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Research article

Critical range of soil organic carbon in southern Europe lands under desertification risk

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ABSTRACT

Soil quality is fundamental for ecosystem long term functionality, productivity and resilience to current climatic changes. Despite its importance, soil is lost and degraded at dramatic rates worldwide. In Europe, the Mediterranean areas are a hotspot for soil erosion and land degradation due to a combination of climatic conditions, soils, geomorphology and anthropic pressure. Soil organic carbon (SOC) is considered a key indicator of soil quality as it relates to other fundamental soil functions supporting crucial ecosystem services. In the present study, the functional relationships among SOC and other important soil properties were investigated in the topsoil of 38 sites under different land cover and management, distributed over three Mediterranean regions under strong desertification risk, with the final aim to define critical SOC ranges for fast loss of important soil functionalities. The study sites belonged to private and public landowners seeking to adopt sustainable land management practices to support ecosystem sustainability and productivity of their land. Data showed a very clear relationship between SOC concentrations and the other analyzed soil properties: total nitrogen, bulk density, cation exchange capacity, available water capacity, microbial biomass, C fractions associated to particulate organic matter and to the mineral soil component and indirectly with net N mineralization. Below 20 g SOC kg⁻¹, additional changes of SOC concentrations resulted in a steep variation of all the analyzed soil indicators, an order of magnitude higher than the changes occurring between 50 and 100 g SOC kg⁻¹ and 3–4 times the changes observed at 20–50 g SOC kg^{-1} . About half of the study sites showed average SOC concentration of the topsoil centimetres <20 g SOC kg⁻¹. For these areas the level of SOC might hence be considered critical and immediate and effective recovery management plans are needed to avoid complete land degradation in the next future.

1. Introduction

Healthy and productive soils are at the basis of ecosystem long term sustainability (Blum, 2005; CEC, 2006) and provide key ecosystem

services which support ecological, economic and social management goals (MEA, 2005; Comerford et al., 2013; UNCCD, 2016; Adhikari and Hartemink, 2016; Baer and Birgé, 2018). Maintaining and restoring soil quality is a fundamental task for land-based management frameworks

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and policies, such as the Common Agricultural Policy and the land degradation neutrality scheme proposed by UNCCD (UNCCD, 2013). Soil quality is defined through several soil chemical, physical and biological characteristics and their interactions (Adhikari and Hartemink, 2016). Among these, soil organic carbon (SOC) is recognized as one of the most relevant and universal indicators to assess soil quality (Bünemann et al., 2018) and land degradation (UNCCD, 2013; Lorenz et al., 2019). SOC is linked with the most fundamental soil properties and functions, such as soil structure, aeration, nutrient storage, water holding capacity, plant health and productivity, microbial biomass and activity, carbon sequestration (Wander, 2004; Comerford et al., 2013; Murphy, 2015; Adhikari and Hartemink, 2016). SOC, its fractions and dynamics have been widely used as adequate indicators to evaluate land management influence and to select the most appropriate agronomic practices to maintain and/or restore soil functionality (Duval et al., 2013). Soil loss and in particular SOC loss is one of the key environmental issues of this century, which, together with climate change, poses serious risks for ecosystem sustainability and food security of many world regions (Cherlet et al., 2018). Within the European context, high soil erosion rates (Panagos et al., 2015), high risk of land degradation (Zeng et al., 2021) and high to very high sensitivity to desertification (Mirzabaev et al., 2019) characterize several areas of the Mediterranean region (Stolte et al., 2016), as a consequence of the combination of climate factors (Russo et al., 2019), geomorphological features, soil type, land cover/management (Zdruli et al., 2004; Rodriguez-Martin et al., 2016; Calvo de Anta et al., 2020). On average, the Mediterranean region exhibits lower SOC values compared with other European regions, with many areas showing from very low ($\leq 1\%$) to low ($\leq 2\%$) SOC concentrations (Van-Camp et al., 2004; De Rodeghiero et al., 2011; de Brogniez et al., 2015). SOC values between 2% and 1% have been considered a major threshold below which potentially serious decline in soil quality might occur and primary productivity might be significantly reduced (Kemper and Koch, 1966; Greenland et al., 1975, Hamblin and Davies, 1977; Johnston 1986; Loveland and Webb, 2003; Oldfield et al., 2019). The SOC critical threshold, however, depends on soil properties, environmental conditions and land management practices (Körschens et al., 1998; Loveland and Webb, 2003). As suggested by Lal (2015), the restoration of SOC pool to threshold levels of at least 1.1%-1.5% by weight is crucial to reduce soil and environmental degradation risks.

To restore soil quality and improve SOC concentration above critical threshold levels it is important to establish the baseline or current soil status (Hessel et al., 2014). This allows to assess the degree of soil quality degradation thus providing useful information to support protection and restoration measures (van Lynde et al., 2016).

This work aimed at assessing the soil quality in agricultural lands and natural and semi-natural areas under desertification risk using SOC and other related soil parameters as quality indicators (DIS4ME, 2004; Álvaro-Fuentes et al., 2008; Huber et al., 2008; Kosmas et al., 2013; Ferreira et al., 2018; Oduor et al., 2018) to support landowners of the project LIFE Desert-Adapt (www.desert-adapt.it) in defining land management plans and climate adaptation measures to revert land degradation and support long term land sustainability (Francaviglia et al., 2018). The project works with landowners located in the Southern Mediterranean areas of Portugal, Spain and Italy which are experiencing land degradation, reduction of land productivity and increasing aridity. The specific objectives of the present work were: i) to quantify the current level of the investigated soil properties in order to evaluate the current soil quality status; ii) to assess the relationship between SOC and other physical, chemical and biological soil properties to estimate variations of soil properties in functions of SOC accumulation/degradation patterns in the analyzed sites; iii) to establish a critical value of SOC for the areas below which significant shifts of important soil characteristics might occur.

