



## Research paper

# Soil micro-biological indicators separated land use practices in contrast to abiotic soil properties at the 50 km scale under summer warm Mediterranean climate in northern Italy



Oliver Dilly<sup>a,b,\*</sup>, Letizia Pompili<sup>c</sup>, Anna Benedetti<sup>c</sup>

<sup>a</sup> DLR – Deutsches Zentrum für Luft- und Raumfahrt e. V., Projektträger, Bereich “Umwelt und Nachhaltigkeit”, 53227 Bonn, Germany

<sup>b</sup> Dr. Joachim und Hanna Schmidt Stiftung für Umwelt und Verkehr, Eichholz 56, 20459 Hamburg, Germany

<sup>c</sup> Consiglio per la Ricerca in Agricoltura e l'Analisi dell'Economia Agraria – Centro di Ricerca Agricoltura e Ambiente, Via della Navicella 2, 00184 Roma, Italy

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## ABSTRACT

Soil abiotic properties represent important framing conditions of ecosystems and soil microbiological biomass and microbial activity are the biological driving forces that respond sensitive to environmental impacts. In environmental research and monitoring programmes, approaches for spatial classification were elaborated poorly. Therefore, soil abiotic and microbiological indicators were clustered for a 2965 km<sup>2</sup> region in northern Italy with the aim to identify the key spatial characteristics. Forty one sampling points for 30 cm soil depth were selected at the European Grid of the Land Use Cover Area frame Statistical survey (LUCAS). Soil abiotic indicators included physical (clay content and soil bulk density) and chemical (Cd, Pb and Hg content, organic carbon and total nitrogen) properties. The values varied between 3–47 g clay 100 g<sup>-1</sup> soil, 1.27–1.62 Mg m<sup>-3</sup>, 0.69–3.47 g organic C 100 g<sup>-1</sup> soil and 0.07–0.31 g total N 100 g<sup>-1</sup> soil. Our cluster analysis with the abiotic characteristics separated two core classes derived from four clusters. In contrast, soil microbiological indicators including microbial carbon, respiration values and microbial energetic quotients, separated three well-defined core classes derived from five clusters. The microbial properties values ranged between 24–431 µg microbial C g<sup>-1</sup> soil and 0.08–1.82 µg CO<sub>2</sub>-C g<sup>-1</sup> soil h<sup>-1</sup> and corresponded to those in other European agricultural soils. The three soil microbiological classes separated low, typical and high ranges for soil microbial biomass (values < 120, 50–250 and > 320 µg microbial C g<sup>-1</sup> soil), for soil respiration rates (values < 0.4, 0.4–0.7, and > 1.4 µg CO<sub>2</sub>-C g<sup>-1</sup> soil h<sup>-1</sup>) for metabolic quotient (values < 3.5, 3.5–5.0, > 5.0, mg CO<sub>2</sub>-C g<sup>-1</sup> C<sub>mic</sub> h<sup>-1</sup>) and for microbial C to organic C ratio (values < 0.7, 0.5–1.7 and > 1.0%). The soil microbiological indicators responded more evidently to land cover and soil management practices while soil abiotic indicators that referred mainly to the framing conditions such as geology and climatic conditions.

## 1. Introduction

Soil microorganisms give useful information of changing system conditions (Hargreaves et al., 2003). They are considered as sensitive drivers and thus indicators of soil health (Yakovchenko et al., 1996; Bloem et al., 2006). Thus, measurements of microbial activities are included in international monitoring programmes on soil quality (Powelson, 1994; Dilly, 2006a). Soil quality can be broadly defined as the capacity of a soil to function, within land use and ecosystem boundaries, to sustain biological productivity, maintain environmental quality, and promote plant, animal, and human health (Doran and Parkin, 1994). An understanding of the spatial variability of microbial processes is an important factor in the development of meaningful global circulation models

