



Comparison of absolute biochemical parameters of undisturbed soils in Mediterranean environments (NE Spain) with corresponding parameters relative to soil organic carbon

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ABSTRACT

The study of soil quality requires the establishment of quality standards. To this end, several authors have highlighted the need to create databases of quality indicators, such as biochemical properties, for different types of undisturbed soils under various climates and to establish standardised methodologies for their development. In Spain, studies of the quality of native soils were initiated > 15 years ago by several groups of authors from differing locations, but little is known regarding the biochemical characteristics of native soils in Catalonia (NE Spain). This study examines representative, minimally disturbed soils from Catalonia with a wide range of organic carbon contents. We examined the total and extractable organic carbon contents, total and extractable carbohydrates contents, enzyme activities (β -glucosidase, β -galactosidase, BAA-protease and urease), microbial biomass carbon and basal respiration of ten selected soils. Statistical analyses were applied to absolute values (i.e., per g of soil) and relative values (i.e., per g of soil organic carbon). The aim of this work was to determine the dependence of these properties on the organic matter content and the suitability of the relative values as soil quality indicators. The biochemical and microbiological parameter values of the native Catalan soils showed unusually wide ranges, although all of the values were similar to those already published for native soils in other Mediterranean climate areas. Overall, the sampled soils could be distinguished by their contents of organic carbon and total and extractable carbohydrates, rather than by their enzyme activities or microbiological variables; nevertheless, when the relative values were considered, the soils could be distinguished by their specific enzyme activities, particularly that of β -glucosidase, and by the labile proportion of organic matter. With the exception of the total carbohydrates/C ratio, the biochemical and microbiological parameters, expressed as functions of soil organic carbon content, were useful in distinguishing groups of native soils according to field observations and soil physicochemical properties.

1. Introduction

The lack of established quality standards is a critical issue for the study of soil quality. The choice of native (undisturbed) soils as references is based upon the association of maximum quality with a sustainable balance between soil components, under characteristic climate and vegetation conditions, and subject to little or no human disturbance (Doran and Parkin, 1994; Karlen et al., 1997). The biological and biochemical parameters of soils are particularly suitable as indicators of their quality because they respond to both natural and human-induced changes (Elliot et al., 1996; Gregorich et al., 1997; García et al., 2000;

Filip, 2002; Gil-Sotres et al., 2005; Bastida et al., 2008b).

Early recommendations for basic indicators of soil quality already included biological characteristics. According to Melé and Crowley (2008), who examined 52 soil quality monitoring programmes developed worldwide through the end of 2003, 29% used biological indicators. Gil-Sotres et al. (2005) found that 40% of the publications on soil quality from 1990 to 2003 reported general biochemical parameters, while approximately 60% used specific ones (e.g., hydrolytic enzyme activities). More recently, 55–80% of studies (which considered only agricultural, forest and land use change) included biological indicators, according to a revision of the most common indicators used in

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soil quality assessment over the last 15 years (Zornoza et al., 2015).

The review by Bastida et al. (2008b) of the biological aspects of the quality of non-agricultural soils indicates that the most relevant works have been performed by Italian and Spanish authors. In Spain, studies of the quality of native soils were begun > 15 years ago and have been undertaken by several groups of authors on soils from various geographic conditions (García et al., 2000, 2003). In Galicia (Spain), the study of native soils has focused on Umbrisols under Atlantic oak-woodland vegetation and a humid climate (Trasar-Cepeda et al., 1998, 2000, 2008a, 2008b; Leirós et al., 1999, 2000). In a Mediterranean climate, some authors studied soils in Murcia and Alicante (Spain), a rather heterogeneous territory, with high variability of climate and vegetation, including areas at risk of desertification (García et al., 1994; García and Hernández, 1997; Zornoza et al., 2007a, 2007b, 2008). All these studies of native soils from Spain have greatly contributed to the development and interpretation of soil quality data. However, no database exists that covers the whole Spanish territory and its lithological, climatic and vegetative diversity.

Scarce data are available concerning the biochemical characteristics of native soils in Catalonia (NE Spain) so we first performed a study of minimally disturbed soils of this territory in a previous work (Jiménez et al., 2012). Representative soils in our territory were studied including those covering a wide gradient of organic matter content. In this work, we provided preliminary information about biochemical properties; the results indicated that the studied biochemical parameters presented high and positive correlations between themselves, but analysis of organic carbon partial correlations indicated that these parameters were highly dependent on soil organic matter content. Consequently, we studied the same native soils, focusing on the behaviour of parameters expressed as a function of their soil organic carbon content (i.e., relative parameters), and present our conclusions herein.

The aim of this study was to elucidate, in non-modified soils developed under Mediterranean climate (NE Spain), the i) degree of influence of the soil organic carbon content on biochemical and microbiological parameters and ii) suitability of the relative parameters (i.e., per g of soil organic carbon) for distinguishing soils' characteristics. Thus, our hypothesis was that relative parameters would be able to group soils according to their general characteristics more accurately than absolute values.