2. Materials and methods

2.1. Soil properties used as indicators of soil quality

SOC, soil total nitrogen, available water capacity, cation exchange capacity, microbial biomass, net N mineralization, bulk density, pH were selected as indicators of soil quality on the basis of their relevance for soil ecosystem services (Table S1; Adhikari and Hartemink, 2016) and indications provided by previous studies in vulnerable areas exposed to land desertification (DIS4ME, 2004; Huber et al., 2008; Kibblewhite et al., 2008; Zornoza et al., 2015; Constantini et al., 2016; Obalum et al., 2017; Sillero-Medina et al., 2020). Additionally, SOC fractions, particulate organic carbon (POC) and mineral associated organic carbon (MAOC), and soil nitrogen (TN) fractions, particulate organic nitrogen (PON) and mineral associated organic nitrogen (MAON) were also analyzed as they are considered more sensitive indicators of soil quality changes compared with variations of bulk soil C and N (Cotrufo et al., 2015; Lavallee et al., 2020). Soil C and N fractions have been used together with microbial biomass carbon (MBC), to assess land degradation (Obalum et al., 2017; Duval et al., 2018; Bünemann et al., 2018; Bongiorno et al., 2019) and to better detect the effect of grazing (Ferreira et al., 2018; Oduor et al., 2018) and tillage (Álvaro-Fuentes et al., 2008; Raiesi and Kabir, 2016) in drylands.

2.2. Study sites and soil sampling

The study analyzed soil quality of 38 sites (Table 1), part of the project LIFE Desert-Adapt, distributed in three southern European regions under strong desertification risk: Alentejo in Portugal, Extremadura in Spain and Sicily in Italy (Fig. 1). According to Koppen's climate classification, climate of these regions is classified as warm temperate, with hot and dry summers (Beck et al., 2018). The sites are characterized by very different geological substrates and soils (Table 1), the latter including Lithosols, Luvisol, Cambisol and Regosol (European Soil Bureau Network, 2005). The sites belongs to both public entities and private companies and include different land covers/uses, which were grouped in 4 main typologies, land areas where the dominant cover was represented by trees, pastures dominated by grass cover, shrublands and croplands (Table 1, Fig. 2). The first group included three coniferous afforestation stands (>50 years old) (CS) characterized by a close canopy cover, 2 in Italy (Pinus halepensis Mill) and 1 in Spain (Pinus pinaster Aiton) and three re-forested broad-leaved stands (BS), including one holm oak stand (IT), one eucalyptus stand (>30years; PT), and one agroforestry system with cork (Quercus suber L.) and holm oak (Quercus ilex subsp. rotundifolia) (PT). The latter ecosystem represents an example of traditional agroecosystems of the Iberian Peninsula called "montado" in Portugal and "dehesa" in Spain, characterized by a savanna-like physiognomy, with a canopy cover varying on average from 20 to 80 trees per hectare (Pinto-Correia and Mascarenhas, 1999; Pinto-Correia et al., 2011). These systems have developed through centuries of human alteration by animal grazing of the original broadleaved forest system, which coexists with a species-rich annual herbaceous vegetation and scattered shrubs (Bugalho et al., 2009). Eight additional sites in Portugal and Spain, dominated by herbaceous plants with few trees per hectare, represented a degraded condition of montado/dehesa systems, where the pressure of grazing was very high. Such sites were grouped as pastures-grasslands (PG) category along with three Italian pasture sites. Shrubland sites (S) included eight areas in Portugal and Spain, post-disturbance (deforestation, pasture abandonment, fires) stages of montado/dehesa systems, and two garrigue areas located in Lampedusa island (IT). Croplands (C) represented cultivated sites and included olive groves, prickly pear cultivations and herbaceous crops. All the cropland sites were tilled for seeding or weeding, with the exception of the prickly pear stands.

Field measurements and soil sampling were carried out in April–May 2018. The study sites covered a variable surface, from less than a hectare

Table 1

Location and main features of the 38 sites analyzed in this study from the three southern Europe areas under desertification risk. Temperature and precipitation are mean average for the period 1976–2005. Crop typologies refer to the cover crop in the year of sampling.

