



Early warning indicators of changes in soil ecosystem functioning



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ABSTRACT

In the last decades, soil is facing numerous environmental threats and climatic changes that are causing a rapid decline of soil fertility and biodiversity. Soil organic matter (SOM), has the most widely recognized influence on soil quality, but it hardly puts in evidence processes associated to the new soil threats, because of its insensitivity in assessing soil quality changes in the short-term. A series of chemical and biochemical analyses were carried out in agricultural and forestry soil ecosystems subjected to different threats, to identify the parameters that better evidence changes in soil characteristics in a short term, but the identification of basic universal indicators and the choice of the number of estimated measures are still under investigation and discussion. The main aim of this paper was to identify biochemical markers to be used routinely and applicable to different soil ecosystems, as early warning indicators of alteration in soil ecosystem functioning. The results obtained allowed to identify three indicators, microbial biomass (MBC), water soluble phenols (WSP), and fluorescein diacetate hydrolase (FDA), as effective tools in the evaluation of soil quality changes in the short term, showing also a threat-indicator specificity. MBC reflected changes mainly induced by abiotic stress, FDA displayed modification caused by climate, and WSP pointed out alteration due to the organic amendment.

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1. Introduction

Soils are the most significant non-renewable geo-resource we have and that are facing numerous environmental threats while trying to resist to climatic changes. Interest in evaluating the quality and health of our soils has been stimulated by increasing awareness that soil is a critical important component of the earth's biosphere, functioning not only in the production of food and fiber but also in the maintenance of global sustainability and environmental balance (Glanz, 1995). Soil is also the basis of agricultural and of natural plant communities. Thus, the thin layer of soil covering the surface of the earth represents the difference between survival and extinction for most land-based life (Doran and Parkin, 1996). Whilst the majority of countries have criteria to evaluate the quality of the air and water, the same does not occur for the quality of the soil. Traditionally, soil quality is associated with productivity (Karlen et al., 1997), but recently it has been defined in terms of sustainability (Tóth et al., 2007), that is, the capacity of the soil to absorb, store and recycle water, minerals and energy in such a way that the production of the crops can be maximized and environmental degradation minimized. Nevertheless, a significant decline in soil quality has occurred

throughout the entire world as a result of adverse changes in its physical, chemical and biological properties, caused by human activity and climate changes (Van Camp et al., 2004; EC, 2006). According to Steer (1998), in the last decades of the last century, about 2 billion of the 8.7 billion agricultural lands, permanent pastures, forests and wild native lands have been degraded. Soil degradation processes constitute a serious problem on a worldwide basis, with significant environmental, social and economic consequences. Many economic activities such as agriculture, industry and tourism depend both directly and indirectly on soil quality, which has been proposed as a prime indicator for characterizing and defining management factors contributing to soil degradation. Many constraints cause short-term disturbances that are detrimental to soil quality (IPCC, 2007; EEA-JRC-WHO, 2008) as they increase the emissions of greenhouse gases (i.e., CO₂, NO, or N₂O), cause nitrate accumulation and leaching, and/or modify soil microbial community structure in a way that decreases the retention of organic C and N (Liu et al., 2006). Generally, soil quality has been related to the SOM (Gao et al., 2013), microbial activity, total nitrogen, and C/N ratio (Molope and Page, 1986; Eash et al., 1994; Roberson et al., 1995; Murphy et al., 2011), but these soil parameters not necessarily change as a result of changing external conditions or use (Muscolo et al., 2014), and hardly address short term changes in soil processes associated to the new environmental threats. To rise the challenge of soil resource degradation, there is an urgent need to develop common, simple

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and transparent method to identify changes in soil characteristics in response to the main environmental constraints. Soil-quality assessment, based on inherent soil factors and focused on dynamic aspects of soil system (Paz-Ferreiro and Fu, 2014; Muscolo et al., 2014) is an effective method for evaluating the environmental sustainability (Hamblin, 1991) of land use and management activities. In these scenarios, the overall goal of this paper was to compare data on soils subjected to different types of use and environmental constraints, in order to find out biochemical markers to be used routinely and applicable to different soil ecosystems, as early warning indicators of changes in soil ecosystem functioning. A series of chemical and biochemical analyses were carried out in forest managed soils, amended agriculture soils, soil irrigated with brackish water and forest soil influenced by seasonal variation to identify the parameters that better reflect changes in soil quality, in the short term. The assessment was comparative because of the lack of specific criteria or guidelines available in the literature for interpretation of most soil property indices measured. The starting hypothesis was that natural soils have developed, over time, an equilibrium with the environment reaching the maximum quality and the greatest degree of balance in their properties (Fedoroff, 1987), but soil use and the new environmental constraints alter this balance by affecting soil biochemical properties even in a short time. (Fedoroff, 1987).

