has been found to be related to microbial community composition. The accumulation of different hydrophobic compounds might be causing the shifts in microbial community structure (Lozano et al., 2014). In this study we investigated the effects of SWR persistence on soil microbial community structure and enzyme activity under *Pinus halepensis* forests.

Materials and Methods

The study areas are located in three sites which differ in soil characteristics and mean annual precipitation: Petrer, Gorga and Javea (Alicante, E Spain) (Table 1).

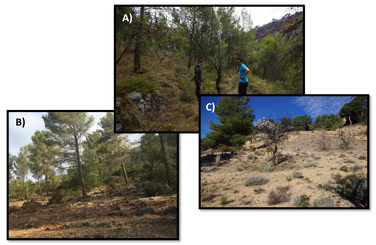

Fig. 1: Soil sampling sites: A) Gorga, B) Javea and C) Petrer.

Soil samples were collected from the first 2.5 cm of the mineral A horizon at microsites beneath *P. halepensis* (n=15 per site).

Table 1: Main characteristics of the study sites areas.

Site	Location Coordinates	Mean Annual Precipitation (mm)	Lithology	Soil classification (SSS, 2010)
Petrer	38°30'48"N, 0°46'33"W	371	Cretaceous limestone	Psamment
Gorga	38° 43'94"N, 0° 22'58"E	500	Jurassic limestone	Xerorthent
Jávea	38°48'7"N, 0°06'11"E	675	Cretaceous limestone	Rhodoxeralf

Water Drop Penetration Times were classified in intervals and in classes according to Bisdom et al. (1993).

Wettable WR< 5 s
Slight WR >5s < 60 s
Strong WR > 60s

Table 2: Mean of Water Drop Penetration Time (WDPT) and standard deviation of each samples and site.

Sample	Javea	Gorga	Petrer
1	230 ±27.8	71±12.1	223±15.7
2	40 ±20	9±1.7	1±0
3	3 ±0	31.3±3.2	1±0
4	1.7 ±0,57	13±1	6.7±1.5
5	164.7 ±4,5	20,7±3.7	1±0
6	1±0	1±0	8±1
7	6±1.7	22,3±2.5	1±0
8	8.7±2.5	5±0	8±1
9	1±0	6±2.1	66.3±7.1
10	43±13.9	2±0.6	30.7±1.5
11	6.3±2.3	7±2.3	62. 7±3
12	1±0	3±0	4.7±0.5
13	10.3±2.1	4±1.2	1±0
14	9.7±3.8	1±0	1±0
15	1±0	1±0	1525±63.6

Glomalin Related Soil Protein (GRSP): Easily extractable glomalin (EEG) was extracted from soil autoclaving with citric acid buffer and detected with a Braddford assay (Wright & Updahyaya, 1998) (Fig. 2)

Phospholipid fatty acids (PLFA) Lipids were extracted from 1.5 g soil using the procedure described by Bligh and Dyer (1959) and Frostegård et al. (1991).

Laboratory Methods

Soil organic content (Walkley & Black,1934) and **pH** (1:2.5 w/v)

The activities of **enzymes** which degrade cellulose trough cellobiohydrolase (Cell) and 1,4-b-glucosidase (Glu), chitin trough 1,4-b-N-acetylglucosaminidase (Nag) were determined using Methylumbelliferone-linked substrates (MUB) (modifying methods of Marx et al. (2001) and Sinsabaugh et al. (2000, 2003)).

Soil Water repellency (SWR) was measured by the Water Drop Penetration Time test (WDPT) (Wessel, 1988) under controlled conditions (20ºC and 50% of air humidity) (Fig. 2)

Fig. 2: Water drops over water repellent soil. Photo. V. Arcenegui

Results and Discussion

Microbial community structure

The results of this study showed different patterns of soil microbial community structure between sites. The RDA of Javea soil explain the 55% of the distribution. Axis 1 and 2 explain the 47.8% and 7.2 % respectively. Most of water repellent samples were located along the positive part of axis 2. SWR and GRSP were the variables more related with the Axis 2 (Fig. 3). Certain PLFAs were strongly associated with water repellent samples; PLFAs i15, i16, i17, which are mainly representative of Gram (+) bacteria,17:0 10m associated with actinobacteria (Fig. 3). The PLFA 18:2w9, representative of fungi, was also associated with these samples.

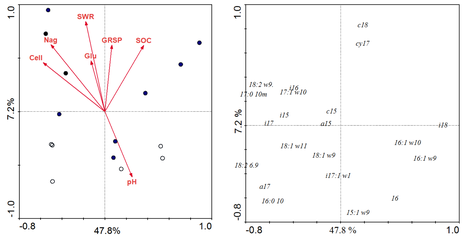

Fig. 3. A) Javea soil samples and soil characteristics biplots from RDA performed on the relative concentration of PLFA's. B) Samples and loadings plots from RDA performed on the relative concentration of PLFAs SOC: soil organic carbon; SWR: soil water repellency. GRSP: glomalin related soil protein; Cell: cellobiohydrolase; Glu: 1,4-b-glucosidase; Nag:1,4-b-N-acetylglucosaminidase. Blue circles: slight soil water repellent samples. Dark circles: Strong water repellent samples. Empty circles: wettable samples.