| Country region | Municipality | Geological substrate | Altitude (m a.s.l.) | Annual temperature (°C) mean (min –max) | Total annual precipitations (mm), aridity index | Site Code | Land cover | Coordinates (centroid of sampling area) |
|---------------------------|------------------------------|--------------------------|------------------------|---|---|----------------|---|--|
| Portugal (PT) Alentejo | Cabeça Gorda (PT1) | Schist, Quartzite | 150–180 | 16.1 (21.9–10.9) | 501, 0.38 | PT1-1 | Broad-leaved stand | 07°47′31''E, 37°54′29''N |
| | | | | | | PT1-2 | Docturo | 07° 47′ 48''E, 37° 54′ 29''N 07° 47′ 47''E |
| | Mertola (PT2) | | 105–190 | 16.2 (22.2–10.8) | 456, 0.35 | PT1-3 | grassland Cropland | 07°47'47''E, 37°54'29''N 07°39'59''E, |
| | | | | | | PT2-2 | (Lupinus) Pasture- | 37°48′33''N 07°39′58''E, |
| | | | | | | PT2-3 | grassland | 37°48′31''N 07°40′03''E, 27°48′27''N |
| | | | | | | PT2-4 | | 07°43′56''E, 37°43′44''N |
| | | | | | | PT2-5 | | 07°43′52''E, 37°43′44''N |
| | Comp (DT2) | | 100 165 | 16 1 (22 2 105) | 511 0 28 | PT2-6 | Cropland (Fodder) | 07°43′25''E, 37°43′37''N |
| | Serpa (P13) | | 120-105 | 10.1 (22.2–105) | 511, 0.38 | PT3-2 | grassland | 07°37°01°E, 37°48′29''N 07°36′58''E. |
| | | | | | | PT3-3 | Shrubland | 37°48′27''N 07°37′08''E, |
| Spain (SP) | Hoyos (SP1) | Biotite | 520–990 | 12.7 (17.8–8.2) | 1204, 1.13 | SP1-1 | Shrubland | 37°48′31''N 06°43′39''E, 40°10′35''N |
| Extrematura | | granitolus | | | | SP1-2 | | 40°10'33''E, 40°10'34''N |
| | Valverde del fresno (SP2) | | 410–485 | 11.7 (16.3–7.5) | 1427, 1.42 | SP2-1 | Conifer stand | 06°54′20''E, 40°12′38''N |
| | | | | | | SP2-2 SP2-3 | Shrubland | 06°54'49''E, 40°12'35''N 06°54'19''E. |
| | Malpartide de | Alluvial clayey | 255–285 | 14.3 (19.8–9.3) | 783, 0.67 | SP3-1 | Cropland | 40°12′04''N 06°38′18''E, |
| | Placençia (SP3) | deposits | | | | SP3-2 | (herbs) Shrubland | 40°00'17''N 06°38'26''E, 40°00'17''N |
| | | | | | | SP3-3 | | 40'00'17' N 06°37′50''E, 40°00′57''N |
| | | | | | | SP3-4 | Pasture- grassland | 06°37′58''E, 40°00′55''N |
| Italy (11) Sicily | Lampedusa e Linosa (IT1) | marls | 0–125 | 16.6 (17.8–15.3) | 352, 0.29 | IT1-1 IT1-2 | Conifer stand | 12°32′50′′E, 35°31′23''N 12°32′06''E. |
| | | | | | | IT1-3 | Shrubland | 35°31′22''N 12°35′54''E, |
| | | | | | | IT1-4 | Conifer stand | 35°31′03''N 12°32′26''E, 35°31′15''N |
| | Caltanissetta (IT2) | Clays and marly clays | 250–375 | 14.7 (20.4–9.8) | 484, 0.43 | IT2-1 | Cropland (olive) | 14°03′21''E, 37°25′41''N |
| | | | | | | IT2-2 | Pasture- grassland | 14°03′31''E, 37°25′37''N |
| | | | | | | IT2-3 | Cropland (Sulla, Avena) Cropland (olive | 14°03'40''E, 37°25'39''N 14°03'34''E. |
| | | | | | | IT2-5 | groove) Broad-leaved | 37° 25′ 43''N 14° 03′ 19''E, |
| | Enna (IT3) | sandy clays and | 410–755 | 13.5 (19.0–8.9) | 486, 0.42 | IT3-1 | stand Pasture- grassland | 37°25′48''N 14°13′57''E, 37°34′47''N |
| | | marry clay | | | | IT3-2 | grassianu | 14°13'45''E, 37°34'40''N |
| | | | | | | IT3-3 | Cropland (cheakpea) | 14°14′08''E, 37°34′58''N |
| | Caltagirone (IT4) | Yellow sands | 382-430 | 14.87 (20.6–9.7) | 561. 0.49 | IT3-4 IT4-1 | Cropland (olive groove) Cropland | 14°14'24''E, 37°34'29''N 14°33'24''F. |
| | | | | | | IT4-2 | (prickly pear) | 37°11′27''N |

(continued on next page)

Table 1 (continued)

| Country region | Municipality | Geological substrate | Altitude (m a.s.l.) | Annual temperature (°C) <i>mean (min</i> <i>–max)</i> | Total annual precipitations (mm), aridity index | Site Code | Land cover | Coordinates (centroid of sampling area) |
|----------------|--------------|-------------------------|------------------------|---|---|--------------|--|--|
| | | | | | | IT4-3 | Cropland (prickly pear) Cropland (Fruit) | 14°33'19''E, 37°11'28''N 14°33'15''E, 37°11'33''N |
| | | | | | | IT4-4 | Cropland (bamboo plantation) | 14°33′14''E, 37°11′34''N |



Fig. 1. Geographical localization of the studied sites.

to around 20 ha. In each site a minimum of five to a maximum of ten soil sampling points were selected (sampling points for each plot reported in Table S3). Starting from the center of each plot, along a North-South transect, sampling points were chosen with a distance between points of about 100 m. For each sampling point, composite samples of soil were collected using four soil cores (8 cm diameter, 10 cm depth) corresponding to the corners of a 25 m² square area centered on the sampling point. Soil was sampled at least 2 m from the tree trunks and 1 m from shrub canopy. Overall, 241 composite soil samples were collected. Taking into account the shallow depth of soils in several sites (<20 cm), the indicators chosen to analyze soil quality which included biological parameters, and considering the very low input of organic C to the soil in most of the sites, the top 10 cm of mineral soil were sampled, where the majority of microbial activity and SOC accumulation was expected (Jandl et al., 2014). Three undisturbed soil steel ring cores were also collected in the upper 10 cm of mineral soils to estimate soil bulk density (BD) in the central area of each plot. After sampling, soils were immediately shipped to the University of Campania Luigi Vanvitelli for laboratory analyses. After arrival, soil samples were divided in a block of samples which was stored fresh at 4 °C for biological analyses, which occurred within a week. Another block was air dried for subsequent chemical and physical analyses.

2.3. Soil analyses

Chemical and physical analyses were done on air dried and sieved soil (2 mm mesh) (Soil Survey Staff, 2014a). Soil pH was measured by potentiometric method in aqueous solution (soil/water 1:2.5). Cation exchange capacity (CEC) was determined by saturation with BaCl₂ at pH 8.2 (ISO 11260:1994). Total soil organic carbon (SOC) was determined by wet sulfochromic oxidation followed by FeSO₄ titration (ISO 14235:1998). Soil total nitrogen (TN) was determined on pulverized dry samples by flash dry combustion with a CN-soil elemental analyzer (Thermo FLASH1200). Calibration was performed by BBOT (72.53% C, 6.51% N) (AOAC Official Method 972.43).

To estimate bulk density BD, soil in steel rings was oven-dried at $105 \,^{\circ}$ C for 48–72 h. When samples were missing (PT3), BD values were calculated by pedo-transfer functions (Hannam et al., 2009).

