



Original article

Soil microbiological properties affected by land use, management, and time since deforestations and crop establishment

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ABSTRACT

Deforestation is a common practice worldwide in order to gain agricultural land. In Filyria, Kilkis, Greece, three major deforestations took place in 1933, 1971 and 1980. Cultivation effects, referring to soil microbial properties are studied, in deforested fields, using the adjacent remaining oak forest as reference. The soils are cultivated with cotton (conventional tillage and irrigation, alternation with wheat every two years) or wheat (reduced tillage and no irrigation). The estimation and interpretation of the microbial properties were based on the analyses of soil organic carbon (SOC), microbial biomass carbon (MBC), the MBC/SOC ratio, potential soil respiration (SR), and the metabolic quotient (qCO_2). The forest ecosystem appears to provide better conditions for microbial growth and activity, having significantly greater SOC (31.8 g C kg^{-1} soil in forest versus 9.6 g C kg^{-1} soil in crop plots), MBC ($1080 \text{ mg C kg}^{-1}$ soil in forest versus 492 mg C kg^{-1} soil in crop plots) and SR ($4.78 \text{ mg C CO}_2 \text{ kg}^{-1}$ soil d^{-1} in forest versus $2.99 \text{ mg C CO}_2 \text{ kg}^{-1}$ soil d^{-1} in crop plots), and significantly lower qCO_2 rates than the crop plots although its organic matter quality results to a lower MBC/SOC ratio. The number of years since deforestation played a major role for most of the parameters analyzed, although after many years the equilibrium reached by the ecosystem appeared to moderate the differences. Disturbance through cultivation decreases soil quality. Reduced tillage without irrigation of the wheat crops leads to more suitable conditions for the microbial populations (as expressed by the microbiological properties) than conventional tillage with irrigation of the cotton crops.

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1. Introduction

Losses of soil organic carbon (SOC) often occur when converting forest to cultivated land, mainly due to lower inputs of organic matter, reduced physical protection of SOC as a result of tillage and changes in soil temperature and moisture regime which accelerate decomposition rates [29]. Soil tillage involves the physical disturbance of the upper soil layers, breaking down soil aggregates, thus influencing C stability in the soil [24]. Conservation tillage techniques seem to increase soil organic matter (SOM) in the upper layer, thus increasing the micro-aggregation and aggregate stability. These practices therefore could promote an enhancement of C sink at a global scale. Changes in SOC content occur slowly and do

not always provide adequate information of changes in soil quality that may occur [6]. It is therefore important to identify SOM fractions more sensitive to a change of land use or management which can be applied as early indicators of the dynamics of the soil C. Soil microbiological properties have been reported as a reliable tool in order to estimate early changes in the dynamics and distribution of soil microbial processes in different land use systems [16]. Microbial biomass carbon (MBC), potential soil respiration rate (SR), metabolic quotient (qCO_2 , ratio of respired C to biomass C), ratio of microbial biomass C to total organic C (MBC/SOC), are variables that have been suggested as indicators for assessing soil management effects on soil quality [1]. The microbial quotient (MBC/SOC) proved to be a reliable soil microbial parameter for describing changes in man-made ecosystems and more sensitive than its single components, MBC and SOC [2]. The metabolic quotient indicates the maintenance energy requirements [1] or generally stress by different factors [4].

Euro-Mediterranean regions are currently threatened by global changes [25]. Minetos and Polyzos [21] carried out a regional analysis of forest land use changes in Greece during the last

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decades. They observed that the prefecture of Kilikis, where the present study is located, showed a high rate of depletion of their forest site. There is a scarcity of studies addressing land use change dynamics and its effect on SOC and microbial properties in these semi-arid regions.

The aim of this paper is to determine the changes in terms of soil microbiological properties that resulted from three stages of deforestation which occurred 25, 34 and 72 years before the experiment, in Northern Greece, by measuring and assessing SOC, MBC, MBC/SOC ratio, SR and qCO_2 . The big amount of time that passed since the first deforestation, the availability of data from three different deforestation dates (1933, 1971 and 1980) and the remaining undisturbed forest, offer an interesting opportunity of studying these changes, along with assessing factors such as land use (forestry vs agriculture) and type of crop (wheat vs cotton). We have hypothesized that the effects on SOC and microbiological properties are similar in terms of amount of time since deforestation, but vary according to the subsequent type of crop installed.

