Responses of soil quality indicators to innovative and traditional thinning in a beech (Fagus sylvatica L.) forest

Manuscript Details

Manuscript number	FORECO_2019_1821
Title	Responses of soil quality indicators to innovative and traditional thinning in a beech (Fagus sylvatica L.) forest
Article type	Full Length Article

Abstract

Soil has a pivotal role in keeping high the productivity of forest ecosystem but its physical and chemical properties are highly influenced by changes in forest species composition and forest management practices. Thinning is the most effective silvicultural practice used in Europe to increase the ecological and economic value of forest stands. In this study, biological indicators were used to assess the effects of innovative (T3) and traditional (T2) thinning on soil properties with respect to unmanaged forest (T1), because forest practices are among the main causes of soil fertility and biodiversity loss. The aim was to identify the most appropriate forestry practices to sustainably manage beech (Fagus sylvatica L.) forest. Results showed that T2 had the highest dissolved organic carbon (DOC) and the lowest water content (WC), organic carbon (OC) and nitrogen (N) amount. The humification index was the highest in T2 as well as fungi, ergosterol fluorescein diacetate hydrolase (FDA) and catalase. The highest values of NO3-, SO4--, Na+, Mg2+ and Ca2+ were found in T2. QBS-ar was significantly higher in T2 and T3 than in T1. In short, our results evidenced that organic matter, total nitrogen, C/N ratio and water content cannot be considered alone or in combination indices of quality to evaluate the effect of thinning on soils. Rather, is the data crossing of microbiota and ions with organic matter fractions (stable and labile) that can give important and accurate information on how thinning can affect soil biological properties that are strictly correlated to soil fertility and quality.

Keywords	Biodiversity, QBS-ar, thinning intensities, soil biological indicators

Corresponding Author	A. Muscolo
Corresponding Author's Institution	Mediterranea University
Order of Authors	Federico Romeo, Giovanna Settineri, Maria Sidari, Carmelo Mallamaci, A. Muscolo

Suggested reviewers	PAOLO PORTO, Diego Pizzeghello, María-Belén Turrión, Miroslava Mitrovic

Submission Files Included in this PDF

File Name [File Type]

Cover letter FORECO 2019.docx [Cover Letter]

Highlights FORECO THINNING.docx [Highlights]

graphycal abstract FORECO THINNING.docx [Graphical Abstract]

Responses of soil quality indicators Romeo et al FORECO.docx [Manuscript File]

To view all the submission files, including those not included in the PDF, click on the manuscript title on your EVISE Homepage, then click 'Download zip file'.

Reggio Calabria, 03-10- 2019

Dear Editor-in-Chief,

I am sending you our manuscript entitled "**Responses of soil quality indicators to innovative and traditional thinning in a beech (*Fagus sylvatica* L.) forest.**" by Romeo et al., which we would like to submit for publication in **Forest Ecology and Management**. The aim of this work was to individuate early warning indicators of soil quality to be applied for identifying the most appropriate forestry practices to manage a beech (*Fagus sylvatica*) forest in a sustainable way. Thus, we assessed the effect of two different thinning practices (traditional and innovative) on soil properties in respect to unmanaged forest. Soil organic matter, microbial biomass C, fungal biomass C, water soluble phenols (WSP), fluorescein diacetate hydrolase, dehydrogenase, catalase, ergosterol, humification indices, QBS-ar and micro-arthropod groups have been used to asses soil quality. Results showed that traditional thinning had the highest dissolved organic carbon and the lowest water content, organic carbon and nitrogen amount. The humification index was the highest in traditional thinning as well as fungi, ergosterol, fluorescein diacetate hydrolase and catalase. The highest amount of ions were found in the traditional thinning. QBS-ar was significantly higher in both thinned area than in unmanaged area. In short, our results evidenced that organic carbon, nitrogen and microbial biomass, cannot be considered alone or in combination indices of quality to evaluate the effect of thinning on soils. Rather, is the data crossing of microbiota and ions with organic matter fractions (stable and labile) that can give important and accurate information on how thinning can affect soil biological properties that are strictly correlated to soil fertility and quality.