The available water capacity (AWC), which represents the capacity of the soil to store water available for plants, is the difference between the soil volumetric water content at field capacity (θ fc) and at permanent wilting point (θ pwp) (Rawls et al., 2003), ranging usually from -0.33 to -15 bars, respectively (USDA-NRCS, 2013). To calculate AWC, soil texture and SOM were used to estimate θ fc and θ pwp according to Rawls et al. (2003). Sand, silt and clay fractions were separated by the pipette method and wet sieving following pre-treatment with H₂O₂ and sodium hexametaphosphate.



Fig. 2. a) Pine afforestation site (IT1-2); b) agroforestry system with cork oak (PT1-1); pastures in (c) dry (SP3-4) and wet (d) season (SP3-4); e) garrigue area (IT1-1) in dry season; f) *Cistus ladanifer* shrubland (SP2-3).

Soil C and N fractions were measured on a subset of samples from sites where the LIFE Desert-Adapt project planned a change of land management, in order to be able to determine in 3 years' time any eventual modifications in the C and N distributions in the soil fractions. The physical fractionation method was used to determine the particulate organic carbon (POC) content, associated with the sand fraction (2000–53 µm), following procedures by Cambardella and Elliott (1992) reported by the Soil Survey Staff (2014b). 10-g of sieved (<2.0 mm) air-dried soil were dispersed with 30 ml of 0.5% sodium hexametaphosphate solution and shaken for 15 h (overnight) at 200 oscillations min^{-1} at room temperature. The slurry was passed through a 53 μm sieve using a jet distilled water. The material retained on the sieve was dried at 45 °C for 48 h in a forced air oven. The oven dried material was ground and analyzed for organic C by the wet oxidation method while TN was determined by dry combustion. The mineral associated organic C (MAOC) and N (MAON) contents in the soil fine fraction (<53 µm) were calculated as the difference between SOC and POC and TN and PON, respectively (Franzluebbers and Stuedemann, 2002; Duval et al., 2013; Soil Survey Staff, 2014b). The fraction (f) was calculated as the amount of C or N in sand (POC and PON) and fine fractions (MAOC and MAON) divided by the total SOC or TN concentration, respectively, measured on the same bulk soil. Net N mineralization was determined by incubating fresh soil samples (20 g) aerobically (55% of soil water holding capacity) in the dark at 25 °C and extracting inorganic nitrogen $(NH_4^+ \text{ and } NO_3^-)$ at t₀ and after 14 days in order to calculate the mineralization rates using a time vs. concentration curve (Kandeler, 1995). Soil NH⁺ and NO⁻ concentrations were determined on K₂SO₄ solution filtered extracts (1:5 soil extract v/v) by potentiometric analysis using ion-selective electrodes (Castaldi et al., 2011). Microbial biomass carbon (MBC) was measured by the fumigation-extraction method (Vance et al., 1987) and the organic C content was determined on filtered extracts (Whatman 42 filters) by humid digestion with 66 mM K₂Cr₂O₇ at 160 °C.

2.4. Statistical analysis

To compare the analyzed soil properties under different land covers, normality test (Shapiro-Wilk) and equal variance test (Levene) were first performed on the data set of each analyzed soil indicator (Table S2). When the two required conditions, i.e. normality and homoscedasticity, were respected a one-way ANOVA was run with an all pairwise comparison using the "Student–Newman–Keuls test" at p < 0.05. When the data were not normally distributed and/or did not exhibit the same variance an ANOVA on ranks (Kruskal–Wallis test) was run and Dunn's method was selected for all pairwise comparison. Simple linear regression, multiple linear regression and non-linear regression analyses were performed to find the relationship between independent and dependent variables. Statistical analyses were carried out using the software programs SigmaPlot 12.5 (Sigma Stat, Jandel Scientific) and Statistica 7.1 (StatSoft Inc Development, 2006).

3. Results

3.1. Indicators of soil quality in the study sites

Average SOC concentration in the top 10 cm of soil of the 38 study sites varied from a minimum of 6.5 ± 1.2 g C kg⁻¹ to a maximum of 184.4 \pm 26.2 g C kg⁻¹ (mean \pm SD) and decreased in the order coniferous tree stands > shrublands > broad-leaved tree stands > pastures > croplands (Table 2, Table S3). The intra group SOC variability was higher than inter group variability (Table S3) and no significant differences in SOC concentration were observed among soils of different groups, with the exception of conifers stands vs soil of pastures and croplands.

Average soil TN content decreased in the order coniferous tree stands > broad-leaved tree stands> shrubland > pastures > croplands (Table 2), with the highest value measured in the IT1-4 coniferous stand, $8.8 \pm 0.6 \text{ g N kg}^{-1}$, and the lowest value, $0.7 \pm 0.1 \text{ g N kg}^{-1}$, measured in the SP2-3 shrubland site (Table S3). No significant differences of TN content were observed among groups, except for coniferous stands where TN values were significantly higher than in all the other sites (Table 2).

Soil C/N ratio varied from 24.6 \pm 4.2, under coniferous cover, to 4.6 \pm 1.1 in pasture/grassland systems (Table S3); no significant differences of soil C/N were observed among groups (Table 2).

Soil under coniferous trees (CS) were characterized by the lowest bulk density (0.64 \pm 0.01 g cm $^{-3})$ and the highest CEC (33.1 \pm 7.1

Table 2

Mean, median, standard deviations (SD) and range (min-max) values of soil chemical and physical properties measured in the top 10 cm of soil of the study sites grouped by dominant land cover.