2. Material and method

2.1. Experiments and soil sampling

Four separate experiments were carried out to identify early warning indicators that better reflect changes in soil chemistry and biochemistry parameters related to soil quality. The experiments were conducted both in forest and agriculture soils underwent to different management practices and climate. The first experiment (named case study 1) was conducted in field, in order to evaluate the effects of artificial brackish water at different concentrations (0, 0.5%, 1%, 1.5%) on chemical and biochemical properties of a Haplic Kastanozem (IUSS, 2006) located in the Agricultural Farm of “Mediterranea University”, Reggio Calabria, Southern Italy. Soil during the dry season (June–August), have been irrigated, three times a week, with synthetic brackish water ($EC\ 4\ dS\ m^{-1}$) prepared using $NaHCO_3$, $NaCl$, Na_2SO_4 , and $MgSO_4$ with $Cl:SO_4$ ratio of 1:1 and $Ca:Mg$ ratios of 4:1 to maintain the 70% of field capacity. Three months after the irrigations with brackish water, soil samples were collected and analyzed for the chemical and biochemical parameters. Six composite soil samples (0–20 cm) for each treatment were taken from the agricultural farm of Mediterranean University of Reggio Calabria, Italy. The samples were brought to the laboratory on the same day of the collection, and kept in the refrigerator at 4 °C for up to 24 h until processing. Prior to the soil analysis, except for FDA hydrolysis and MBC, all the soil samples were air-dried, sieved (<2 mm), and visible roots were removed.

The second experiment (named case study 2) was performed in climatic chamber for 40 days, in plastic pots (10 cm diameter × 7 cm height). The soil (Haplic Kastanozem) used was taken from the agricultural farm of Mediterranean University of Reggio Calabria, Italy, in spring. Each pot was filled with 350 g of soil, in order to evaluate the effects of amendment with digestate at different concentrations (0, 25, 50, 75%) on soil chemical and biochemical properties. The digestate was obtained by a bio-gas energy plant with 998 kWel of installed power, supplied with animal manure (poultry, cow and sheep), milk serum, maize silage and in minor amount with olive waste and citrus pulp. During the experiment, the soil humidity was maintained at 70% of the field capacity in all treatments. The soils differently treated (6 replicates) were

air-dried and sieved (<2 mm) prior to the chemical analysis. Soil samples for the biochemical determination (microbial biomass and enzyme activities) were stored in the refrigerator at 4 °C for up to 24 h until processing.

The third experiment (named case study 3) was carried out in field, in the Calabrian Apennine Forest, Southern Italy, to investigate if artificial gaps and in particular the size of the gaps affected the soil chemical and biochemical parameters related to natural forest regeneration. The research area was in the Regional Park of Serre (Calabrian Apennines, Southern Italy at an elevation of 900–940 m. Soils, were classified as Haplic Phaeozem (IUSS, 2006). The natural forest is dominated by silver fir (*Abies alba* Mill) and beech (*Fagus sylvatica* L). In this forest, three small (185 m²) and three medium (410 m²) gaps were created by felling trees and removing boles. The treatments were named as follow: A = medium gaps; B = canopy cover sites; and C = small gaps. Gap sites were paired with an adjacent site under canopy cover. Soil were sampled 3 months after gap opening and were analyzed for chemical and biochemical properties. Soil samples were collected from 0 to 30 cm depth in each gap and in its adjacent forest canopy cover site. Each soil sample consisted of a mixture of six sub-samples taken at random. Prior to the soil analysis, except for soil moisture content, microbial biomass and FDA, all soil samples were air-dried and sieved (<2 mm).

The fourth experiment (named case study 4) was carried out in field. The study area was located in the Peripoli Mountain (San Lorenzo) of Aspromonte Mountains (Calabria, Southern Italy), 1270 m above sea level. The climate is predominantly Mediterranean, with dry hot summers and cold winters. The average seasonal precipitation are typically highest during the winter (1100) and autumn (1500) compared to spring (900) and summer (600). The soil were Haplic Phaeozem (IUSS, 2006) with a xeric soil regime moisture and a vegetal cover of *Pinus laricio* Poir et ssp. Calabrica. The effects of seasons (autumn, winter, spring and summer) were evaluated on soil chemical and biochemical parameters as described below. Soil profiles were carefully excavated, different (layers) horizons were thoroughly separated from the top to the bottom of the profile on the basis of morphological differences that could be perceived by the naked eye. Every 15 days, soil samples (1 kg) were taken from each horizon over a year (24 times in a year). The samples were brought to the laboratory on the same day of the collection, and kept in the refrigerator at 4 °C for up to 24 h until processing. Prior to the soil analysis, except for FDA hydrolysis and MBC, all the soil samples were air-dried, sieved (<2 mm), and visible roots were removed. Data presented are the means of three replicate determinations.

2.2. Soil chemical analysis

Organic C was estimated by the Walkley–Black procedure (Nelson and Sommers, 1982) and was converted to organic matter by multiplying the percentage of C by 1.72; total N was measured by the Kjeldahl method (Bremner and Mulvaney, 1982). Humic substances were extracted with 0.1 N NaOH (solid:liquid ratio 1:10); the suspension was shaken for 16 h at room temperature and centrifuged at 5000 rpm for 30 min; the extract was dialysed by Wisking tubes against distilled water to pH 6.0. Subsequently, the solution was filtered through a column of Amberlite IR 120H⁺. The fractionation of humic substances was carried out as follows: aliquots of extracts were acidified to pH 2.0 with dilute H₂SO₄; the humic acids precipitated and were removed by centrifugation, while the fulvic acids corresponded to the supernatants (Bettany et al., 1980). The C content of humic and fulvic acids was determined by dichromate oxidation (Nelson and Sommers, 1982). Phenols were extracted with distilled water as this is the most realistic extractant in allelopathic studies (Kaminsky and Muller,

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