



Effects of *Quercus rubra* L. on soil properties and humus forms in 50-year-old and 80-year-old forest stands of Lombardy plain

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Abstract

• **Key message** Besides the well-known effects on the native plant community, red oak may also impact the soil; the effects of afforestation with red oak involve both organic layers and mineral soil, resulting in changes in organic carbon quantity and quality and in soil acidification.

• **Context** Many alien species have become widespread in Europe; among these, red oak is a common invader of temperate forests.

• **Aims** The effects of substitution of natural mixed forest by red oak forest on humus forms and soil properties were investigated in two paired plots: a 50-year-old (Bosco Vacaressino) and 80-year-old (Bosco Ginestre) forest stand.

• **Methods** Soil sampling was performed from 3 layers at 40 and 49 points in Bosco Vacaressino and Bosco Ginestre respectively to determine humus forms, soil pH, organic carbon stock, carbon-nitrogen ratio (C:N), available phosphorus, and texture.

• **Results** Red oak resulted in a shift from Mull to Moder humus forms; soil acidification, higher C:N ratio, and soil organic carbon stock were observed compared with mixed forests.

• **Conclusion** The major changes were reflected in a change toward less active humus forms; the effects of vegetation conversions were also visible in mineral layers; many of the modifications were more evident with increasing stand age.

Keywords Alien species · Red oak · Forest Conversion · Humus form · Soil spatial variability · Mixed model

1 Introduction

Alien invasive plant species have significant effects on the structure and function of ecosystems (Ehrenfeld 2003; Hejda et al. 2009). Invasive species can threaten biological diversity in particular by reducing genetic variation through the endangering of endemic species and by altering habitat and ecosystem functioning. Because of human intervention, many alien species have become widespread in Europe in the last century;

among these, red oak (*Quercus rubra* L.) is a common invader of European temperate forests. Native to North America, red oak was introduced in Europe in the eighteenth century as timber species and ornamental plant. Red oak grows up to 60% faster than common oak (*Quercus robur* L.) and sessile oak (*Quercus petraea* (Matt.) Liebl.) (Magni Diaz 2004; Vansteenkiste et al. 2005), flourishes in acidic conditions, and may tolerate compacted and periodically flooded soils; a thick litter layer protects the acorns against decay and inhibits the renewal of other tree species.

Compared with the extensive literature on the effects of invasive species on native plant community, structure, and understory environment (Chmura 2013; Hejda et al. 2009; Lenda et al. 2013; Vilà et al. 2011), studies on the impact of red oak on soil properties are still limited and provided sometimes conflicting results (Bonifacio et al. 2015; Miltner et al. 2016; Riepsas and Straigyte 2008; Stefanowicz et al. 2017). Unlike vegetation dynamics, which occur rapidly and are easily observable and interpretable, the understanding of soil dynamics and processes is complex, getting more difficult the formation of any generalizations on the invasion influence on soil. The great spatial

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variability in pedological and pedoclimatic parameters and the need to have a high number of samples to significantly characterize these parameters generate additional issues in understanding soil dynamics.

Vegetation affects soil characteristics mainly through litterfall, root turnover, and exudation of different compounds. The rate of decomposition of plant residues and the incorporation of organic matter in mineral horizons depend on forest types. In a forest ecosystem, the humus form derives from the equilibrium between litter production, decomposition, and humification, resulting from the biological action of bacteria, fungi, and soil fauna. The diversity of humus forms may be related to the adoption of different strategies for the acquisition and use of resources by ecosystems (Ponge 2003). It follows that the humus form can act as a field indicator of environmental variations, such as substitution of vegetation.

In northern Italy, mesophytic mixed oak forests are among the most threatened by artificially introduction of red oak, which rapidly spreads due to its ability to compete with shade-tolerant species, to quickly grow and resist to disease and water stress.

We assumed that the presence of red oak may modify soil characteristics starting from the organic layers and deepening with the duration of its permanence. The aim of this work was to evaluate and quantify the impact of such alien plant on humus forms and soil properties, considering variability in soil characteristics; we present here the results of two paired plot studies in the Lombardy plain (Italy), comparing natural mixed forests representing the original vegetation of the Po Valley with 50- and 80-year-old red oak stands.

2 Material and methods

2.1 Study sites

The two study sites were Bosco Vacaressino (at 45° 20' 47" N, 8° 56' 37" E, elevation 94 m a.s.l.) and Bosco Ginestre (at 45° 20' 39" N, 8° 55' 18" E, elevation 87 m a.s.l.); they are located within the Parco Regionale del Ticino, Italy, near Morimondo and about 7 km northwest of the city of Vigevano. The distance between the sites is 1700 m (Fig. 1).

The soils, developed on alluvial deposits of the Ticino River, show a limited degree of pedogenesis (Tables 2 and 3 in Annex 2; Fig. 5 in Annex 1): according to the World Reference Base (WRB) classification (IUSS Working Group WRB 2015), they are Regosols, Umbrisols, and Cambisols, rich in organic matter and desaturated in bases at the surface, but tending to saturation in depth, with coarse sandy texture at the surface and often with

abundant gravel in depth. From pedogenetic point of view, less evolved soils (Regosols) are found on younger surfaces (Bosco Ginestre, recent alluvial terrace), and most evolved ones (Cambisols), which show cambic horizon, are found on older surfaces (Bosco Vacaressino, ancient alluvial terrace). The climate is temperate continental, as monitored by the meteorological station at Vigevano (1970–2015), with yearly average rainfall of 1032 mm (maximum in October–November, minimum in January–February) and mean air temperature of 11.3 °C. As the water reserve of the soil (AWC) is low (shallow soils, coarse texture), in the warmer years with little rain (e.g., 2015), the water deficit can be high (Fig. 6 in Annex 1).

The natural land cover of the area is Padanian-Ilyrian hardwood forest in transition to mesophytic Padanian mixed oak forest (Bohn et al. 2000). The landscape was mostly forested (dominated by deciduous oaks, *Quercus* spp., and hornbeam (*Carpinus betulus* L.) until late Middle Age clearings (Ravazzi et al. 2013). Currently, a few areas have maintained the past forest cover that has not been replaced by agricultural crops and, according to historical and cadastral documents (dating back to 1722: Teresian Cadastre), has never changed, at least in the last 300 years.

Mixed forest (MF) is nowadays dominated by common oak and hornbeam; in the shrub layer, there are common hazel (*Corylus avellana* L.), black cherry (*Prunus serotina* Ehrh.), wild cherry (*Prunus avium* L.), black locust (*Robinia pseudoacacia* L.), spindle (*Euonymus europaeus* L.), and elder (*Sambucus nigra* L.); periwinkle (*Vinca minor* L.) and lily of the valley (*Convallaria majalis* L.) are often present in the herbaceous layer.

In some parts of the study sites, red oak was introduced in order to take advantage of its rapid growth and disease resistance, resulting in the creation of red oak forest spots where native species were completely replaced, surrounded by mixed forest. In the two study sites, it was therefore possible to investigate the effect of red oak on soils by comparing data with neighboring mixed forest areas.

The vegetation of the red oak forests is dominated in the tree layer by red oak trees of about 50 years (Bosco Vacaressino) and 80 years (Bosco Ginestre), as estimated by dendrochronological analyses on collected increment cores using a Pressler borer. Red oak stands are dominant in the vigorous rejuvenation layer too, where there is also common hawthorn (*Crataegus monogyna* Jacq.), common hazel, black locust, and black cherry; among the herbs, red bracken (*Pteridium aquilinum* L.) is often present.

At Bosco Vacaressino, the mixed forest zone is separated from the red oak forest by a relict riverbed, about 20 m wide, which is constantly humid and without trees.