| LAND COVER | | BD g·cm ⁻³ | SOC $g \cdot kg^{-1}$ | TN $g \cdot kg^{-1}$ | C/N | CEC meq $\cdot 100 g^{-1}$ | AWC % |
|--------------------------|--------|-----------------------|-----------------------|----------------------|-----------|----------------------------|-------------|
| Coniferous tree Stands | Mean | 0.64 | 143.6 | 7.3a | 21.5 | 33.1 | 21.2 |
| | Median | 0.64a | 133.1a | 7.0 | 25.5 | 33.1a | 21.1a |
| | SD | 0.01 | 36.7 | 1.4 | 7.7 | 7.1 | 2.1 |
| | Range | 0.64-0.65 | 113.4-184.4 | 6.0-8.8 | 12.6-26.4 | 26.0-40.2 | 19.2-23.3 |
| Broad-leaved tree Stands | Mean | 1.24 | 25.7 | 3.8b | 7.0 | 14.9 | 14.8 |
| | Median | 1.26 ab | 18.8ac | 2.8 | 6.6 | 11.2ac | 14.7 ab |
| | SD | 0.18 | 14.8 | 2.3 | 1.9 | 7.1 | 1.1 |
| | Range | 1.05-1.40 | 15.6-42.6 | 2.1-6.3 | 5.5–9.1 | 10.5-23.0 | 13.7-15.9 |
| Shrublands | Mean | 1.18 | 30.6 | 2.9b | 12.5 | 14.0 | 15.7 |
| | Median | 1.21 ab | 25.1ac | 3.2 | 12.5 | 12.6ac | 15.6a |
| | SD | 0.21 | 14.5 | 1.2 | 6.9 | 3.9 | 1.7 |
| | Range | 0.73-1.39 | 13.5-60.5 | 0.7-4.3 | 6.5-25.3 | 7.9–21.6 | 13.8 - 18.1 |
| Pastures/Grasslands | Mean | 1.32 | 17.7 | 2.5b | 7.7 | 11.5 | 14.1 |
| | Median | 1.34b | 14.7bc | 2.4 | 6.8 | 10.7bc | 14.2 ab |
| | SD | 0.10 | 7.2 | 1.1 | 2.1 | 6.2 | 1.5 |
| | Range | 1.05-1.45 | 10.9-32.3 | 1.2-4.3 | 5.0-11.5 | 5.3-23.3 | 11.6–17.4 |
| Croplands | Mean | 1.20 | 15.5 | 2.1b | 7.6 | 12.1 | 13.0 |
| | Median | 1.16 ab | 11.7bc | 1.7 | 7.2 | 9.5bc | 13.7 b |
| | SD | 0.22 | 10.0 | 1.1 | 2.7 | 7.9 | 3.0 |
| | Range | 0.79–1.51 | 6.5–33.9 | 0.9–4.3 | 4.6-12.6 | 3.4-26.0 | 6.3–19.6 |

BD = bulk density; SOC = soil organic carbon content; TN = total nitrogen; C/N = soil organic carbon: total nitrogen ratio; CEC = cation exchange capacity; AWC = available water content. Different lower-case letters show statistically significant differences at p < 0.05 among data in the same column. Choice of parametric or not parametric ANOVA based on data reported in Table S2.

meq·100 g⁻¹) (Table 2, Table S3). In the other sites, BD ranged from a 0.73–1.51 g cm⁻³ (Table S3), with the highest average BD values estimated in pasture systems, while the lowest average CEC values were measured in the cropland sites (Table S3). For both variables no significant differences were observed among different groups.

No significant differences of average soil net N mineralization rates and soil microbial biomass carbon concentrations were observed among different groups, except for coniferous stands, where both indicators were from 3 to 4 times higher than in all the other sites (Table 3, Table S4). A great variability was observed within each land cover group (Table S4) for both parameters. Considering the 38 sites separately, net N mineralization rates varied from close to zero to around 10 mg N kg⁻¹d⁻¹, whereas microbial biomass C ranged from 112 to 1325 mg C kg⁻¹.

Soils under coniferous tree cover were characterized by the lowest proportion of the most stable fraction *f* MAOC (<0.5) (Table S5). In all the other sites, *f* MAOC was the prevailing form of C accrual with an average *f* MAOC value of 0.70 (Table S5). However, in terms of stored C, average MAOC concentration in coniferous tree stands was 71.4 g C kg⁻¹, compared with 12.9 g C kg⁻¹ in broad-leaved tree stands, 17.8 g C kg⁻¹ in shrublands, 12.8 g C kg⁻¹ in pastures and 8.6 g C kg⁻¹ in croplands. Croplands sites showed the highest ratio variability between the two fractions, for both C and N. In the majority of sites, N accumulated in soil organic matter mainly in form of *f* MAON (all sites *f*)

Table 3

Mean (±1 SD) and median values of soil net N mineralization rate (mg N kg⁻¹d⁻¹) and soil microbial biomass carbon (MBC - mg C kg⁻¹) measured in the study sites grouped by dominating land cover.

| | Net N mineralization rate Mean \pm SD (Median) | MBC Mean \pm SD (Median) |
|-----------------------------|--|------------------------------------|
| Coniferous tree stands | 7.1 ± 2.9 (7.1) | 900.2 ± 371.8 (<i>1007.8</i>) |
| Broad-leaved tree stands | 1.4 ± 0.5 (1.4) | 284.0 ± 104.9 (225.6) |
| Shrublands | 1.7 ± 1.9 (0.8) | 360.5 ± 180.4 (281.8) |
| Pastures/ Grasslands | 1.5 ± 1.3 (1.4) | 306.8 ± 221.2 (231.3) |
| Croplands | 1.7 ± 1.4 (1.4) | 390.3 ± 312.1 (282.0) |

MAON average 0.64). The cropland sites, which used rotations with leguminous crops, showed the highest values of f MAON (0.73–0.90) among cropland sites, but on average the balance between the two soil N fractions was even in cropland sites (mean f MAON 0.46). Excluding the conifer stands, the only significant difference of C or N accrued in the two fractions was observed between shrublands and croplands.

3.2. Relationships between SOC and the other soil indicators

Soil total N (TN) increased steeply for increasing SOC concentrations between 0 and 20 g C kg⁻¹ soil. Above 20 g SOC kg⁻¹ soil, the accumulation of N in soil per unit of SOC gradually levelled off (Fig. 3A, Table S6). The fitting line representing the relationship between the two variables was a "rise to the max" curve where, however, the plateau was reached at very high SOC concentrations.

Excluding data of coniferous tree stands, SOC content was negatively correlated to the soil bulk density (BD) (Fig. 3B, Table S6). Between 5 and 60 g SOC kg⁻¹, BD varied between 1.5 and 0.7 g cm⁻³, while in all the three coniferous tree stands the BD remained in the range of 0.6 g cm⁻³.