We confirm that this manuscript has not been published elsewhere and is not under consideration by another journal. All authors have approved the manuscript and agree with its submission to **Forest Ecology and Management.**

We look forward to hearing from you at your earliest convenience.

With my best regards,
Sincerely yours,
Prof.ssa Adele Muscolo

Please address all correspondence to:

Prof. Adele Muscolo

Dipartimento di Agraria
Università degli Studi “Mediterranea” di Reggio Calabria
Feo di Vito 89124 Reggio Calabria ITALY
Tel: 0965/1694364
Fax: 0965/1694550
e-mail: amuscolo@unirc.it

Highlights

- Biological indicators were used to assess the effects of thinning on soil properties.
- Thinning decreased organic C and water content in a *Fagus sylvatica* dominated-soil.
- QBS-ar was significantly higher in the thinned soils than in the control area
- High thinning intensity reduced the activity of soil enzymes and MBC
- The highest values of ions were found in the traditional thinning

Improved

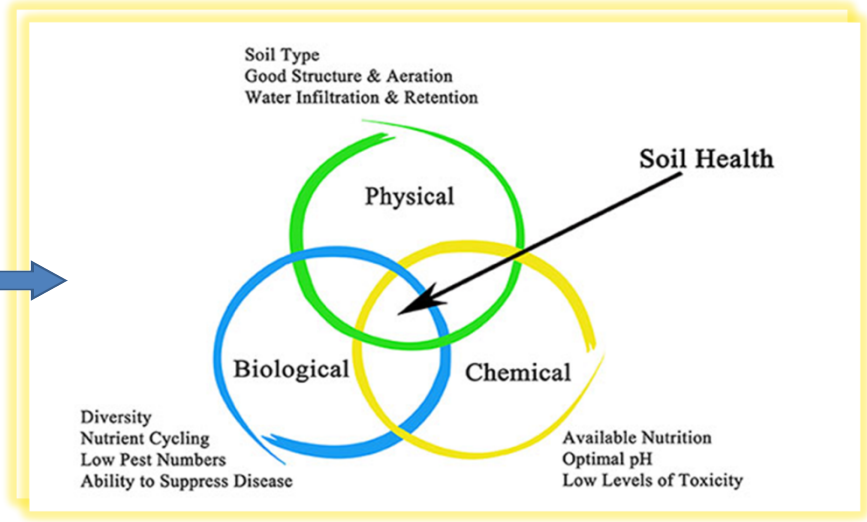
Organic carbon, nitrogen, water content, phenols, C/N ratio, MBC

Increased

DOC, nutrients FBC, enzymes, HI, QBS-ar

decreased

Organic carbon, nitrogen,water content, phenols, C/N ratio, MBC,DOC, nutrients FBC, enzymes, HI



Responses of soil quality indicators to innovative and traditional thinning in a beech (*Fagus sylvatica* L.) forest

Federico Romeo, Giovanna Settineri, Maria Sidari, Carmelo Mallamaci and Adele Muscolo*

Department of AGRARIA - Mediterranea University, Feo di Vito, 89122 Reggio Calabria Italy

* Corresponding author. Tel: +39-1694364, Fax: +39-1694550, e-mail: amuscolo@unirc.it

Abstract

Soil has a pivotal role in keeping high the productivity of forest ecosystem but its physical and chemical properties are highly influenced by changes in forest species composition and forest management practices. Thinning is the most effective silvicultural practice used in Europe to increase the ecological and economic value of forest stands. In this study, biological indicators were used to assess the effects of innovative (T3) and traditional (T2) thinning on soil properties with respect to unmanaged forest (T1), because forest practices are among the main causes of soil fertility and biodiversity loss. The aim was to identify the most appropriate forestry practices to sustainably manage beech (*Fagus sylvatica* L.) forest. Results showed that T2 had the highest dissolved organic carbon (DOC) and the lowest water content (WC), organic carbon (OC) and nitrogen (N) amount. The humification index was the highest in T2 as well as fungi, ergosterol fluorescein diacetate hydrolase (FDA) and catalase. The highest values of NO_3^-, $SO4^{--}$, Na^+, Mg^{2+} and Ca^{2+} were found in T2. QBS-ar was significantly higher in T2 and T3 than in T1. In short, our results evidenced that organic matter, total nitrogen, C/N ratio and water content cannot be considered alone or in combination indices of quality to evaluate the effect of thinning on soils. Rather, is the data crossing of microbiota and ions with organic matter fractions (stable and labile)

21 that can give important and accurate information on how thinning can affect soil biological
22 properties that are strictly correlated to soil fertility and quality.