(Parkin, 1993). Most of the work at the tempo-spatial scale was done for trace gases or C and N dynamics in the field and pH value or other abiotic parameters (Mc Bratney and Webster, 1983; Röver et al., 1999; Bruckner et al., 1999; Wang et al., 2007). These studies were helpful to quantify and to model the heterogeneous nature of soils. In contrast, soil microbial biomass, structure of microbial communities (e.g. biodiversity) and microbial activity are driving forces such as for the release of nutrients in agricultural and natural ecosystems. However, their use as potential sensitive indicators of soil quality changes at spatial scale is still poorly developed (Bonmati et al., 1991; Wich et al., 1998; Mummey et al., 2002). The spatial soil variability may control variations of soil processes. Particularly specific environmental conditions and human management practices may increase variation.

\* Corresponding author at: DLR – Deutsches Zentrum für Luft- und Raumfahrt e. V., Projektträger, Bereich “Umwelt und Nachhaltigkeit”, 53227 Bonn, Germany.  
E-mail address: [oliver.dilly@gmail.com](mailto:oliver.dilly@gmail.com) (O. Dilly).

Cluster analysis is a robust data analysis technique that combines characters, observations and indicators in ‘acceptable homogeneous sets’. Cluster analysis is applied for environmental, economic and social sciences and sustainability analysis (Dilly and Hüttl, 2009). In particular in K-means analysis, the observations in the dataset are divided into K partitions, and each portion represents a cluster (Sharma 1996). Here, K-means analysis was selected to detect spatial patterns and at their controls. This was a study within the research project “Biodiversity and bio-indication in Pavia province” addressing soil quality with spatial evaluation (Cenci and Senna, 2006). This study included data on soil pollution, basic physical and bio-chemical properties and land use. These parameters were the basis for separating core classes related to climatic, abiotic pedogenic and environmental conditions, land cover and land management practices. It was hypothesised that the soil microbiological indicators allow a better evaluation of soil quality at a spatial level. Soil abiotic and biotic indicators related mainly to carbon and energetic characteristics to assess an integrative “energomics” view (Dilly, 2006b). Last but not least, high-to-low value ranges of microbial carbon and microbial basal respiration and the respective quotients should be identified for the Pavia province’s soils.

## 2. Materials and methods

### 2.1. Study area and sampling

Soil samples were collected in the north of Italy, in the Pavia province with a surface area of 2965 km<sup>2</sup> and within the framework of monitoring actions between 2004–2006 (Brenna and Filippi, 2006; Cenci and Senna, 2006; Renzi et al., 2017). Forty-one samples were collected. Approximately 70% of the territory is flat land, included in Pianura Padana, which is located at north of Po river. In the north-western area of the province, land is mostly cultivated by rice (n = 9) because of the high availability of water due to two important rivers Sesia and Ticino and numerous channels for irrigations; corn (n = 7), wheat (n = 4) and soy (n = 1) are the other crops. In the central-eastern area, poplar plantation and meadows were present. Modern agricultural management practices tended to maximise yield by using mineral fertilisers and pesticides. Soils were classified as *Luvisols*, *Calcisols* and *Arenosols* (IUSS Working Group WRB, 2006). The remaining 30% of the territory is hilly and mountainous and it is located in the south reaching the Appennini chains. This area is mainly covered by mixed natural woods, chestnut woods (forest n = 11) and pastures (meadows/pastures n = 9). Soils were classified as *Calcisols* and *Cambisols*. Climate of entire area is warm-temperate with fully humid and summer warm Mediterranean (Köppen Cfb) climate. Winter are fairly cold (January is the coldest month), summer are warm and moist. Fog is typical characteristic of the climate in Pavia province because of the presence of water basins.