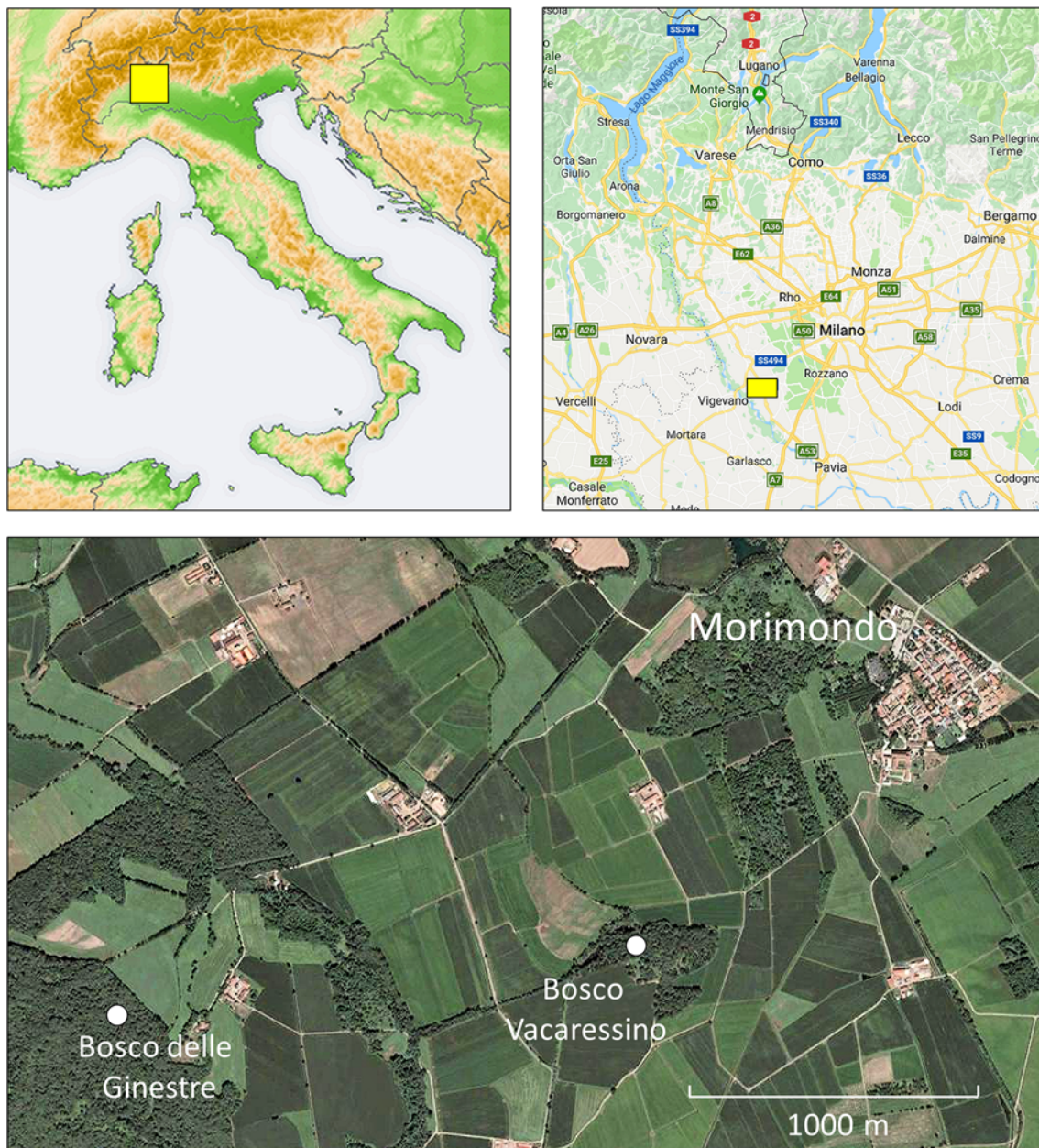


Fig. 1 Study sites: Bosco Vacaressino (BV) and Bosco Ginestre (BG)

2.2 Sampling and analyses of organic horizons and mineral layers

The effects of conversions from mixed to red oak forest were investigated by sampling and comparing neighboring forests. Humus forms and soil properties were assessed accounting for site spatial variability. Due to the albeit limited variability in thickness and typology of mineral soil horizons, sampling by layers (3 layers) was chosen.

Sampling was performed using a cylindrical core sampler (5.4-cm diameter) for the first two layers and a gouge auger (Eijkelkamp; 2.5-cm diameter) for the deeper layer. The first

layer was chosen because it corresponded to the surface portion of soil, rich in organic matter and more involved by the change of vegetation cover, the last one as apparently not involved in the conversion and the intermediate layer to evaluate the vertical trend of the impact; for the two study sites, different layer thicknesses were identified, basing on characteristics of representative pedological pits (Table 2 in Annex 2).

At Bosco Vacaressino, sampling was performed during summer 2015 over an area of about 2 ha, at 40 (20 for each forest type) georeferenced sampling points based on a random scheme. According to soil pit description, for this study area, the 3 investigated layers were as follows: 0–10 cm

(corresponding approximately to A and AB horizons), 10–30 cm (corresponding to AB and Bw horizons), and 30–60 cm (corresponding to BC horizon).

At Bosco Ginestre, the sampling was performed during late summer 2014 over a total area of about 2 ha. Soil samples were collected at 49 (20 at mixed forest and 29 at red oak forest) randomly selected georeferenced points, from 3 layers: 0–10 cm (corresponding approximately to A horizon), 10–35 cm (mainly corresponding to transition AC horizons), and 35–60 cm (corresponding to C1 and C2 horizon).

The coring survey was carried out to uniformly assess the impact of forest conversion by comparing equivalent soil volumes and, concerning soil stock only, equivalent soil masses; in fact, in this study, SOC_{stock} was compared between the different forest types by evaluating equivalent masses (Poepflau et al. 2011), accounting for differences in soil BD since major errors have been found in quantifying changes in SOC_{stock} to fixed depth compared with quantification in equivalent soil mass (Ellert and Bettany 1995). For each site, the largest soil masses (considering a thickness of 30 and 35 cm for Bosco Vaccaressino and Bosco Ginestre respectively) were identified and considered as reference mass; all the soil masses in correspondence of the sampling points were then aligned to the reference mass.

Soil samples were air dried, sieved (2-mm mesh), and analyzed to determine soil organic carbon (SOC) and total nitrogen (TN) content (Flash EA 1112 NCSoil, Thermo Fisher Scientific elemental analyzer, Pittsburgh, PA, USA), pH in water (soil to water ratio of 1:2.5), particle-size distribution by sieving and sedimentation (Burt 2004) (sand, 0.05–2 mm; silt, 0.002–0.05; clay, < 0.002 mm), for a total of 44 and 32 sampling points at Bosco Vaccaressino and Bosco Ginestre respectively. Available phosphorus (Olsen et al. 1954) was determined in the 0–10 cm layer only. List and abbreviations of all investigated parameters are shown in Table 1.

For each monitoring point, soil bulk density (BD) was determined for the first two layers with the cylindrical core method (core diameter 5.4 cm) on undisturbed core samples, considering the volume of stones, which were mainly absent or few at Bosco Ginestre and common (mean \pm SD, $12.3 \pm 4.7\%$) at Bosco Vaccaressino. Considering BD and stone volume, SOC_{stock} was computed on an area basis for mineral layers.

At each soil sampling point, humus form was described in the field and then classified according to Zanella et al. (2011, 2018), considering the presence and characteristics of OL, OF, and OH organic horizons and of A mineral horizon.

Organic horizons were sampled using a 30×30 cm frame and the collected biomass was oven-dried at 70 °C for 48 h and weighed. The organic carbon content (OC) of organic horizons was determined by combustion with a muffle furnace at 550 °C for 4 h and converted to content on area basis (kg m^{-2}).

The Humus Index (Hum_{ind}), which was proposed and designed for the transformation of a scale of discrete humus forms

in a numerical parameter (Ponge et al. 2002), was used; the humus forms were classified and scaled in order of increasing accumulation of organic matter in the O horizons and decreasing burrowing activity in the A horizon as follows: 1: Eumull (not present in our study sites), 2: Mesomull, 3: Oligomull, 4: Dysmull, 5: Hemimoder, 6: Eumoder, and 7: Dysmoder.

In text and figures, for the comparison between forest types, we used average values and standard deviations.

2.3 Statistical analyses

In order to evaluate the effects of forest conversion on soil properties and humus forms, the mixed effect model procedure was performed (Bolker et al. 2009), testing for autocorrelation among the model residuals (Searle et al. 2009). Each variable response in relation to forest type was evaluated considering forest type as a fixed effect in the linear mixed model. Statistical analyses were performed with PROC MIXED of SAS software package (release 9.4; SAS Institute) (Littell et al. 2006). The simultaneous estimates of covariance parameter and fixed effect coefficients were obtained by restricted maximum likelihood (REML) estimation (Littell et al. 2006). The spatial covariance function of residuals was determined iteratively using the statement REPEATED, by estimating partial sill, range, and nugget effect parameters. Residual spatial correlation was found for SOC_1 , CN_1 , and CN_2 at Bosco Vaccaressino and for pH_{w1} , SOC_{stock} , and CN_1 at Bosco Ginestre.

If linear model assumptions on residuals distribution were not satisfied, as in the case of pH_w of all three investigated layers at Bosco Vaccaressino and of OC_{OF} , OC_O , and CN_1 at Bosco Ginestre, Gaussian anamorphosis transformation of the response variable was performed using ISATIS software package (release 3.01; Geovariances, 2016).