23

24 *Keywords:* Biodiversity, QBS-ar, thinning intensities, soil biological indicators

25

26

27 **1. Introduction**

28 Forests provide crucial services for human well-being and economic development. In addition to
29 wood and fiber, they provide numerous non-wood products such as food, freshwater, and fuel, or
30 services as climate and diseases regulation, recreation and preservation of biodiversity, driving the
31 sustainable growth (IUCN). Now a days, a prominent challenge is how to manage forests for timber
32 and bioenergy production maintaining, at the same time, long-term conservation/implementation of
33 the forest ecosystem functioning. Soil has a pivotal role in keeping high the productivity of forest
34 ecosystem but its physical and chemical properties are highly influenced by changes in forest stand
35 structure, density and composition of species which correlates with thinning intensity, interval, and
36 method. The reduction in tree density due to thinning is reported to alter microclimate (Masyagina
37 et al., 2006; Trentini et al., 2017), increasing soil temperature and decreasing soil humidity (Ma et
38 al., 2010) but also to affect soil properties, increasing compaction (Elliot et al., 1998; Picchio et al.,
39 2012; Marchi et al., 2014), soil bulk density and decreasing water holding capacity and nutrients.
40 Thinning can also affect organic matter (Smolander et al., 2013) which govern most of the soil
41 properties, and hence soil quality and health. Soil organic matter (SOM) changes in a short time are
42 difficult to measure directly because they don't vary only on the basis of flux by which carbon enter
43 or leaves soil ecosystem but also for the flux of carbon that occurs in the soil ecosystem (Chapin III
44 et al., 2002), total SOM by itself is thus not able to highlight changes in soil ecosystem functioning.
45 Organic matter contains numerous fractions (phenolics, microorganisms, enzymes) each with a

different residence time and different functional roles in soil (Zagal et al., 2009) which singularly could give a measure of subtle, or early changes in soil quality. Organic matter fractions can be considered as fine indicators of soil quality that influence soil function in specific ways and that are much more sensitive to changes in soil management practices (Muscolo et al., 2014). Generally, changes in SOM fractions can sensitively respond to changes in plant vegetation, climate and land use in agroforestry ecosystems (Schmidt et al., 2011; Wang and Wang, 2011; Wagg et al., 2014). Muscolo et al. (2014) identified organic matter fractions such as microbial biomass carbon (MBC), water soluble phenols (WSP), and fluorescein diacetate hydrolase (FDA), as effective tools in the evaluation of soil quality changes, in the short term, in a coniferous stand of southern Italy. Additionally, Muscolo et al. (2015) showed in *Pinus laricio* Poiret ssp. *calabrica*, *Abies alba* Mill. and *Fagus sylvatica* stands in Calabrian Apennines, southern Italy, that these three indicators reflected soil quality change due to different factors. MBC primarily showed variation related to vegetation, FDA evidenced changes caused by climatic factors, and WSP was influenced by changes related to soil depth. Thinning might have a great impact on the different fractions of SOM in the soil. Recently, there were a number of studies on the effects of thinning on soil respiration and SOM. Settineri et al. (2018), studying the effects of different thinning intensities on soil carbon stock showed that high intensity thinning was the best silvicultural practice-approach method to manage *Pinus laricio* forest for increasing soil carbon storage. Kim et al. (2019) highlighted the interaction between soil properties and microbes evidencing that inconsistent thinning effects on soil properties changed microbial biomass and enzymes in thinned oak and larch forest ecosystems. Zhang et al. (2018) analyzing 53 published studies on forest thinning, evidenced that thinning did not significantly change total carbon, and MBC, but enhanced soil temperature, soil total nitrogen and decreased the soil C/N ratio. Since there is still little information on the effect of thinning on organic matter fractions, further studies on the variations in soil organic matter fractions and soil biodiversity are thus crucial for evaluating the stability and the sustainability of forest production following a thinning treatment. In Italy, thinning of beech forests represent the most used

silvicultural treatment to enhance the economic value of these stands maintaining biodiversity (Lombardi et al., 2018). Therefore, this research aims to understand how thinning affects organic matter and each of its component pools in beech forest ecosystem. The objectives of this study were to: (1) compare the effects of two different thinning practices (traditional and innovative) to unmanaged beech (*Fagus sylvatica*) forest; (2) determine the variation of soil organic matter fractions under different thinning practices, to identify which type of forest management practice was more sustainable for maintaining/improving soil quality in Calabrian beech forests.