Soil was sampled according to the European Grid of the Land Use Cover Area frame Statistical survey (LUCAS). This approach is a statistical method proposed by Eurostat (2003) in close co-operation with the General Directorate of Agriculture of European Commission. It is based on the observation of sample points, which are used to representative of the entire area under investigation. The LUCAS survey is based on a two-stage sampling design: the first step identifier, 7 points as cells of a regular grid with a size of 18 × 18 km (Principal Points, PP); the step identifier, 34 points distributed in a grid and each cell is 9 × 9 km (Secondary Points, SP). Soil was sampled at 0–30 cm depth, air-dried and sieved at 2 mm mesh.

### 2.2. Soil abiotic properties

Average clay content and soil bulk density were derived from soil geo-referenced database (scale 1:250.000) of the Lombardy region (ERSAF, 2004). In particular each cartographic polygon is linked to a dominant Soil Typological Unit (STU) to whom other subordinated STU

are related. Total contents of Pb and Cd were determined by Atomic Adsorption Spectrometry (AAS), total Hg by Optical Plasma (ICP-OES), and total N by CHN Analyzer. Total organic C, C<sub>org</sub>, was determined as reported by Springer and Klee (1954).

### 2.3. Soil microbiological properties

Soil microbial estimates in the laboratory included microbial carbon, C<sub>mic</sub>, (Vance et al., 1987) and soil respiration (Isermeyer 1952). Soil microbial respiration was measured daily during a 14 days period. The respiration of the last day was considered as basal respiration, CO<sub>2</sub>-C<sub>bas</sub>, and expressed on an hourly basis. A non-linear least-square regression analysis,  $C_{-min} = C_0 [1 - \exp(-k_{MIN} \cdot t)]$ , was used to calculate kinetic parameters (mineralization kinetic rate, k<sub>MIN</sub>, and potentially mineralizable carbon, C<sub>0</sub>) from cumulative data of C-mineralization, C<sub>-min</sub>, respect to time (t = days) according to Riffaldi et al. (1996). We also calculated the specific respiration of soil microbial communities, the metabolic quotient qCO<sub>2</sub> representing the C use efficiency of the microbial communities (Anderson and Domsch, 1993) and the percentages of organic C respired as carbon CO<sub>2</sub>-C<sub>bas</sub>, the maintenance requirement qM (Dommergues, 1960) and biotic carbon, C<sub>mic</sub>/C<sub>org</sub> × 100 (Anderson and Domsch, 1989).

### 2.4. Statistical analysis

Spatial data were processed by the K-means analysis, which is a classification system where K is considered the centroid of the cluster and observations of the cluster are all closer one to other. The K value must be given *a priori* by considering the nature of the observations. Relatively homogenous groups of areas with similar indicator profile should be obtained by a reasonable and non-redundant number of clusters. The selected K value was chosen according to the number of observations (n = 41), and the classification of microbial indicators.

A soil data-matrix was used to compare indicator units against to each other. Mean, standard deviation (SD) and standard error (SE) were calculated to evaluate the overall variability for biotic properties inside the clusters.

Statistical analysis were performed by Statistica 6.0 (data analyses software system) distributed by Stat Soft Italia (2001).

## 3. Results

### 3.1. Soil abiotic characteristics

Four clusters with 5–15 observations each were separated with Euclidean distances between 0.724 and 1.463 (Table 1). The observations were located closely within each cluster. Clay, organic C and total N were the most important factors separating the ‘abiotic’ clusters (Fig. 1). Heavy metals contents and bulk density were less important here (data not shown).

Although the four clusters showed some overlapping organic C and total N values, the spatial classification could be further advanced and simplified based on the Euclidean distances and the inclusion of indigenous and local knowledge (ILK). The Euclidean distances and ILK separated most evidently two core classes for the soil abiotic properties

**Table 1**  
The number of observations (n) and Euclidean distances when clustering soil abiotic characteristics with K means of 4. The highest numbers represents most distant clusters.

	n	Cluster 1	Cluster 2	Cluster 3
Cluster 1	5			
Cluster 2	15	1.189		
Cluster 3	12	0.937	0.724	
Cluster 4	9	1.463	0.793	1.284

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