3 Results

3.1 Humus forms

The main humus forms found at Bosco Vaccaressino were, in order of decreasing biological activity according to Hum_{ind} (Ponge et al. 2002; Zanella et al. 2018): Mesomull, Oligomull, Dysmull, Hemimoder, and Eumoder. Mixed forest was characterized by Mull forms ($Hum_{ind} \leq 4$) characterized by fast biodegradation and rapid disappearance of litter from the topsoil by mainly anecic and endogeic earthworms and bacteria, with prevalence of Mesomull, distinguished by the absence of OF horizon, followed by Dysmull and Oligomull. In the red oak forest besides Mull forms (mainly represented by Dysmull), Moder forms (Hum_{ind} 5–6), where slower biodegradation by arthropods,

Table 1 Investigated soil properties and their abbreviations

Soil properties	Unit	Abbreviation
Organic carbon stock of OL organic horizon	kg m ⁻²	OC _{OL}
Organic carbon stock of OF organic horizon	kg m ⁻²	OC _{OF}
Organic carbon stock of OH organic horizon	kg m ⁻²	OC _{OH}
OC _{OL} + OC _{OF} + OC _{OH}	kg m ⁻²	OC _O
Soil organic carbon content of the 0–10 cm mineral soil layer	%	SOC ₁
Soil organic carbon content of the 10–30 cm or 10–35 cm mineral soil layer	%	SOC ₂
Soil organic carbon content of the 30–60 cm or 35–60 cm mineral soil layer	%	SOC ₃
Soil organic carbon stock of the 0–30 cm or 0–35 cm mineral soil layer	kg m ⁻²	SOC _{stock}
C:N ratio of the 0–10 cm mineral layer		CN ₁
C:N ratio of the 10–30 cm or 10–35 cm mineral layer		CN ₂
C:N ratio of the 30–60 cm or 35–60 cm mineral layer		CN ₃
pH in water of the 0–10 cm mineral layer		pH _{w1}
pH in water of the 10–30 cm or 10–35 cm mineral layers		pH _{w2}
pH in water of the 30–60 cm or 35–60 cm mineral layers		pH _{w3}
Available phosphorus content of the 0–10 cm mineral layer	mg kg ⁻¹	P _{av}
Humus index		Hum _{ind}
Bulk density of the 0–10 cm mineral layer	g cm ⁻³	BD ₁
Bulk density of the 10–30 cm or 10–35 cm mineral layers	g cm ⁻³	BD ₂

enchytraeids, and fungi resulted in the appearance of an OH horizon overlying a biomeso- or biomicrostructured A horizon, were found (Fig. 2).

At Bosco Ginestre, the humus forms found were as follows: Mesomull, Oligomull, Dysmull, Hemimoder, and Dysmoder (Fig. 2). Mixed forest was dominated by Mull

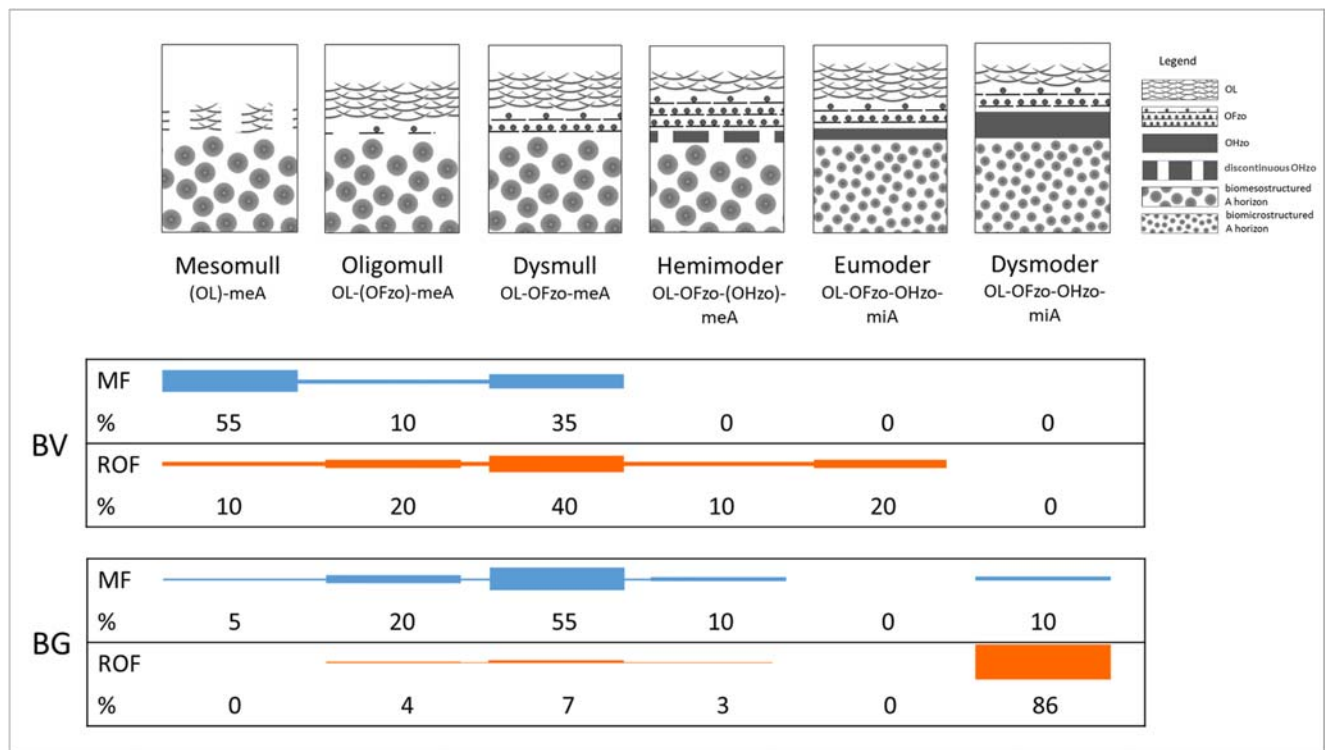


Fig. 2 Comparison of type and presence percentage of humus forms between natural mixed forest (MF) and red oak forest (ROF) types at Bosco Vacaressino (BV) and Bosco Ginestre (BG). zo: zoogenic horizon; meA: biomesostructured A horizon; miA: biomicrostructured A horizon

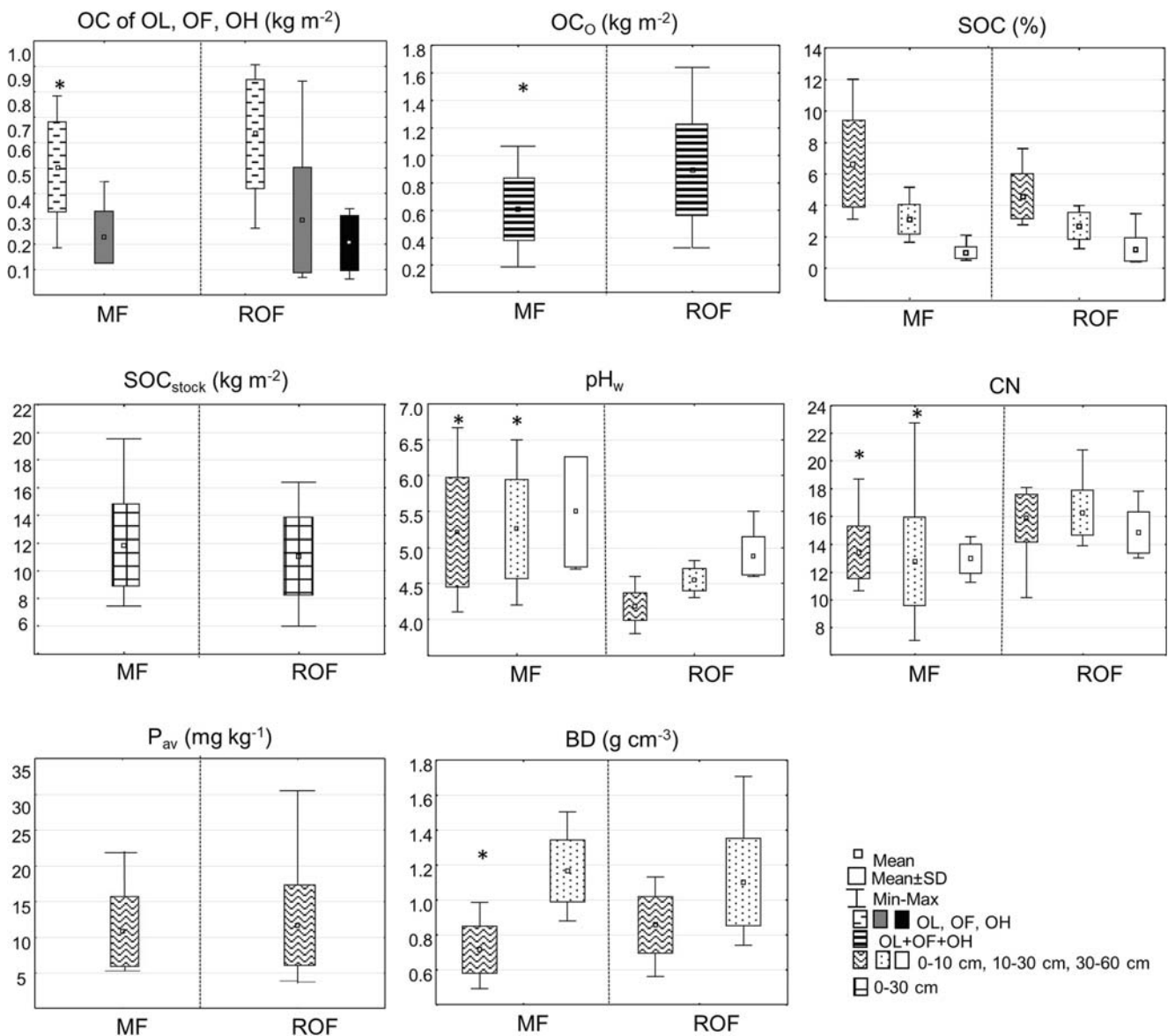


Fig. 3 Box plots for comparison of properties of organic and mineral layers between soil forest types (mixed forest, MF; red oak forest, ROF) at Bosco Vacaressino (BV). Asterisk indicates significant differences ($p < 0.05$) of response variable between forest types in the mixed model

forms (Hum_{ind} 2–4); the most represented Mull type was Dysmull with a continuous OF horizon overlying a biomesostructured A horizon. Dysmoder (Hum_{ind} 7) was the main humus form found in red oak forest (Fig. 2).