Cation exchange capacity (CEC) increased logarithmically for increasing SOC concentration between 5 and 60 g SOC kg⁻¹ (Fig. 3C, Table S6), with the steepest increase of CEC per unit of SOC observed between 0 and 20 g SOC kg⁻¹. Soil CEC values could also be predicted by a linear combination of clay and SOC content (CEC (meq. g⁻¹) = 4.03 + (0.26 * clay %) + (1.38 * SOC %), R² = 0.61 P < 0.001), although the multi-linear equation showed a more relevant role of SOC than clay in determining CEC variability.

The variation of AWC% in function of SOC content followed a logarithmic relationship (Fig. 3D, Table S6), with the biggest AWC variations per unit of SOC change estimated between 0 and 20 g C kg⁻¹.

Soil net N mineralization was significantly correlated with soil TN but not with SOC. The rates of N mineralization significantly differed in acid and neutral/alkaline soils, the latter having net N mineralization rates more than twice the rates observed in the acid soils (Fig. 4A Table S6).

Soil microbial biomass showed significant differences in acid vs. not acid soils (Fig. 4B, Table S6), but only for the latter a significant regression was observed. In the range of $0-50 \text{ g SOC kg}^{-1}$, the increase of soil microbial biomass per unit of SOC was three times higher in



Fig. 3. Values of mean SOC content plotted vs total nitrogen (TN) content (A), bulk density (BD) (B), cation exchange capacity (CEC) (C) and available water capacity % (AWC) (D). Dotted lines represent the best fit of mean data from the 38 study sites, continuous lines show the best fit of data without values from conifer stands (CS). Equations and statistical parameters are reported in Table S6.

neutral-basic soils compared with acid soils, exception made for the soils under coniferous tree cover, where the extremely high values of SOC corresponded to very high values of microbial biomass (Table S4).

In order to better represent the relationship between the soil fractions of C and N vs. SOC and TN content, the equations derived from the best fit of experimental data reported in Table S6 were used to calculate a continuous range of f MAOC, f POC, f MAON and f PON values in function of the observed range of mean values of SOC and TN.

Data reported in Fig. 5 A, B, show that the relative amount of soil C and N stored as particulate or as mineral associated fractions, varied with increasing content of SOC and soil TN, respectively. C stored as f POC increased exponentially with increasing SOC, whereas f MAOC saturated above 100 g SOC kg⁻¹. Soil C was stored more efficiently as f MAOC for values of SOC <120 g C kg⁻¹. Above this value, SOC accumulated mainly in form of POC. The distribution of soil N in the two fractions in functions of the TN concentration showed several differences compared with C trends (Fig. 5B). Both fractions increased their N content linearly with increasing TN up to 2.5 g N kg⁻¹ dry soil, but N accumulated as MAON was twice the amount accrued as PON and MAON remained the major form of soil stored N up to 10 g N kg^{-1} of TN. Above 10 g TN kg⁻¹, N accumulated mostly in form of PON, while MAON levelled off (Fig. 5B). Bulk soil C/N was found to increase exponentially for increasing C/N of the mineral associated organic matter component (Table S6). No significant correlation was found between the concentration of MAOC or MAON and the total amount of silt and clay or the soil C/N ratio, alone or in combination with the other soil variables.

mineralization rates and MAON concentration (Table S6). As no significant difference was found between acid and not acid soils, all the data were plotted together. On the contrary no significant relationship was found between N mineralization rates and PON concentrations, even when acid and not acid soils were analyzed separately. Overall, soil TN seemed a better soil property to predict N mineralization rates than PON or MAON. No significant correlation was found between microbial biomass content and C content of any of the two SOC fractions.

3.3. Use of soil indicators to define a critical SOC range for soil quality in the studied sites

To better understand the impact that SOC concentration reduction might have on soil quality, we calculated the variation (%) of each soil property, used as indicator of soil quality, in function of a SOC variation of 1% (10 g C kg⁻¹). Estimates were averaged for three different SOC ranges 0–20, 20–50, 50–100 g C kg⁻¹. Results show that between 0 and 20 g C kg⁻¹ all the analyzed soil properties varied between 35 and 66%, excluding the BD which showed relative low variations overall (Table 4). Such changes were about an order of magnitude higher than the changes occurring in the SOC range of 100–50 g C kg⁻¹, and 3–4 times the changes observed at 50-20 g C kg⁻¹ (Table 4). The highest variation per 1% SOC change, was observed at very low SOC concentrations (<2%) for the soil quality indicators more dynamically related to the input of new biomass, *i.e.* soil microbial biomass, N mineralization, MAOC and MAON.

A significant exponential relationship was found between net N



Fig. 4. Net N mineralization rate plotted vs soil total nitrogen (TN) content A) and soil microbial biomass carbon (MBC) content plotted vs soil organic carbon (SOC) content (B), for sites with acid soil pH (black symbols) and sites with neutral to basic soil pH (white symbols); equations of the fitting curves are reported in Table S6.

4. Discussion

4.1. Range of soil C and N content in bulk soil and its fractions in the analyzed sites

With the exclusion of the three coniferous tree stands, characterized by high levels of C in the topsoil (Johnson and Curtis, 2001; Krishna and Mohan, 2017), soils sampled in the other 35 sites had medium to low concentrations of SOC (Table S3), in accord with previous observations from several Mediterranean areas of Europe (Van-Camp et al., 2004; Zdruli et al., 2004; De Rodeghiero et al., 2011; Tóth et al., 2013; De Brogniez et al., 2015; Cotrufo et al., 2019). Fourteen sites had average SOC concentrations between 20 and 60 g C kg^{-1} and twenty-one had SOC concentrations below 20 g C kg⁻¹. As a general trend, shrublands and broad-leaved tree stands had higher SOC concentrations compared to pastures and croplands, as both systems have on average higher incorporation rates of plant residues and are considered important C sinks in Mediterranean areas (Rodriguez et al., 2009; Durán Zuazo et al., 2014; Ruiz-Peinado et al., 2017). The estimated values of SOC concentration were comparable to data reported for European Mediterranean semiarid soils (0–15/20 cm) by Tóth et al. (2013), for shrublands of 23 \pm 16 g C kg^{-1}, 17 \pm 12 g C kg^{-1}, pastures and 12.5 \pm 7.5 g C kg^{-1} croplands. Pastures showed on average low SOC value (pasture mean value 17.7 \pm 7.2 g SOC kg⁻¹) but also high variability among different sites, with values ranging from 10 to 32 g SOC kg⁻¹. This variability



Fig. 5. Simulated distribution of soil C and N content in particulate (POC and PON) and mineral associated (MAOC and MAON) fractions in function of the total soil organic carbon (SOC)(A) and nitrogen (TN) (B) content, based on empirical relationships calculated from the experimental data of the analyzed sites.

