MOLECULAR AND BIOCHEMICAL CHARACTERIZATION OF SOILS FROM SEMIARID MEDITERRANEAN ECOSYSTEMS

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1. INTRODUCTION AND OBJECTIVES

Microorganisms play an essential role in the maintenance of ecosystems - whether directly, in biogeochemical cycles, or indirectly, by participating in the processes of degradation and transformation of soil organic matter.

In spite that the environmental functions of microorganisms in soil have been widely analyzed by biochemical properties and the advances in molecular biology have made possible to study these microorganisms, until now the relationships between the structure of the microbial communities and their environmental functions have not been established.

The objectives of the present study are: i) To compare the information provided by molecular and biochemical methods, in order to obtain a better understanding of the relationships between the structure of microbial communities and the functions of these microorganisms in soil; ii) To determine whether these molecular properties can be used to assess human impact in semiarid Mediterranean soils located at different altitudes and under different types of land use and vegetation.

2. MATERIALS AND METHODS

Soils samples of an arid-semiarid zone in the southeast of Spain under different land uses and vegetation covers were collected: natural pine forests of *P. halepensis* Mill. (1M) and *P. nigra* Arnold (6M), natural evergreen oak forests (5M, 8M), juniper shrubs (2M), scrub (3M), reforested *P. halepensis* pine stands less than 20 years old (7M), and almond tree crops (4M).

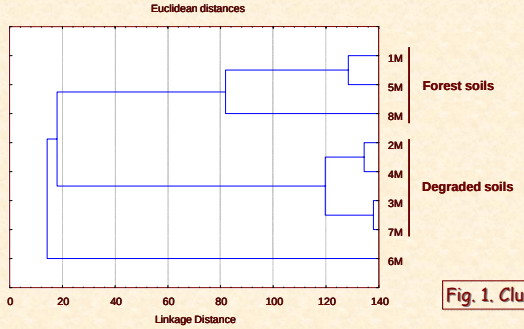
Physico-chemical (total C, total N, textural class, carbonates and pH), biochemical (Microbial biomass carbon, Soil respiration (SR), Nitrogen mineralization and some enzymatic activities [dehydrogenase (DESH), catalase (CAT), phosphodiesterase (DIES), β-glucosidase (GLUC), urease (URE) and casein-protease (CAS)] and molecular (16S RNA-based phylogenetic analysis) methods were applied in all soil samples (Table 1).

Table 1. Physico-chemical and biochemical soil properties

	1M	2M	3M	4M	5M	6M	7M	8M
Altitude (m)	892	988	1141	1230	1398	1520	1053	1122
Total C*	71.7	43.0	34.7	16.4	68.0	102.8	23.5	72.9
Total N*	4.63	3.10	3.48	1.64	5.57	5.24	2.01	3.74
pH in water	7.80	8.10	8.30	8.40	7.70	8.00	8.00	7.70
Sand*	262	207	391	319	292	251	458	68
Clay*	393	455	237	394	431	296	253	612
$CaCO_3$*	415	341	528	159	245	418	537	11
Biomass-C#	1264	709	808	565	1490	2173	579	1460
SR$	317	117	137	112	329	454	137	259
DESH+	0.49	0.19	0.32	0.23	0.38	0.56	0.21	0.44
CAT&	3.31	2.77	2.32	2.92	3.30	4.20	1.75	3.32
DIES^	1.24	0.61	0.75	0.67	1.26	1.52	0.47	1.61
GLUC^	2.18	0.80	1.56	1.67	2.58	2.52	0.96	1.85
URE##	11.09	5.15	5.95	4.64	9.95	17.06	4.32	12.14
CAS$$	0.76	0.16	0.31	0.18	0.94	1.29	0.33	0.53

*, g kg^{-1}; #, Biomass-C (mg kg^{-1}); $, mg CO_2-C kg^{-1} 10 d^{-1}; +, µmol INTF g^{-1} h^{-1}; &, mmol H_2O_2 g^{-1} h^{-1}; ^, µmol *p*-nitrophenol g^{-1} h^{-1}; ##, URE, µmol NH_3 g^{-1} h^{-1}; $$, µmol tyrosine g^{-1} h^{-1}.

3. RESULTS AND DISCUSSION



Analysis of the biochemical parameters microbial biomass carbon, soil basal respiration, qCO_2 and enzymatic activities enabled differentiation of soils according to vegetation/land use (forest soils and degraded or cultivated soils) (Fig 1).

Fig. 1. Cluster analysis based on biochemical properties.

Analysis of nitrogen mineralization also enabled differentiation of soils according to altitude (Fig 2).

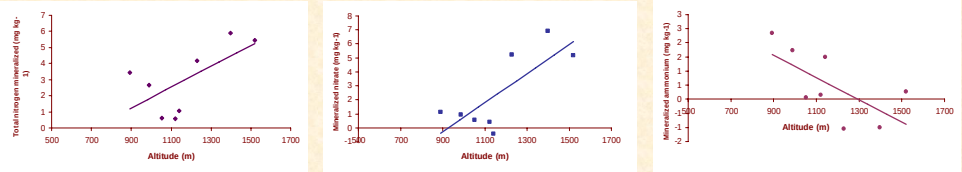


Fig. 2. Trend of total nitrogen, ammonium and nitrate mineralized of soil samples with the altitude.

The study of microbial structure by PCR-TGGE with specific primers (alpha and beta-proteobacteria) also revealed that some subpopulations of microbes behaved differently from others, depending on environmental variables (vegetation/land use and altitude). Beta-proteobacteria profiles grouped the samples according to land use and vegetation, whereas alpha-proteobacteria profiles clearly grouped the samples according to altitude (Fig 3).

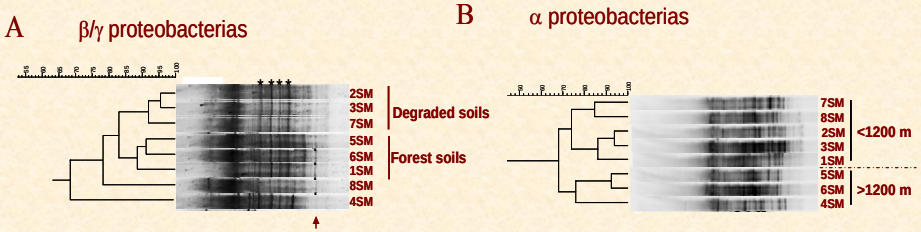


Fig .3. TGGE gel of PCR amplicons from soil samples and the corresponding cluster analysis; (A) primers specific to β/γ-Proteobacteria; (B) primers specific to alfa-Proteobacteria.

4. CONCLUSIONS

The results suggests that, in general, there is concordance between the structure of microbial communities estimated by PCR-TGGE of 16S rDNA genes and the functions these microorganisms have in the environment, as estimated from their biochemical properties. The methodology used may provide new lines of research that could be developed and simplified in the future (use of primers specific to different taxonomic units) and therefore, with potential for use as indicators of anthropogenic activity.