3.2 Carbon stock of organic horizons

At Bosco Vacaressino, OC_{OL} was statistically higher in red oak forest compared with mixed forest, whereas no statistical differences in OC_{OF} were found between the two forest types. The average OC_{OH} of the thin or discontinuous OH horizon in the red oak forest was $0.21 \pm 0.11 \text{ kg m}^{-2}$ (Fig. 3 and Tables 4 and 5 in Annex 2). In its entirety, the OC_O, obtained summing the OC of all the present organic horizons, was statistically

higher in red oak forest compared with mixed forest for both study sites with differences between red oak forest and mixed forest more accentuated in Bosco Ginestre (1.31 kg m^{-2}) than in Bosco Vacaressino (0.28 kg m^{-2}).

At Bosco Ginestre, the average OC in red oak forest was $0.58 \pm 0.25 \text{ kg m}^{-2}$ and $0.14 \pm 0.08 \text{ kg m}^{-2}$ in OL and OF horizons, respectively, statistically higher ($p < 0.05$) than OC_{OL} and OC_{OF} of mixed forest (Fig. 4 and Tables 4 and 5 in Annex 2). The OH horizon of red oak forest showed an average OC_{OH} of $1.28 \pm 1.05 \text{ kg m}^{-2}$.

The variability in humus forms within each type of forest was reflected in the variability of the OC_O: the coefficients of variation ranged from a minimum of 38% (Bosco Vacaressino) to a maximum of 65% (mixed

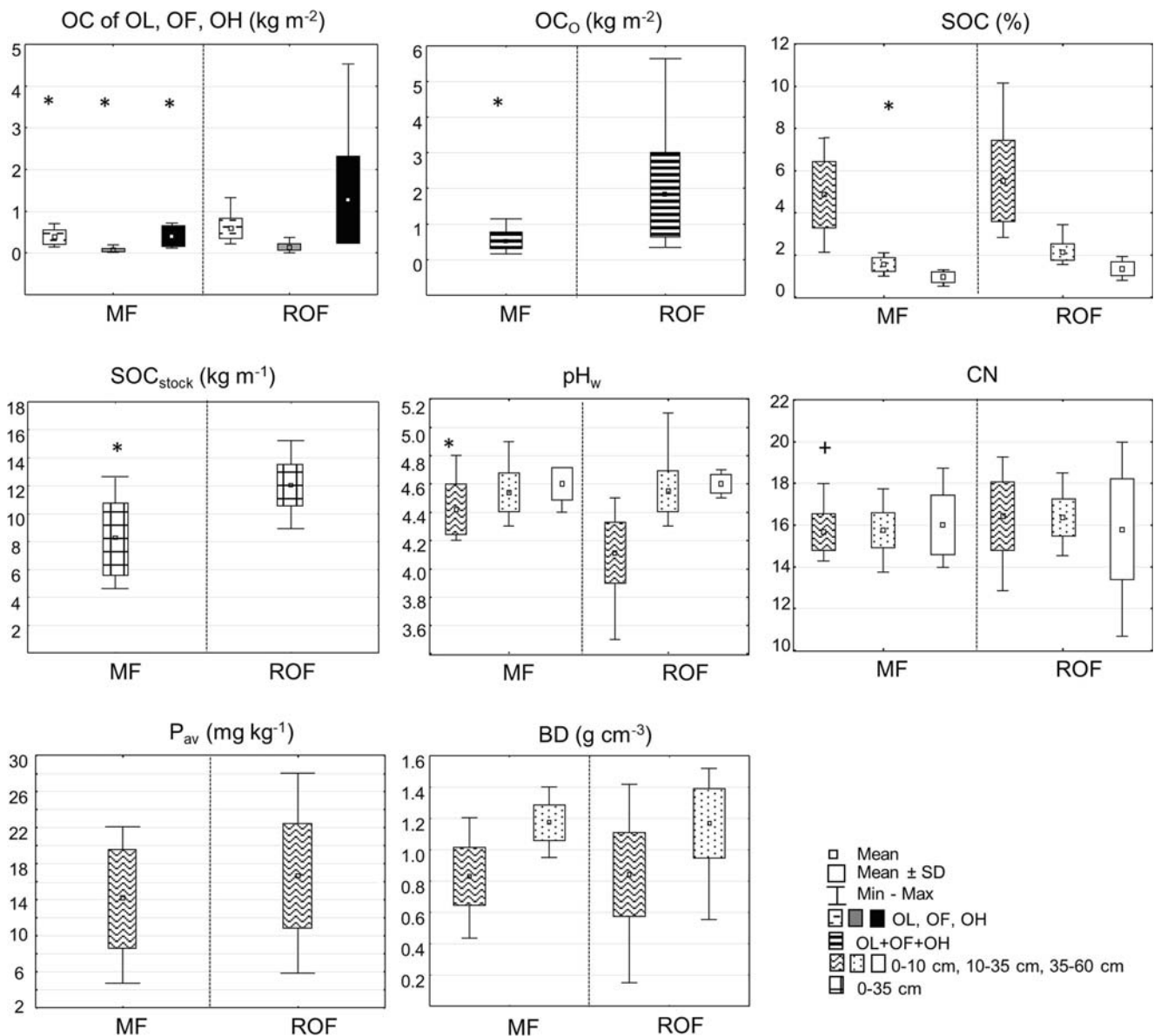


Fig. 4 Box plots for comparison of properties of organic and mineral layers between soil forest types (mixed forest, MF; red oak forest, ROF) at Bosco Ginestre (BG). Asterisk and plus sign indicate

significant differences ($p < 0.05$ and $p < 0.1$, respectively) of response variable between forest types in the mixed model

forest at Bosco Ginestre) with higher variations in OF and OH horizons than in OL horizons.

3.3 Mineral soil

In mixed forest at the Bosco Vaccaressino site, the SOC averaged to $6.6 \pm 2.8\%$, $3.1 \pm 1.0\%$, and $1.17 \pm 0.80\%$ in the first, second, and third mineral layer respectively (Fig. 3 and Tables 4 and 5 in Annex 2). BD₁ averaged 0.71 ± 0.14 g cm⁻³, increasing in the second investigated layer to 1.16 ± 0.18 g cm⁻³. The average pH_w value of the first layer was 5.2 ± 0.8 , remaining stable in the second layer, and increasing to 5.9 ± 0.7 in the third layer. C:N ratios were similar for all the

investigated layers, averaging 13.3. The average P_{av} content was 10.82 ± 4.94 mg kg⁻¹. No significant differences among forest types were found in P_{av} and SOC at Bosco Vaccaressino, but the type of organic matter (as shown by C:N values) soil reaction, and BD₁ discriminated red oak forest from mixed forest (Fig. 3 and Tables 4 and 5 in Annex 2).

In the mixed forest at Bosco Ginestre, the SOC of the mineral soil was high in the first two layers, averaging $4.85 \pm 1.58\%$ and $1.54 \pm 0.34\%$, respectively, and decreased to $0.94 \pm 0.27\%$ in the 35–60-cm layer (Fig. 4 and Tables 4 and 5 in Annex 2); pH_w value was 4.4 ± 0.2 in the surface mineral layer slightly increasing with depth. The C:N ratio was 15.7 ± 0.9 until a depth of 35 cm. The average P_{av} was

$14.1 \pm 5.5 \text{ mg kg}^{-1}$, ranging between 4.7 and 22.1 mg kg^{-1} . BD was $0.83 \pm 0.19 \text{ g cm}^{-3}$ in the 0–10-cm layer and $1.17 \pm 0.12 \text{ g cm}^{-3}$ in the 10–35-cm layer.

SOC was higher in red oak forest than in mixed forest; the first layer showed an average value of $5.5 \pm 1.9\%$ but no statistical differences were found between the two forest types, differently from the second layer where SOC was statistically higher in red oak forest. The third layer showed an average SOC not different from that of the mixed forest (Fig. 4 and Tables 4 and 5 in Annex 2). Statistical differences between the two vegetation cover types were also found in pHw₁ ($p < 0.05$) and CN₁ ($p < 0.1$), lower and higher, respectively, in red oak forest than in mixed forest.

The third investigated layer showed similar soil characteristics in both forest types of each investigated site, suggesting a non-involvement of deep soil layer in forest conversion effects. For this reason, only the SOC_{stock} of the first two layers together, by comparing equivalent soil mass, was considered. At the Bosco Vacaressino, the SOC_{stock} of the two forest types was not statistically different, while at Bosco Ginestre, red oak forest showed an average value of 12.04 kg m^{-2} (ranging between 9.8 and 15.7 kg m^{-2}), statistically higher than that of mixed forest ($8.31 \pm 2.83 \text{ kg m}^{-2}$).