2. Material and methods

2.1. Study area

The study area is located in the Marchesale Biogenetic Reserve (Natura 2000 site) within the highest slope of the Calabrian "Serre" mountains, in Mongiana (VV) (38° 30’ N, 16° 14’ E). The entire reserve, that covers 1234 hectares, is managed by the National Forest Service of Italy and consists mainly of 75-year-old high beech forest *Fagus sylvatica*.

Being located in the upper part of the mountain system facing the Tyrrhenian Sea (1100 m a.s.l.) and exposed to North-West, its microclimate is influenced by the interception of fog, wet winds and precipitation (Becagli et al., 2013). The mean values of annual rainfall and temperature, based on the data obtained at Mongiana (VV) meteorological station (920 m a.s.l.) and available for the period 1928-2018, are respectively of 1801.5 mm e 11.4 °C. The warmest month is August (18.6 °C), the coldest one is January (3.8 °C). The climate is typically Mediterranean (Csb, sensu Koppen, 1936).

This area is geologically characterized by Paleozoic granitoid deeply fractured and with deep, versus shaped slopes (Conforti et al., 2015). According to USDA (soil classification), the predominant soils are Inceptisols and Entisols (Soil Survey Staff, 2010).

96 Between 2012 and 2013, an experimental forest management was carried out on a surface of 30
97 hectares of the entire reserve, chosen subsequently as the object of our study. In this area three
98 different sub-areas of about 3 ha each have been identified for each different silvicultural thinning
99 (3 treatments x 3 replicates). Specifically, an unmanaged area for over 30 years (T1) was used as
100 control; a traditional treatment (T2) and an innovative treatment (T3) have been identified.
101 Traditional treatment was a thinning from below with a moderate intensity which removed all the
102 dominated trees and the worst dominant trees (on average, ca. 12% of total volume resected). The
103 innovative treatment was oriented to retain the 50 best trees per hectare and improve the structural
104 biodiversity, collecting 5 or 6 trees closer to them, regardless of their social position (on average,
105 ca. 27% of the total volume removed). No significant differences were found between the
106 dendrometric parameters in these areas before the silvicultural interventions (Picchio et al., 2016).
107 The three treatment sites are fully comparable in terms of slope, orientation and soil types.
108
109 *2.2. Sampling procedure*
110 For each of the three treatment sites (T1, T2 and T3), 3 representative plots (1000 m^2 in size) were
111 established for sampling (3 treatments x 3 plots = 9 plots). The soil sampling campaign was carried
112 out in May 2018, and consisted of collecting soil cores for chemical-physical and biological
113 analysis. The cores were collected in areas with similar slope using a 10-cm-diameter steel core
114 tube inserted up to a depth of ca. 35 cm.
115 Each soil core (9 in total) used for the chemical-physical and biological analyses, was divided into 2
116 layers (0-15 and 15-30 cm). The corresponding layers were merged in order to obtain 3 final
117 representative cores (one for each treatment site) and all analysis made tripled. Separate samples
118 (10 cm depth), following the same scheme, were performed to establish the micro-arthropods QBS-
119 ar index. All the samples were air dried and sieved to separate the < 2 mm fraction, except for
120 microbial biomass, QBS-ar (fresh soil) and water content (oven drying).
121

122 *2.3. Soil chemical and biochemical analysis*

123 Soil water content (WC) was determined within 24 h of samples collection with a difference
124 between fresh and dry weight after oven drying at 65°C for 72 hours; pH was measured in distilled
125 water (soil solution ratio 1:2.5) with a glass electrode.