Table 4

Percentage reduction of each specific soil properties, chosen as indicator of soil quality, for a SOC reduction of 10 g of SOC kg⁻¹, calculated over three SOC intervals. Estimates are based on the relationships reported in Table S6.

| Soil properties used as indicators of soil quality | Variation (%) of soil quality indicators for a SOC change of 10 g of SOC kg^{-1} (1% SOC) | | | | |
|--|---|---|--------------------------------|--|--|
| | 0-20 SOC kg ⁻¹ | $\begin{array}{c} \text{20-50 SOC} \\ \text{kg}^{-1} \end{array}$ | 50-100 SOC kg ⁻¹ | | |
| Total soil N content | 48.0 | 15.7 | 5.3 | | |
| Bulk density | 10.8 | 8.4 | 5.2 | | |
| Cation exchange capacity | 35.3 | 10.6 | 3.9 | | |
| Available water capacity (%) | 48.3 | 5.1 | 1.2 | | |
| MAOC concentration ^a | 66.6 | 18.6 | 8.0 | | |
| MAON concentration ^{a,b} | 57.5 | 14.0 | 3.5 | | |
| Net N min rate (acid soils) ^{a,c} | 56.1 | 17.3 | 5.7 | | |
| Net N min (not acid soils) ^{a,c} | 44.4 | 15.4 | 5.2 | | |
| Soil microbial biomass (not acid soils) | 63.5 | 12.9 | 4.5 | | |

^a Range 0.5-2% SOC.

^b Estimated indirectly with a 2 step analysis TN vs SOC and MAON vs TN and.

^c Estimated indirectly with a 2 step analysis TN vs SOC and Net N min vs TN.

most probably reflected differences in factors affecting soil organic matter formation, losses and balance like, soil texture, geomorphology, slope, organic inputs (Zdruli et al., 2004) but also the different levels of grazing intensity, which could influence grass cover richness and productivity and hence quantity and quality of C inputs to the soil (Steinbeiss et al., 2008; Simón et al., 2013; Francaviglia et al., 2017). SOC variations among cropland sites were even wider (6–34 g C kg⁻¹). As expected, croplands showed the lowest levels of SOC concentration among the analyzed sites, due to the significant level of disturbance by tillage and management practices and the reduced inputs of fresh organic C.

N accrual in the soils of the studied sites increased proportionally to increasing SOC content up to 20 g SOC kg⁻¹, thereafter the relative increase of N per unit of SOC became progressively smaller, levelling off at concentrations measured only in few of the analyzed sites. Overall, values of TN were in the lower end of the range of soil N data reported in Lucas dataset for European soils (Tóth et al., 2013), with the exception of soils under coniferous cover. Excluding the latter, soil C/N ratio was below 20 in 33 out of 35 sites. A C/N < 20 is generally considered a condition favorable to soil organic matter mineralization (Robertson and Groffman, 2007). In croplands and pastures, fertilization, leguminous rotations and animal dejections might have contributed to the observed low C/N ratio (Guimarães et al., 2013; Francaviglia et al., 2017). However, soils within SOC range of $0-50 \text{ g C kg}^{-1}$ also showed a tendency to accumulate more soil organic matter in the more stable fraction associated to the mineral component, considered more N rich than particulate organic matter (Lavallee et al., 2020). Below 20 g SOC kg^{-1} , C stored as MAOC was about 3 times the amount of C stored as POC. In the same SOC range, the ratio MAON/PON was around 2. From 20 to 80 g SOC kg⁻¹ the ratio MAON/PON increased, whereas the ratio MAOC/POC decreased progressively to arrive to a value of 1 at 130 g SOC kg⁻¹, which was generally higher than the SOC content in most of the analyzed sites. A prevalence of MAOC over POC has been previously observed in soils with relatively low C content, like semi-arid zones of Mediterranean areas, seasonally dry and arid ecosystems under different land management (Cambardella and Elliott, 1992; Álvaro-Fuentes et al., 2008; Purakayastha et al., 2008; Awale et al., 2017; Cappai et al., 2017; Ferreira et al., 2018; Cotrufo et al., 2019). In terms of ecosystem services provision, the prevalence of MAOC and MOAN as form of C and N storage in such poor soils entails several benefits for the whole ecosystems. Low molecular weight compounds present in the mineral associated organic fraction are generally more nutrient dense (Tipping et al., 2016) and easier to process, than the POC and PON fractions, having lower activation energies of decomposition (Williams et al., 2018) and a simpler enzymatic and catabolic pathway of assimilation for microbes and plants once the link with the mineral component has been destabilized (Kleber et al., 2011, 2015). However, MAOC and MOAN are highly persistent in soil system with minimal disturbance, with a residence time from decades to centuries as they are protected from decomposition through association with soil minerals via chemical bonds and/or occlusion within micropores or small aggregates (<50-63 μm) (von Lützow et al., 2007; Kogel-Knabner et al., 2008; Kleber et al., 2015). For this reason, the prevalence of MAOC and MOAN as a form of C and N storage in soils of arid lands can be considered very important for C sequestration and long term N storage in the system, but might represent a less readily and useful source for plant primary productivity (Jilling et al., 2018; Lavelle et al., 2020).