4 Discussion

4.1 Site comparability and spatial variability

Reliable evaluations of changes in soil characteristics due to changes in vegetation cover are possible if the comparability of sites is guaranteed. However, the spatial variability that characterizes the soil properties increases the uncertainty of such estimates, especially when dealing with forest ecosystems where soil variability is even more accentuated by heterogeneity in tree and shrub composition (Ferré et al. 2014; Kounda-Kiki et al. 2008). To satisfy the requirements for assessing the impact of vegetation cover changes on humus forms and soil properties, we selected the two sites because each of them includes both mixed and red oak forest stands with comparable land use history until time of conversion, and same pedological conditions. Since soil texture does not depend on the forest type but on the nature of the parent material from which the soil has developed, the common pedological origin of soils within each study site was verified and confirmed by soil texture comparability. For both sites, the texture (USDA class) of the subsoil was mainly sandy loam with no differences between red oak forest and mixed forest.

The detailed sampling performed at the study sites enabled considering the spatial variability in humus forms and soil properties, ensuring the representativeness of the obtained results. Considerable variability in humus forms and soil properties was found not only in each paired plot, but also within each forest stand. The spatial distribution of the organic and mineral

layer properties probably reflected the heterogeneity of forest vegetation, particularly in mixed forest, where species were various and scattered in all forest layers. Often, a spatial structure of residuals was not found, due to an inherent variability on a spatial scale shorter than that of the measurements.

4.2 Effects of forest conversion on humus forms and organic horizon carbon stock

Replacing mixed oak-hornbeam forest with red oak forest caused a shift from Mull to Moder forms (Gentili et al. 2019) mainly as a consequence of a recalcitrant litter in red oak forest types (Jonczak et al. 2015). Decomposition of plant litter is a complex process which is controlled by climate, litter quality, and microbial communities (Couteaux et al. 1995; Regina and Tarazona 2001). In our case, in which we may safely exclude climate as factor of variation, litter quality mainly influenced the decomposition rate, but also the dynamics of nutrient mineralization and immobilization (Regina and Tarazona 2001), which in turn may have affected microbial communities (Graça and Poquet 2014) directly involved in litter decomposition. Some studies reported relationships between the rate of leaf litter decomposition and leaf litter C:N ratios (Hobbie 2015; Steffen et al. 2007). The effects of litter chemistry on microbial activity also involve the role of polyphenolic compounds (Steltzer and Bowman 2005). In particular, concerning red oak, Talbot and Finzi (2008) observed that litter tannins, during periods of large protein inputs (following root turnover, or after leaf senescence), play an important role in lowering the rate of soil nutrient cycling by binding proteins into recalcitrant complexes.

Evaluating the effect of the introduction of red oak in areas of mixed broadleaved forests of northwestern Italy, Bonifacio et al. (2015) found a relationship between the different transformation of organic layers of the compared forest types and differences in litter biochemical composition consisting of a larger presence of tannins and a higher aromatic C/O-alkyl C and alkyl C/O-alkyl C ratios, which were indicative of lower degree of litter decomposition, in red oak forests than in native forests.

The Intergovernmental Panel on Climate Change recognized litter carbon as one of five C pools in forest ecosystems included in the Agriculture, Forestry, and Other Land Use sector of the annual national greenhouse gas inventories (IPCC 2006). The litter layer accounts for about 5% of all forest ecosystem carbon stocks worldwide (Pan et al. 2011), so changes in the litter carbon pool have important implications for global carbon budgets. The presence of the complete sequence of organic horizons (OL, OF, and OH), visible in part of the red oak forest at Bosco Vacaressino and in most of the red oak forest at Bosco Ginestre, and the significantly higher OC in red oak forest of both study areas, supported the lower turnover of red oak organic matter and the capacity of red oak forest organic layers to act as a carbon sink. As a consequence of 50 years of red oak persistence, we found an

increase in OC_O of about 50% compared with the natural mixed forest, whereas in the 80-year-old red oak forest, the OC_O almost tripled with respect to OC_O of the mixed forest. The shift of humus forms toward a decrease in biological activity was more emphasized at Bosco Ginestre compared with the Bosco Vacaressino site: in the older red oak forest, in addition to higher accumulation of OC in the organic layers, substantial increases in Hum_{ind} were also observed, in accordance with what was reported by Ponge and Chevalier (2006), who found a positive relationship between the Humus index and the age of forest stands.

4.3 Effects of forest conversion on mineral layers

The conversions from mixed to red oak forests affected the mineral topsoil, although, concerning some parameters, to a lesser extent than humus forms, since changes in soil characteristics are much slower than changes in humus forms as suggested by some space-for-time studies (Bernier and Ponge 1994; Dimbleby 1962; Willis et al. 1997).

Soil acidification occurred in red oak forest at both sites. Similarly, Miltner et al. (2016) found a decrease in pH_w values when investigating the effects of red oak on soil properties at forest sites in the Czech Republic. In particular, in Moder forms like those that predominantly characterized the ROF at the Bosco Ginestre site and part of red oak forest at the Bosco Vacaressino site, most of microbial biomass is fungal due to the more acidic conditions than in Mull (Nagel-de-Boois and Jansen 1967). Fungi produce antibiotics, leading to breakage of bacteria and excrete organic acids resulting in further acidification of the soil (Takao 1965). However, lowering of soil pH_w due to “acidifying” red oak litter was particularly marked in originally less acid soils probably due to the fact that the acidification trend of a soil decreases at lower soil pH (Wiklander and Andersson 1972).

Moder humus forms are generally characterized by smaller, lower abundance, and lower diversity macrofauna compared with Mull humus forms (Schaefer 1991). The biological activity of organic matter was lower in red oak forest compared with mixed forest, resulting in organic matter accumulation and slower incorporation into the mineral soil. For the 50-year-old red oak forest, vegetation effects on organic carbon stock were observed only in organic horizons; for the 80-year-old red oak forest, effects of vegetation were visible in the mineral soil, too. As a consequence of 80 years of red oak forest, we found an increase in soil SOC_{stock} of about 45% compared with the native forest.

Vegetation conversion also affected soil organic matter quality as shown by the higher soil C:N ratio in red oak forests than in mixed forests. Similar to our findings but concerning the spruce forests in the Italian Alps, Salmon et al. (2006) found increases in soil carbon content and C:N

ratio that were accentuated in mature spruce stands compared with younger ones.

Different from what was reported in previous studies (Bonifacio et al. 2015; Miltner et al. 2016), we found no difference between forest types in the P_{av} of the investigated mineral layers. This finding could be related to the fact that the spatial distribution of this soil characteristic was highly variable.

5 Conclusions

We studied the effects of afforestation with red oak on humus forms and soil properties in two paired plots through a sampling scheme that considered the horizontal and vertical spatial variability to ensure representativeness of the obtained results. The comparability of soil properties between the two neighboring forests was guaranteed by performing soil sampling by depth increments and, concerning SOC stock, by comparing equivalent soil masses.

Besides the well-known effects of red oak on the native plant community, whose structure is altered and biodiversity is significantly reduced in tree, shrub, and herbaceous layer, this alien species may also impact the soil. In forest management aimed at the establishment of red oak plantations, it is important to take into account that, if on the one hand, the recalcitrant nature of the litter favors the accumulation of the organic residues on topsoil, promoting the carbon sink role of soil, on the other hand, it inhibits the processes of transformation and incorporation of the organic matter into mineral soil, which occur more slowly than in native forests, and favors mineral soil acidification.

In detail, major changes in soil characteristics mainly involved organic layers: vegetation changes were reflected in changes in litter quantity and quality. The substitution of natural mixed forest by red oak forest strongly changed organic horizons, resulting in a shift of humus forms from Mull to Moder, i.e., a shift toward less active humus forms.

The effects of vegetation conversions were also visible in mineral layers: soil acidification, increase in C:N ratio, reflecting change in organic matter characteristics toward a quality worsening, and higher SOC_{stock} in red oak forest compared with mixed forest were observed. Many of the detected modifications, whether they involved organic or mineral horizons, increased according to stand age. Although a wider range of investigated tree ages would permit a better assessment of age impact, we observed that the changes in the humus forms and associated parameters, such as organic matter accumulation in the topsoil and increase in SOC stock, were more evident with increasing time elapsed from the red oak forest establishment.