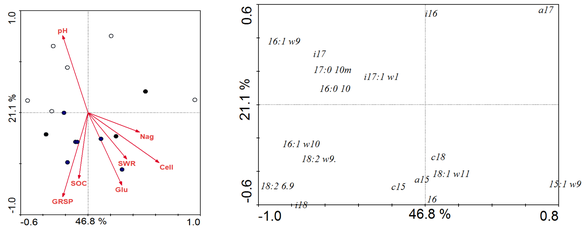

Fig. 4. A) Petrer soil samples and soil characteristics biplots from RDA performed on the relative concentration of PLFAs in . B) Samples and loadings plots from RDA performed on the relative concentration of PLFAs SOC: soil organic carbon; SWR: soil water repellency. GRSP: glomalin related soil protein; Cell: cellobiohydrolase; Glu: 1,4-b-glucosidase; Nag:1,4-b-N-acetylglucosaminidase. Blue circles: slight soil water repellent samples. Dark circles: Strong water repellent samples. Empty circles: wettable samples

The RDA of Petrer samples explain the 68%% of the distribution. Axis 1 and 2 explain the 46.8% and 21.1 % respectively. Almost all water repellent samples were located along the negative part of axis 2. GRSP and cellobiohydrolase were the variables more related with the Axis 2.(Fig. 4). The PLFA's 18: 2 w 6,9 and 16:1 w 10, representative of fungi, were associated with SWR samples.

The distribution of microbial community in Gorga samples did not showed any influence of presence of SWR (Fig. 5), a lower persistence of SWR could be a reason (table 2). pH and Nag were the variables which mainly explained the results.

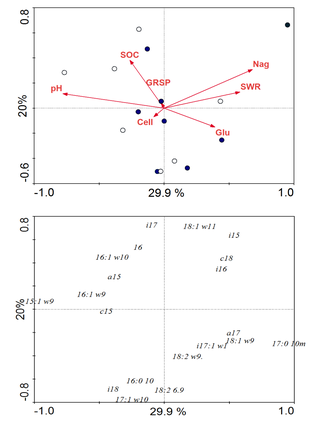

Fig. 5. A) Gorga soil samples and soil characteristics biplots from RDA performed on the relative concentration of PLFAs . B) Samples and loadings plots from RDA performed on the relative concentration of PLFAs. SOC: soil organic carbon; SWR: soil water repellency. GRSP: glomalin related soil protein; Cell: cellobiohydrolase; Glu: 1,4-b-glucosidase; Nag:1,4-b-N-acetylglucosaminidase. Blue circles: slight soil water repellent samples. Dark circles: Strong water repellent samples. Empty circles: wettable samples

Total PLFA's was higher in slight water repellent samples in the three sites studied. In Gorga and Petrer, the lower total content was found in strong water repellent samples (Fig. 6).

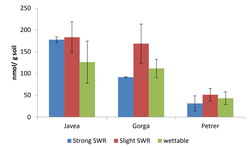

Fig. 6. Total PLFA's (nmol/ g soil) at each site

Enzyme activity was higher in water repellent samples. The mean activity of 1,4-b-glucosidase was higher in slight water repellent samples (Fig. 7). Both total PLFA's and enzymes activity were much lower in Petrer site. Results in this site showed a clear influence of SWR in the activity of enzymes, which was always higher in samples with slight water repellency.

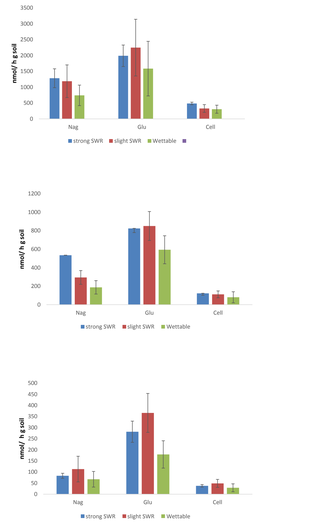

Fig. 7. Enzyme activity (nmol/ h g soil) in samples grouped by the different soil WR class. (A) Javea site; B) Petrer site; C) Gorga site. Standard errors in bars. ; SWR: soil water repellency; Cell: cellobiohydrolase; Glu: 1,4-b-glucosidase; Nag:1,4-b-N acetylglucosaminidase

Conclusions

In the Mediterranean forest context, SWR was found to be partially related to microbial community composition. The accumulation of different hydrophobic compounds might be influencing the microbial community structure and function or *viceversa*. However, soil texture could be an important parameter influencing such effect. Differences in the soil parameters (moisture, temperature, GRSP accumulated, quality of SOM) at the spatial microscale might be also responsible for the shifts in the microbial community composition, due to their role in the SOM mineralization. The accumulation of specific organic compounds and less water availability could be influencing the activity and structure of the microbial community. We provided further evidence that structure and fuction of the microbial community are closely linked to the presence of SWR.

References

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