4.2. Critical levels of SOC and relationships with other indicators of soil quality

A significant relationship was found between values of SOC and other soil properties chosen as indicators of soil quality. The empirical relationships allowed to estimate the degree of change that such properties might have per unit of SOC change over the observed range of SOC values. Not being such relationships linear (except BD vs SOC for SOC

<50 g C kg⁻¹), the effect of SOC variation on the other soil properties changed dramatically with the level of SOC (Table 4). At SOC <20 g C kg⁻¹ variations of soil properties in the range of 35 up to 66% were estimated, for 1% SOC change (10 g C kg-1). A serious decline in soil quality might hence occur for relative small variations of SOC content, when concentrations of SOC are quite low, which for the specific case of our sites would be below 20 g C kg⁻¹. Several studies have focused on the identification of critical SOC ranges for soil quality and plant productivity, with a general agreement that critical thresholds depend on site specific factors, soil type, climatic conditions, land use (Körschens et al., 1998; Loveland and Webb, 2003) and their definition requires a local scale approach preferable to a single European SOC threshold (Van-Camp et al., 2004; Huber et al., 2008). Yet, to establish critical minimum SOC levels which pose serious risks for the quality of the overall system, is strongly advised to guide farmers' land management planning (Sparling and Schipper, 2002). Minimum, critical levels of soil organic matter between 5 and 25 g C kg⁻¹ for crop production have been reported (Hijbeek et al., 2017). Spink et al. (2010) indicated 25 g SOC kg⁻¹ as the limit for poor soil quality. A critical range of SOC between 10 and 20 g SOC kg⁻¹ was previously reported in the review by Loveland and Webb (2003).

Among the analyzed properties in the studied sites, the indicators more closely related to the dynamics of soil organic matter, soil microbial biomass, N mineralization, MAOC and MAON, were those more rapidly changing for a variation of 1% SOC, below 20 g SOC kg $^{-1}$. The soil microbial component is critical for decomposition processes involved in the formation of soil organic matter and accrual of SOC (Paul, 2007; Prescott, 2010), in particular for the formation of the most stable fraction of soil C and N, MAOC and MAON (Cotrufo et al., 2013). A growing body of evidence suggests that microbial products, together with low molecular weight compounds leached from plant litter or produced by exo-enzyme depolymerization of plant litter, are the main component of MAOC and MOAN (Kogel-Knabner et al., 2008; Knicker, 2011; Cotrufo et al. 2013, 2019; Lehmann and Kleber, 2015). On the contrary, no specific dominant role is played by microorganisms in POC and PON formation (Lavalle et al., 2020). Low productivity characterizing many semi-arid areas of the Mediterranean basin, in combination with biomass removal by intensive grazing and crop removal by farming activities, might result in low organic matter inputs for the detritus chain but also less favorable conditions for microbial growth and activity, with direct effects on the formation of the most stable forms of soil organic matter (Cotrufo et al., 2019).

Soils which are at a critical level of SOC and soil quality require urgent restoration actions (Romanya and Rovira, 2011). Lal (2015) proposed three basic strategies to recover soil quality and health in degraded areas: (i) to minimize soil losses; (ii) to create a positive soil C budget and (iii) to reinforce water and elemental cycling. Such principles have been adopted following the baseline analysis in the Desert-adapt project sites, in particular in the areas more at risk, to slow down soil degradation, ecosystem services and economic losses (Sanz et al 2017) in accordance with local landowners, integrating their knowledge of local environmental and agronomic conditions. To make some example of the proposed measures, water conservation and erosion control practices were applied at different levels, from swales to key lines, to limit soil losses and increase water retention and plant productivity (Pereira, 2005). Mulching was successfully applied to reduce soil and water losses using local plant pruning materials, improving soil water storage, adding C sources to the soil, and protecting new seedling from excess evapotranspiration during drought periods (Prosdocimi et al., 2016). Management technologies, such as holistic planned grazing (Savory and Butterfield, 1999), were introduced in the pastures in Portugal to promote grassland recovery and increase grass productivity, to strengthen below ground biomass and to increase the overall C input to the soil (Follett and Schuman, 2005). To reduce soil disturbance and to increase plant cover in Spanish areas, where shrubs were usually mechanically eradicated to prevent fire risk, manual shrubs biomass

removal was applied and the plant material recovered was in part used to extract aromatic oils for the local markets. Such initiative provided several benefits reducing fire risk, soil disturbance and erosion, maintaining significant levels of C inputs to the soil and creating net economic benefits from a waste product.

5. Conclusions

The study confirmed that SOC is a suitable indicator of soil quality as it is functionally linked to other soil properties relevant for soil quality and soil ecosystem services. The empirical relationships between SOM and the other soil indicators allowed to define a critical level of SOC (<20 g SOC kg⁻¹) at which significant shifts in soil quality might occur with small changes in SOC concentration. About half of the studies sites were below such critical threshold. Management plans for the recovery of SOC, should be a priority in lands characterized by such low levels of SOC and exposed to increasing aridity conditions, such as those analyzed in this study. Ideally, sustainable measures should be applied well before significant losses of SOC occur, *i.e.* when SOC is still in the range of 20–50 g C kg⁻¹.

Author contributions

Eleonora Grilli: Methods, Investigation, Data Curation, Writing -Original Draft, Project administration Sílvia C. P. Carvalho: Resources, Tommaso Chiti: Resources, Writing - Review & Editing Elio Coppola: Writing - Review & Editing Rosaria D'Ascoli: Investigation, Data Curation, Tommaso La Mantia: Investigation, Writing - Review & Editing Rossana Marzaioli: Investigation, Data Curation, Micòl Mastrocicco: Methods, Investigation, Data Curation, Fernando Pulido: Resources, Writing - Review & Editing Flora Angela Rutigliano: investigation, Writing - Review & Editing Paola Quatrini: resources Writing - Review & Editing, Simona Castaldi: Conceptualization, Formal analysis, Writing -Original Draft, Supervision, Project administration, Funding acquisition

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

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