126 Total water-soluble phenols (WSP) were determined, after water extraction, with Folin–Ciocalteu
127 reagent according to Box (1983). Tannic acid was used as standard and the concentration of water-
128 soluble phenolic compounds was expressed as tannic acid equivalents (μg TAE g^{-1} dry soil).

129 Total soil organic carbon (TOC) was determined according to Springer and Klee (1954). The
130 content of organic carbon (OC) was calculated by back-titration with a solution of 0.2 N $FeSO_4$.
131 Total nitrogen (N) was measured by the Kjeldahl method (1883).

132 Dissolved organic carbon (DOC) was analysed with a visible spectrophotometer (Agilent-8453) at
133 254 nm according to the method described in Brandstetter et al. (1996).

134 Humification index (HI), degree of humification (DH%) and humification rate (HR%) have been
135 detected. In short, humic components were extracted with a solution 0.1 M of sodium
136 pyrophosphate and sodium hydroxide (Official Methods of Soil Chemical Analysis, 1994). TEC
137 was fractioned into humified (humic acid, HA + fulvic acid, FA) and non-humified (NH) fractions
138 (Ciavatta et al., 1990). Humification parameters have been calculated as shown in Eqs. 1–3
139 following the method proposed by Ciavatta et al. (1990) and Sidari et al. (2005).

140 $$HI = NH / (HA + FA) \quad (1)$$

141 $$HR\% = 100(HA + FA) / TOC \quad (2)$$

142 $$DH\% = 100(HA + FA) / TEC \quad (3)$$

143 Humification index (HI) represents the ratio between not humified and humified extracted carbon,
144 while HR is the percent of humification rate and DH is the percent of humified carbon in the
145 extract.

146 The fungal cell membrane component ergosterol (Erg) was extracted following the method
147 described by Gong et al. (2001) with some modifications, 10 g of methanol (Me-OH) was added to

1 g of soil. The suspension was homogenized with mechanical agitation for 15 min, and centrifuged at 3500 rpm for 15 min. The supernatant was filtered using a syringe membrane filter (4 mm, 0.45 μm polytetrafluoroethylene (PTFE) and then kept in the dark until analysis with an Agilent Technologies Infinity 1290 high performance liquid chromatography (HPLC) system (Agilent Technologies, Santa Clara, California, USA). Soil samples were extracted with bidistilled water (ratio soil/water 1:10) (Wang et al., 2013) for 24 h at 25 °C to detect ion concentration by using a chromatography systems (Dionex ICS-1100).

2.4. Fungal and Microbial biomass C

MBC was determined in fresh soil samples by the chloroform fumigation-extraction method (Vance et al., 1987). The filtered soil extracts of both fumigated and non-fumigated samples were analyzed for soluble organic C using the method of Walkley and Black (1934). The estimation of the MBC was made on the basis of the differences between the fumigated and non-fumigated soil, and an extraction efficiency coefficient of 0.38 was used to convert soluble C in biomass (Vance et al., 1987). As reported by Montgomery et al. (2000), measurement of ergosterol concentration in the soil is useful for estimating the soil fungal biomass carbon (FBC) content. Following the method proposed by Montgomery et al. (2000) it was possible to determine the content of living FBC starting from measurements of ergosterol content in the soil. Several mycelial carpets have been studied in different types of soil, and ergosterol transformed by the following formula into FB (fungal biomass):

$$FB\ (\mu g\ g^{-1}\ soil) = Ergosterol\ (\mu g\ g^{-1}\ soil)\ x\ f\ x\ Rf \qquad (4)$$

where $f = 250$ (1/4 x 1000, mg biomass μg^{-1} ergosterol), and $Rf = 1.61$ (correction factor for average percent recovery, 1/0.62).

FB can also be expressed in terms of the C content of the mycelial mats (Montgomery et al., 2000):

$$FB - C = FB \times C \qquad (5)$$

where FB-C = fungal biomass-carbon, and C = 0.43, average C content detected in fungal species.