2. Materials and methods

2.1. Study area

The study area is located in Filyria, prefecture of Kilikis, North Greece (40°54'11.69" N–40°53'41.9" N, and 22°28'47.97"E–22°29'37.10"E). The altitude ranges from 145 to 195 m. The climate is temperate Mediterranean with mean annual temperature of 15.0 °C, (absolute maximum 40.4 °C, absolute minimum –17.4 °C) and mean annual precipitation of 506 mm. Soils have developed from limestone, and are classified as Xeralfs according to Soil Taxonomy. Soil A horizons have clay loam textures, with percentages of sand, silt and clay of 30, 36, and 34%, respectively for the forest topsoil mineral horizon, and 26, 38, and 36%, respectively for the Ap horizon at agricultural land. Mean pH was between 7.7 and 8.0 and concentrations of $CaCO_3$ (method described below) were 1–7% at the forest sites and 1–14% at the cultivated plots. There are no differences between the plots of the study area in terms of basic soil properties. The vegetation is forest of *Quercus* species (especially *Quercus pubescens*). Socio-economic policies resulted to deforestation of the area at 3 stages (1933, 1971, 1980) leaving only 80 ha of natural forest undisturbed. These fields (2–9 ha mean extension) are cultivated with wheat, cotton and cherry trees. Mean slope of the area is 2%.

2.2. Management practices

The agricultural plots selected for this study are cultivated with cotton or wheat. For wheat, management practices include plowing up to 20–30 cm, fertilization with 20–10–0 (applying 300–400 kg ha⁻¹), and NH_4NO_3 (100–150 kg ha⁻¹). No irrigation is applied. Cotton management includes deep plowing up to 30–40 cm in September, followed by surface plowing in March. Fertilization with 11–15–15 or 12–12–17 + micronutrients (250–300 kg ha⁻¹) is applied once per period and NH_4NO_3 (100 kg ha⁻¹) twice per period. In terms of irrigation, every 10–15 days at summer 300–400 m³ of water per hectare is applied by sprinklers, meaning that the whole soil surface of the area is watered. Cotton fields are alterned with wheat every two years (two consecutive years of cotton crops, one year of wheat). Wheat fields are cultivated solely with wheat. Management practices are carried out with the same principles since 1933. History data were collected from the Greek Forestry Service, the National Agricultural Service and from interviews with farmers managing the land since 1946.

Practice of irrigation and tillage are the main differences among the two indicated crop types, hence for this study the cultivated

plots were divided to wheat crops and cotton crops. This division to subcategories relates to reduced tillage practices for the wheat plots (no deep plowing) that are not irrigated, and conventional tillage practices for the cotton plots that are irrigated.

2.3. Experimental design and sampling

Three sampling categories referring to deforestation year (1933, 1971, and 1980) in the cultivated fields and one category referring to the natural remaining forest were considered. For each deforestation year category, two crop subcategories were considered (wheat and cotton), and from each crop subcategory, 12 plots were sampled. In adjacent areas, twelve plots of remaining undisturbed forest (*Q. pubescens*) were selected. A composite sample was taken in each plot (72 cultivated plots and 12 forest plots). The composite sample was obtained by mixing fifteen random subsamples from the 0–15 cm mineral layer. Soil samples were collected at the end of November 2005, after harvest.

2.4. Total organic C analyses

Soil organic carbon was calculated from the total carbon measurement subtracting C from carbonates. Total carbon was measured by dry combustion on a LECO 2000 C/N/H analyzer. Soil total carbonates were determined by elimination with acid previously titrated with 0.5 M NaOH [8].

2.5. Microbiological properties

The fumigation–extraction method [30] was used in order to determine MBC. Microbial biomass C was calculated as a difference in C content in fumigated and non-fumigated sample (EC) using k_{EC} coefficient (MBC = EC: k_{EC}). The value $k_{EC} = 0.45$ was used to calculate microbial biomass C [32].

Potential soil respiration was determined in closed jars and under laboratory-controlled conditions following the Isermeyer method modified by Llorente et al. [17]. Soil samples were wetted to 75% of water holding capacity and incubated in 1 L jars at 29 °C for 3 days.

The metabolic quotient represents the potential soil respiration per unit microbial biomass, and was calculated as reported by Anderson and Domsch [3]. The microbial quotient (MBC/SOC) represented the fraction of MBC with respect to the SOC [3].

2.6. Statistical analysis

All data were subjected to ANOVA and when significant differences were detected ($p < 0.05$), Tukey's test was performed to allow separation of means. A factorial ANOVA with a control, forest, was carried out in order to determine the effect of deforestation, time since deforestation and type of crop. The Eq. (1) shows the factorial analysis of variance used for this model:

$$Y_{jkl(i)} = \mu + \alpha_i + \beta_{j(i)} + \gamma_{k(i)} + \beta\gamma_{jk(i)} + \epsilon_{ijl(i)} \quad (1)$$

Where, $Y_{jkl(i)}$: analyzed soil property, μ : overall mean, α_i : main effect of land use, with two levels (crop and forest), $\beta_{j(i)}$: main effect of time elapsed since deforestation with four levels (deforested in 1933, 1971 and 1980 for crops, and remaining forest), $\gamma_{k(i)}$: main effect of type of crop with three levels (wheat, cotton for crop plots, and forest), $\beta\gamma_{jk(i)}$: interaction between time and type of crop, and $\epsilon_{ijl(i)}$: experimental error. The last two factors (time and type of crop) and their interaction were nested into the land use factor. The normality, independence and homoscedasticity of the residuals were checked. Correlations between studied soil properties and the

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