2. Materials and methods

2.1. Sites and soil sampling

Soil samples were collected from ten locations in Catalonia (NE Spain): *Serres del Camp*, *Balaguer (BL)*, *Serra del Corredor (CR)*, *Conca d'Odena*, *Igualada (IG)*, *Serra de la Picarda*, *La Granja d'Escarp (LG)*, *Serra Litoral (LT)*, *Serra del Montnegre (MN)*, *Serra de l'Ordal (OR)*, *La Panadella plateau (PN)*, *Segre alluvial plain (SG)*, and *Plana de Vic (VC)*. An overview of the sites and their soil characteristics is presented in Tables 1 and 2. We focused on native soils, under autochthonous vegetation (corresponding, as much as possible, to potential vegetation) which had not been disturbed by human action for decades. At all locations, forest and abandoned agricultural soils were distributed over wide zones in a landscape mosaic. To validate soil results, we selected four land uses: undisturbed (or subject to little disturbance) forest; abandoned agriculture field; dry grassland; and steppe.

The climate in these areas is of the semiarid Mediterranean type. The average annual temperature ranges from 9 to 16 °C and rainfall varies from < 400 to 825 mm/year (ICC and SMC, 2008). The common rocks in this area are carbonate rocks (limestone, marls, alluvial and colluvial deposits) together with silica rocks (shales and granodiorite). The dry climatic conditions promote erosion, physical degradation and salinisation of these soils. The vegetation developing on the sampled soils varies from site to site. The *BL* soil supports a xeric shrubland of *Rosmarinus officinalis* L. *IG* and *LG* soils were found in the lowland and

midland dry grasslands, with rocky surfaces. The *LG* soil in particular corresponds to a steppe-like vegetation, well-adapted to low water availability, where the scarcity of rain prevents development of pastures and the vegetation is dominated by herbs and sparse shrubs. The *VC* soil is typical of *Aphyllanthes monspeliensis* L. grasslands, dominated by annual plants and gramineae. *CR* and *MN* soils support a Mediterranean woodland vegetation, dominated by holm-oak (*Quercus ilex* subsp. *rotundifolia* L.) and cork-oak (*Quercus suber* L.). The vegetation on *LT* and *PN* soils consists of holm-oaks (*Quercus ilex* L.). In contrast, the *OR* soil supports conifer-dominated woodlands, typically *Pinus pinea* L., *Pinus halepensis* Mill and *Pinus nigra* Arnold. The *SG* soil is associated with Mediterranean riparian woodlands where the most typical species are alder (*Alnus glutinosa* (L.) Gaertn), ash (*Fraxinus excelsior* L.) and black poplar (*Populus nigra* L.).

A plot of approximately 100 m² was defined in each site, and a sample composed of 20–25 homogeneously mixed sub-samples was collected from the topsoil (0–10 cm) after litter removal. Samples were collected on two consecutive days in spring, then immediately sieved to obtain fine earth (< 2 mm) and homogenised. One part was stored at 4 °C prior to biochemical and microbial analysis (within 15 days of sampling), while another was air-dried for a week and stored at room temperature before analysis of its chemical and physical properties.

2.2. Analytical methods

The main physical and chemical properties of the soil samples were characterised as follows. Texture was determined by the Bouyoucos method (Gee and Bauder, 1986). Electrical conductivity was measured in a 1/5 suspension, pH in a 1/2.5 (soil/water) suspension and total carbonates were measured using a Shimadzu TOC-V-Series analyser with a solid sample module SSM 5000A (Shimadzu Corporation, Kyoto, Japan) by adding diluted H₃PO₄ before heating at 200 °C. Total organic carbon was determined by potassium dichromate oxidation using the Walkley-Black procedure (Nelson and Sommers, 1982).

Carbohydrates were analysed in air-dried samples: total carbohydrates were determined by a double hydrolysis with H₂SO₄ (4 M and 0.5 M), as reported by Cheshire and Mundie (1966); and extractable carbohydrates (soluble in 0.5 M K₂SO₄) as described by Badalucco et al. (1992). Carbohydrate contents were measured by anthrone colourimetry (Brink et al., 1960).

Extractable organic carbon (extractable organic C) was obtained by extraction with 0.5 M K₂SO₄ (1:4 w/v dry soil: extractant ratio) and quantified using a Shimadzu TOC-V-Series analyser. Microbial biomass-C (MBC) was determined using the fumigation extraction procedure (Vance et al., 1987) in samples that had been pre-incubated for 7 days in the dark at 28 °C after being adjusted to 60% of their field capacity. Carbon dioxide emissions were determined in 100 g of soil previously adjusted to 60% field capacity and incubated for 7 days in the dark at 28 °C in sealed jars containing a vial with 10 mL of 0.5 M NaOH to absorb the gas; NaOH traps were removed daily during incubation. The quantity of CO₂ was determined by titration of NaOH with 0.5 M HCl (Hernández and García, 2003), and basal respiration (BR) values were obtained (after checking that daily CO₂ production was constant from the 5th day) by calculating the amount of CO₂ produced between the 6th and 7th days of incubation.

The method of Tabatabai and Bremner (1972) as modified by Nannipieri et al. (1978) was used to determine urease activity. BAA (N-benzoyl-L-argininamide) proteolytic activity was determined by the method of Ladd and Butler (1972) as modified by Bonmatí et al. (1998). β-Glucosidase and β-galactosidase activities were determined as reported by Tabatabai (1982), with calibration plots of p-nitrophenol prepared by using individual soil samples, thus taking into account the relative adsorption of p-nitrophenol by each soil (Vuorinen, 1993).

Results were expressed on two bases: a) dry weight soil (absolute values); and b) total organic C measured in soil (relative values). We designated the relative values of enzyme activities as “specific

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