Annexes

Annex 1

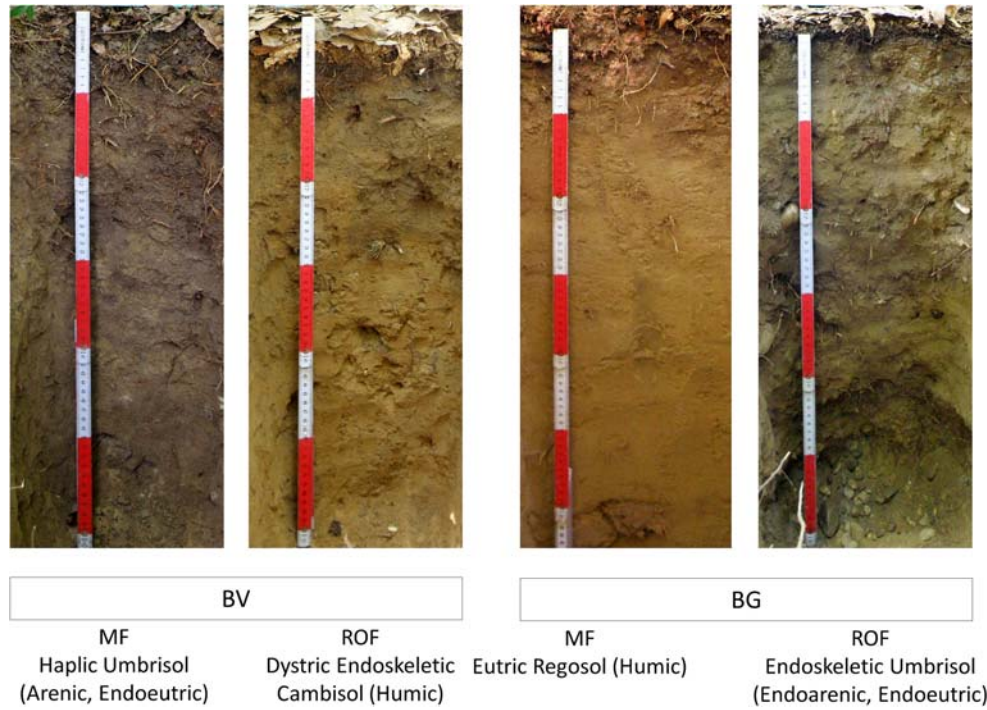


Fig. 5 Soil profiles of mixed forest (MF) and red oak forest (ROF) at study sites Bosco Vacaressino (BV) and Bosco delle Ginestre (BG). Taxonomy of soil profiles according to IUSS Working Group WRB (2015)

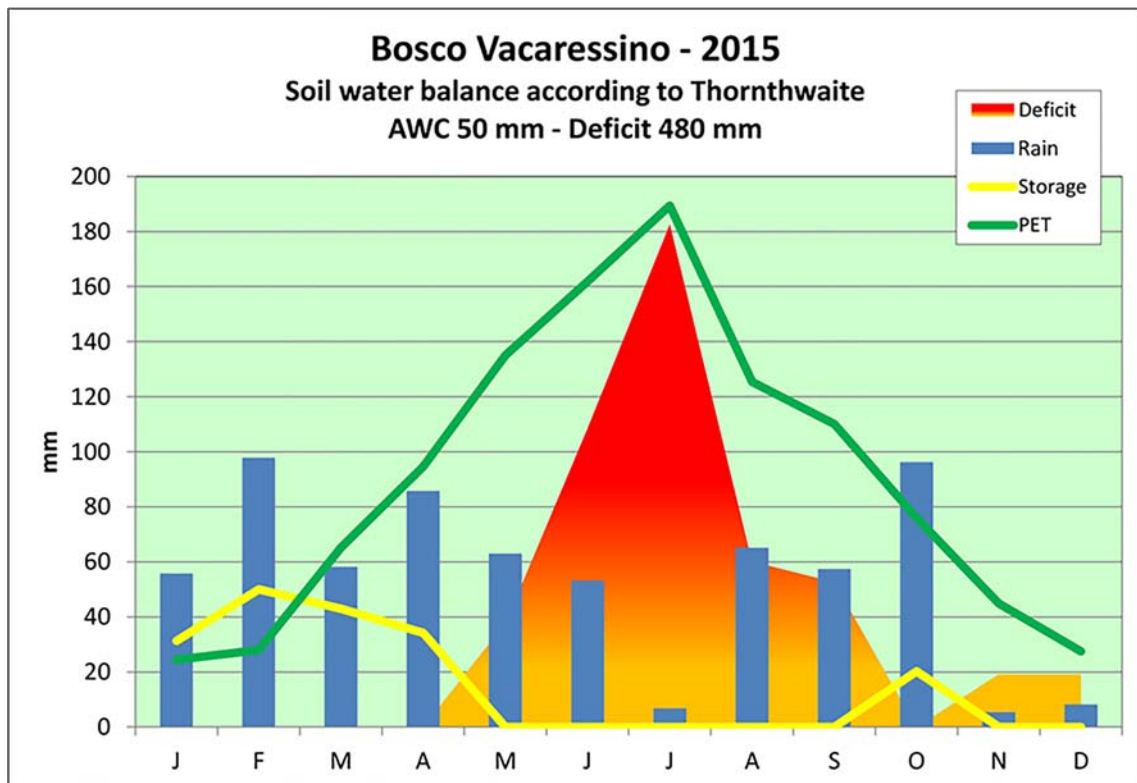


Fig. 6 Soil water balance according to Thornthwaite for Bosco Vacaressino

Annex 2

Table 2 Soil profile descriptions. Taxonomy of soil profiles according to IUSS Working Group WRB (2015)

Profiles and horizons	Color (moist)	Mottles	Rock fragments	Structure	Roots
BV (Bosco Vacaressino)					
MF (mixed forest)—Haplic Umbrisol (Arenic, Endoeutric)					
A (0–3 cm)	10YR 2/2	Absent	Common, f–vf	Granular, m	Many, f–vf
AB (3–27 cm)	10YR 3/3	Absent	Frequent, f–vf	Subang. blocky, m	Common, vf–f
BC (27–60 cm)	10YR 4/4	Absent	Frequent, f–vf	Massive	Few, f–m
ROF (red oak forest)—Dystric Endoskeletal Cambisol (Humic)					
A (0–2 cm)	10YR 2/2	Absent	Few, f–vf	Single grain	Many, f–vf
AB (2–13 cm)	10YR 3/3	Absent	Few, f–vf	Granular, m	Common, f–vf
Bw (13–33 cm)	10YR 4/5	Absent	Few, f–vf	Subang. blocky, m	Common, f–m
BC (33–60 cm)	10YR 3.5/6	10YR 6/6, many	Abundant, f–vf	Massive	Few, m
BG (Bosco Ginestre)					
MF (mixed forest)—Eutric Regosol (Humic)					
A (0–9 cm)	10YR 3/2.5	Absent	Absent	Granular, f	Many f–vf
AC (9–20 cm)	10YR 4/3	Absent	Absent	Subang. blocky, m	Common, vf–f
C1 (20–35 cm)	10YR 4.5/3	Absent	Absent	Subang. blocky, m	Few, vf
C2 (35–77 cm)	10YR 5/4	Absent	Absent	Subang. blocky, m	Few, f–c
C3 (77–90 cm)	10YR 5/3	Absent	Absent	Single grain	Absent
ROF (red oak forest)—Endoskeletal Umbrisol (Endoarenic, Endoeutric)					
A (0–10 cm)	10YR 3/3	Absent	Frequent, f–vf	Subang. blocky, f	Common, f–vf
AC (10–35 cm)	10YR 3/3	Absent	Frequent, f–vf	Subang. blocky, m	Common, f–vf
C1 (35–50 cm)	10YR 4/4	Absent	Frequent, f–vf	Single grain	Few, f–vf
C2 (50–65 cm)	10YR 5/4	Absent	Abundant, v–vf	Single grain	Few, vf

vf, very fine; f, fine; m, medium; c, coarse; vc, very coarse

Table 3 Soil profile analyses

Profiles and horizons	pHw	pHKCl	Org. C (g kg ⁻¹)	Total N (g kg ⁻¹)	C:N	Sand 2–0.05 mm (g kg ⁻¹)	Silt 0.05–0.002 mm (g kg ⁻¹)	Clay < 0.002 mm (g kg ⁻¹)	Text. class	CEC (cmol ₍₊₎ kg ⁻¹)	Exchangeable cations (cmol ₍₊₎ kg ⁻¹)			BS (%)	Available P (g kg ⁻¹)	
											Ca	Mg	Na K			
BV (Bosco Vaccaressino)																
MF (mixed forest)																
A (0–3 cm)	5.2	4.5	59	4.5	13.0	714	245	41	SL	17.9	11.57	1.62	0.08	0.36	76.1	9
AB (3–27 cm)	5.2	4.3	23	1.9	12.2	726	241	33	SL	10.1	3.48	0.52	0.05	0.13	41.6	2
BC (27–60 cm)	6.0	4.3	5	0.3	13.8	829	162	9	LS	5.1	2.90	0.53	0.12	0.08	70.9	1
ROF (red oak forest)																
A (0–2 cm)	3.8	3.1	91	4.7	19.4	694	180	126	SL	24.6	2.03	0.49	0.01	0.05	10.5	20
AB (2–13 cm)	4.3	3.6	22	1.5	15.0	657	311	32	SL	13.0	0.96	0.24	0.08	0.01	10.0	5
Bw (13–33 cm)	5.1	4.0	7	0.5	14.8	627	286	87	SL	4.6	1.06	0.27	0.07	0.02	31.0	3
BC (33–60 cm)	4.5	3.7	3	0.2	14.4	614	344	42	SL	2.8	0.95	0.24	0.06	0.02	45.4	1
BG (Bosco Gimestre)																
MF (mixed forest)																
A (0–9 cm)	4.5	3.6	77	5.4	14.4	698	261	41	SL	21.9	5.27	1.37	0.12	0.18	31.7	17.2
AC (9–20 cm)	4.6	3.8	23	1.5	15.1	633	324	43	SL	10.9	1.30	0.49	0.30	0.13	20.4	4.7
C1 (20–35 cm)	4.6	4.0	8	0.6	14.1	697	236	67	SL	5.8	1.18	0.50	0.34	0.11	36.8	1.7
C2 (35–77 cm)	5.1	4.1	3	0.2	11.3	725	242	33	SL	3.7	1.46	0.88	0.36	0.13	75.9	1.1
C3 (77–90 cm)	5.8	4.6	1	0.1	10.0	986	14	0	S	1.9	1.06	0.48	0.28	0.07	97.4	0.6
ROF (red oak forest)																
A (0–10 cm)	4.6	3.9	29	1.8	15.6	705	264	31	SL	12.7	0.99	0.24	0.06	0.03	10.4	6.6
AC (10–35 cm)	4.4	4.1	26	1.6	16.1	641	334	25	SL	14.3	0.87	0.21	0.14	0.04	8.8	5.3
C1 (35–50 cm)	4.7	4.3	16	1.0	15.2	673	294	33	SL	3.9	0.90	0.43	0.12	0.02	38.1	2.6
C2 (50–65 cm)	5.1	4.7	2	0.1	18.2	972	28	0	S	1.1	0.52	0.28	0.14	0.02	86.5	1.0

SL, sandy loam; LS, loamy sand; S, sand

Table 4 Descriptive statistics of data of organic horizons and mineral soil layers at study sites

	Valid N	Mean	Min	Max	Std.Dev.	Std.Err.	Var. Coeff
BV (Bosco Vaccaressino)							
ROF (red oak forest)							
SOC ₁ (%)	20	4.56	2.77	7.62	1.44	0.32	31.52
SOC ₂ (%)	20	2.67	1.24	3.97	0.87	0.19	32.59
SOC ₃ (%)	20	1.18	0.41	3.46	0.77	0.17	65.75
CN ₁	20	15.9	10.1	18.1	1.7	0.4	10.9
CN ₂	20	16.3	13.9	20.8	1.6	0.4	10.1
CN ₃	20	14.8	13.0	17.8	1.5	0.3	10.1
pHw ₁	20	4.2	3.8	4.6	0.2	0.0	4.7
pHw ₂	20	4.6	4.3	4.8	0.2	0.0	3.5
pHw ₃	20	4.8	4.4	5.5	0.3	0.1	5.5
BD1 (g cm ⁻³)	20	0.86	0.56	1.13	0.16	0.04	19.06
BD2 (g cm ⁻³)	20	1.10	0.74	1.70	0.25	0.06	22.85
SOC _{OL} (kg m ⁻²)	20	0.63	0.26	0.91	0.22	0.05	34.19
SOC _{OF} (kg m ⁻²)	14	0.30	0.07	0.84	0.21	0.06	70.46
SOC _{OH} (kg m ⁻²)	5	0.21	0.06	0.34	0.11	0.05	53.62
SOCO (kg m ⁻²)	20	0.89	0.33	1.64	0.34	0.08	37.69
SOC _{stock} (kg m ⁻²)	20	11.13	5.97	16.34	2.85	0.64	25.65
Pav (mg kg ⁻¹)	20	11.65	3.50	30.50	5.67	1.27	48.62
Rock fragm ₁ (%)	20	8	1	19	5	1	64
Rock fragm ₂ (%)	20	9	1	32	7	1	73
Sand ₂ (g kg ⁻¹)	20	626	518	692	40	9	6
Silt ₂ (g kg ⁻¹)	20	273	222	355	31	7	11
Clay ₂ (g kg ⁻¹)	20	101	64	127	16	4	16
Sand ₃ (g kg ⁻¹)	12	683	575	780	72	21	11
Silt ₃ (g kg ⁻¹)	12	251	158	366	71	21	28
Clay ₃ (g kg ⁻¹)	12	66.0	55	80	8	2	13
SOC ₁ (%)	20	6.58	3.11	12.03	2.79	0.62	42.43
SOC ₂ (%)	20	3.10	1.67	5.17	0.96	0.21	31.03
SOC ₃ (%)	20	1.17	0.50	2.09	0.80	0.18	40.92
CN ₁	20	13.4	10.6	18.7	1.9	0.4	14.2
CN ₂	20	12.7	7.1	22.7	3.2	0.7	25.1
CN ₃	12	13.0	11.3	14.5	1.1	0.3	8.4
pHw ₁	20	5.2	4.1	6.7	0.8	0.2	14.7
pHw ₂	20	5.3	4.2	6.5	0.7	0.2	13.1
pHw ₃	12	5.9	4.7	6.6	0.7	0.2	12.5
MF (mixed forest)							

Table 4 (continued)

	Valid N	Mean	Min	Max	Std.Dev.	Std.Err.	Var. Coeff
BD1 (g cm ⁻³)	20	0.71	0.49	0.98	0.14	0.03	19.21
BD2 (g cm ⁻³)	20	1.16	0.88	1.50	0.18	0.04	15.38
SOC _{OL} (kg m ⁻²)	20	0.50	0.19	0.78	0.18	0.04	35.47
SOC _{OF} (kg m ⁻²)	9	0.23	0.14	0.45	0.10	0.03	45.07
SOC _{OH} (kg m ⁻²)	-	-	-	-	-	-	-
SOCO (kg m ⁻²)	20	0.61	0.19	1.06	0.23	0.05	37.79
SOC _{stock} (kg m ⁻²)	20	11.88	6.93	19.55	3.07	0.69	25.84
Pav (mg kg ⁻¹)	20	10.82	5.30	21.57	4.94	1.10	45.63
Rock fragm ₁ (%)	20	16	1	31	8	2	52
Rock fragm ₂ (%)	20	20	5	25	7	2	35
Sand ₂ (g kg ⁻¹)	20	637	566	681	32	7	5
Silt ₂ (g kg ⁻¹)	20	276	223	312	25	6	9
Clay ₂ (g kg ⁻¹)	20	87	59	122	18	4	21
Sand ₃ (g kg ⁻¹)	12	713	589	852	98	28	14
Silt ₃ (g kg ⁻¹)	12	241	100	354	96	28	40
Clay ₃ (g kg ⁻¹)	12	46	39	57	6	2	13
BG (Bosco Gimestre)							
SOC ₁ (%)	29	5.50	2.82	10.15	1.93	0.36	35.15
SOC ₂ (%)	29	2.13	1.55	3.42	0.39	0.07	18.54
SOC ₃ (%)	15	1.33	0.80	1.92	0.34	0.09	25.22
CN ₁	29	16.4	12.8	19.2	1.7	0.3	10.1
CN ₂	29	16.1	9.2	18.5	1.6	0.3	9.9
CN ₃	15	15.8	10.6	20.0	2.4	0.6	15.4
pHw ₁	29	4.1	3.5	4.5	0.2	0.0	5.3
pHw ₂	29	4.5	4.3	5.1	0.1	0.0	3.2
pHw ₃	15	4.60	4.50	4.70	0.07	0.02	1.45
BD1 (g cm ⁻³)	29	0.84	0.15	1.42	0.27	0.05	31.86
BD2 (g cm ⁻³)	29	1.17	0.55	1.52	0.22	0.04	19.16
SOC _{OL} (kg m ⁻²)	29	0.58	0.22	1.33	0.25	0.05	42.86
SOC _{OF} (kg m ⁻²)	29	0.14	0.01	0.37	0.08	0.01	55.90
SOC _{OH} (kg m ⁻²)	25	1.28	0.42	4.53	1.05	0.21	82.53
SOC _O (kg m ⁻²)	29	1.83	0.33	5.64	1.19	0.22	65.33
SOC _{stock} (kg m ⁻²)	29	12.04	9.80	15.67	1.57	0.29	12.47
Pav (mg kg ⁻¹)	29	16.61	5.86	28.02	5.82	1.08	35.06
Rock fragm ₁ (%)	29	2	0	11	2	0	128

Table 4 (continued)

	Valid N	Mean	Min	Max	Std.Dev.	Std.Err.	Var. Coeff
Rock fragm ₂ (%)	29	2	0	11	2	0	114
Sand ₂ (g kg ⁻¹)	29	689	561	767	54	10	8
Silt ₂ (g kg ⁻¹)	29	248	172	382	50	9	20
Clay ₂ (g kg ⁻¹)	29	64	19	98	23	4	36
Sand ₃ (g kg ⁻¹)	15	728	575	841	89	23	12
Silt ₃ (g kg ⁻¹)	15	220	119	344	78	20	35
Clay ₃ (g kg ⁻¹)	15	52	35	81	14	4	27
SOC ₁ (%)	20	4.85	2.11	7.54	1.58	0.35	32.52
SOC ₂ (%)	20	1.54	0.99	2.08	0.34	0.08	22.03
SOC ₃ (%)	10	0.94	0.52	1.29	0.27	0.08	28.35
CN ₁	20	15.7	14.3	18.0	0.9	0.2	5.7
CN ₂	20	15.7	13.7	17.7	0.8	0.2	5.4
CN ₃	10	16.0	14.0	18.7	1.4	0.5	9.0
pHw ₁	20	4.4	4.2	4.8	0.2	0.0	4.1
pHw ₂	20	4.5	4.3	4.9	0.1	0.0	3.1
pHw ₃	10	4.6	4.4	4.7	0.1	0.0	2.5
BD1 (g cm ⁻³)	20	0.83	0.44	1.20	0.19	0.04	22.44
BD2 (g cm ⁻³)	20	1.17	0.95	1.40	0.12	0.03	9.89
SOC _{OL} (kg m ⁻²)	20	0.38	0.14	0.71	0.18	0.04	47.28
SOC _{OF} (kg m ⁻²)	19	0.06	0.01	0.19	0.05	0.01	79.85
SOC _{OH} (kg m ⁻²)	4	0.39	0.12	0.72	0.26	0.13	65.71
SOCO (kg m ⁻²)	20	0.52	0.16	1.14	0.26	0.06	51.28
SOC _{stock} (kg m ⁻²)	20	8.31	4.6	12.71	2.83	0.63	30.10
Pav (mg kg ⁻¹)	20	14.07	4.71	22.07	5.48	1.22	38.92
Rock fragm ₁ (%)	20	-	-	-	-	-	-
Rock fragm ₂ (%)	20	-	-	-	-	-	-
Sand ₂ (g kg ⁻¹)	20	684	537	821	73	16	11
Silt ₂ (g kg ⁻¹)	20	242	128	342	61	14	25
Clay ₂ (g kg ⁻¹)	20	74	43	122	24	5	33
Sand ₃ (g kg ⁻¹)	10	715	652	756	38	12	5
Silt ₃ (g kg ⁻¹)	10	219	185	267	29	9	13
Clay ₃ (g kg ⁻¹)	10	65	49	81	12	4	18

MF (mixed forest)

Table 5 Mixed model results: estimates and standard error of the fixed effects. The response variables are the properties of organic horizons and mineral layers; the fixed effect is forest types: MF and ROF

Variable	Residual model	Site	Statistic	Intercept	MF	ROF	Variable	Residual model	Site	Statistic	Intercept	MF	ROF	
OC _{OL}	Non-spatial	BV	Estimate	0.6534	-0.1486	0	pH _{w,3}	Non-spatial	BV	Estimate	1.3018	-0.5318	0	
			SE	0.0439	0.0613	.				SE	1.0067	1.9710	.	
	Non-spatial	BG	Pr > t	<.0001	<.0001	.	*	Non-spatial	BG	Pr > t	Pr > t	0.2100	0.7900	.
			Estimate	0.5838	-0.2065	0	Estimate			4.6222	-0.0222	0		
OC _{OF}	Non-spatial	BV	SE	0.0415	0.0651	.	*	Sp. (Sph)	BV	SE	0.0351	0.0531	.	
			Pr > t	<.0001	0.0027	.	Pr > t			<.0001	0.6821	.		
	Non-spatial	BG	Estimate	0.2796	-0.0517	0	CN ₁	Sp. (Sph)	BG	Estimate	15.7999	-2.3944	0	
			SE	0.0486	0.0778	.	SE			0.5393	0.7479	.		
OC _O	Non-spatial	BG	Pr > t	<.0001	0.5136	.	*	Sp. (Sph)	BG	Pr > t	<.0001	0.0086	.	
			Estimate	0.3855	-0.9739	0	Estimate			16.4088	-0.7521	0		
	Non-spatial	BV	SE	0.1610	0.2558	.	*	Sp. (Sph)	BV	SE	0.2595	0.4062	.	
			Pr > t	0.0207	0.0004	.	Pr > t			<.0001	0.0704	.		
SOC ₁	Non-spatial	BG	Estimate	0.8998	-0.2925	0	CN ₂	Non-spatial	BG	Estimate	16.3066	-3.2997	0	
			SE	0.0672	0.09384	.	SE			0.7566	1.0508	.		
	Non-spatial	BV	Pr > t	<.0001	0.0035	.	*	Non-spatial	BV	Pr > t	<.0001	0.0098	.	
			Estimate	0.5893	-1.4438	.	Estimate			16.0981	-0.3535	0		
SOC ₂	Non-spatial	BG	SE	0.1262	0.1976	.	*	Non-spatial	BG	SE	0.2496	0.3906	.	
			Pr > t	<.0001	<.0001	.	Pr > t			<.0001	0.3701	.		
	Non-spatial	BV	Estimate	4.5447	1.7487	0	CN ₃	Non-spatial	BV	Estimate	14.1472	-1.1819	0	
			SE	0.8135	1.1601	.	SE			0.5447	0.8613	.		
SOC ₃	Non-spatial	BG	Pr > t	0.0004	0.1714	.	*	Non-spatial	BG	Pr > t	<.0001	0.1809	.	
			Estimate	5.5028	-0.6568	0	Estimate			15.9222	0.0920	0		
	Non-spatial	BV	SE	0.3338	0.5226	.	*	Non-spatial	BV	SE	0.6984	1.0559	.	
			Pr > t	<.0001	0.2150	.	Pr > t			<.0001	0.9318	.		
SOC ₃	Non-spatial	BG	Estimate	2.6605	0.4369	0	Pav	Non-spatial	BG	Estimate	11.5621	-0.7432	0	
			SE	0.2131	0.2975	.	SE			1.2848	1.7710	.		
	Non-spatial	BV	Pr > t	<.0001	0.1504	.	*	Non-spatial	BV	Pr > t	<.0001	0.6772	.	
			Estimate	2.1293	-0.5851	0	Estimate			16.6067	-2.5337	0		
SOC ₃	Non-spatial	BG	SE	0.0693	0.1086	.	*	Non-spatial	BG	SE	1.0558	1.6526	.	
			Pr > t	<.0001	<.0001	.	Pr > t			<.0001	0.1319	.		
	Non-spatial	BV	Estimate	1.0882	-0.1119	0	BD	Non-spatial	BV	Estimate	0.8784	-0.1638	0	
			SE	0.1148	0.1583	.	SE			0.0347	0.0479	.		
SOC ₃	Non-spatial	BG	Pr > t	<.0001	0.4840	.	*	Non-spatial	BG	Pr > t	<.0001	0.0016	.	
			Estimate	1.1722	-0.1179	0	Estimate			0.8424	-0.0128	0		
	Non-spatial	BV	SE	0.0478	0.0722	.	*	Non-spatial	BV	SE	0.0443	0.0693	.	
			Pr > t	<.0001	<.0001	.	Pr > t			<.0001	0.0016	.		

Table 5 (continued)

Variable	Residual model	Site	Statistic	Intercept	MF	ROF	Variable	Residual model	Site	Statistic	Intercept	MF	ROF
SOC _{stock}	Non-spatial	BV	Pr > t	< .0001	0.1250	.	BD	Non-spatial	BV	Pr > t	< .0001	0.8536	.
			Estimate	10.9149	0.9134	0				Estimate	1.0822	0.0819	0
			SE	0.6688	0.9340	.				SE	0.0488	0.0673	.
pH _{w1}	Sp. (Sph)	BG	Pr > t	< .0001	0.3345	.	g	Non-spatial	BG	Pr > t	< .0001	0.2319	.
			Estimate	12.0364	- 3.7203	0				Estimate	1.1673	0.0054	0
			SE	1.3691	1.5879	.				SE	0.0348	0.0545	.
pH _{w2}	Non-spatial	BV	Pr > t	0.0056	0.0350	.	g	Non-spatial	BG	Pr > t	< .0001	0.9214	.
			Estimate	- 0.6133	1.2496	0				Estimate	1.1673	0.0054	0
			SE	0.1777	0.2480	.				SE	0.0348	0.0545	.
pH _{w2}	Sp. (Sph)	BG	Pr > t	0.0015	< .0001	.	g	Non-spatial	BG	Pr > t	< .0001	0.9214	.
			estimate	4.1371	0.2824	0				estimate	4.1371	0.2824	0
			SE	0.0877	0.1183	.				SE	0.0877	0.1183	.
pH _{w2}	Non-spatial	BV	Pr > t	< .0001	0.0376	.	g	Non-spatial	BG	Pr > t	< .0001	0.9214	.
			Estimate	- 0.4729	1.3076	0				Estimate	1.1673	0.0054	0
			SE	0.4449	0.6607	.				SE	0.0348	0.0545	.
pH _{w2}	Non-spatial	NF	Pr > t	0.3547	0.1245	.	g	Non-spatial	BG	Pr > t	< .0001	0.9214	.
			Estimate	4.5483	- 0.0082	0				Estimate	1.1673	0.0054	0
			SE	0.0265	0.0415	.				SE	0.0348	0.0545	.
pH _{w2}	Non-spatial	NF	Pr > t	< .0001	0.8430	.	g	Non-spatial	BG	Pr > t	< .0001	0.9214	.
			Estimate	4.5483	- 0.0082	0				Estimate	1.1673	0.0054	0

Sp. (Sph), the spatial covariance function of residuals was the Spherical model; non-spatial, the residuals were spatially uncorrelated

* $p < 0.05$ and + $p < 0.1$ denote statistically significant differences between forest types; g defines Gaussian variable

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Data availability The datasets generated and analyzed during the current study are available in the Pangaea repository (Ferré and Comolli 2019) at <https://doi.pangaea.de/10.1594/PANGAEA.905854>.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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