174

175 *2.5. Enzymatic assay*

176 Fluorescein dyacetate hydrolase (FDA) hydrolysis reaction was determined according to the
177 methods of Adam and Duncan (2001). Briefly to 2 g of soil (fresh weight, sieved <2mm) was added
178 15 mL of 60 mM potassium phosphate pH 7.6 and 0.2 mL 1000 mg FDA mL^{-1} and then placed in
179 an orbital incubator at 30,8°C for 20 min. Once removed from the incubator 15 mL of chloroform/
180 methanol (2:1, v/v) was added to terminate the reaction. After this, the content of the flask was
181 centrifuged at 2000 rpm for 3 min. The supernatant was filtered and measured at 490 nm on a
182 spectrophotometer (Shimadzu UV–vis 2100, Japan).

183 Dehydrogenase (DH) activity was determined by the method of von Mersi and Schinner (1991). In
184 brief, to a sample of fresh soil equivalent to 1 g of oven dried (105 °C) soil were added 1.5 ml of 1
185 M Tris–HCl buffer of pH 7.5 followed by 2 ml of 0.5% INT solution (Sigma product No I 8377),
186 and the suspension was kept at 40 °C for 1 h. Then 10 ml of extractant (methanol) were added and
187 the samples were mixed and then leaved in the dark for 10 min. Finally, the solutions were filtered
188 (Whatman's n° 40 paper), and the absorbance of the filtrate was determined at 490 nm.

189 Urease (URE) was determined using the method of Kandeler and Gerber (1988). Soil (5 g fresh
190 weight) was mixed with 2.5 ml of urea (80mM) and 20 ml 0.1 M borate buffer pH (10.0), allowed
191 to react for 2 h in an orbital shaker at 37 °C. After incubation, pipette 2.5 ml of urea to the control,
192 add 30 ml of KCl (2M) to both sample and control, and shake for 30 min. Filter the contents of the
193 flasks through folded filters. Aliquots of 1 ml of the filtered solution were mixed with 9ml of
194 distilled water, 5 ml of sodium/salicylate solution and 2 ml of dichloroisocyanuric acid (Na^+ salt).
195 The colour intensity of the solution was measured at 690 nm. Ammonium concentrations were
196 determined by using a calibration curve of ammonium chloride standard solution.

197 Catalase (CAT) activity was measured by the method of Beck (1971). Results were expressed as
198 O_2% g^{-1} soil $3min^{-1}$.

199

200 *2.6. Microarthropods*

201 Micro-arthropods, included mites and collembola, were analyzed with Berlese-Tullgren selector
202 (Parisi et al., 2005). One soil samples were taken from each site differently managed, including the
203 control site. Each plot was sampled at soil depths of 0-10 cm. Immediately upon returning from the
204 field, the soil samples were transferred to Berlese-Tullgren funnels lined with 4 mm wire mesh.
205 Arthropods were extracted for 7 days and collected in a beaker filled with preservative liquid (2
206 parts 75% ethanol and 1 part glycerol) beneath the funnel.

207 All the arthropods were identified at different taxonomical levels through, the determination of
208 biological forms and calculation of QBS index, using a microscope PCE-MM200. Accordingly, the
209 biological form (morpho-type) that is most adapted to soil was identified. Each morpho-type
210 correspond an ecomorphological index (EMI); as a rule, eu-edaphic (i.e. deep soil living) forms
211 correspond to an EMI=20, hemi-edaphic (i.e. intermediate) forms have an index rating
212 proportionate to their degree of specialization, while epi-edaphic (surface living) have an EMI = 1.
213 The QBS index value was obtained from the sum of EMI index of all the collected groups (*Acari,*
214 *Collembola, Areneae, Diptera, Hemiptera, Coleoptera, Hymenoptera, Protura, Diplura,*
215 *Diplopoda*). If in a group biological forms with different EMI values are present, the higher value
216 (more adapted to the soil form) was selected to represent the group in the QBS calculation (Parisi et
217 al., 2005). EMI is a simplified index, that use the microartopod morphology assessment for
218 generating Soil Biological Quality index (QBS index). This analysis allows assessing the
219 degradation level of soils (Parisi et al., 2005).

220

221 *2.7. Statistical analyses*

222 To test the relationships among soil parameters at two soil depths and for three different
223 silvicultural treatments, datasets were analyzed using Principal Component Analysis (PCA),
224 Multivariate Analysis of Variance (MANOVA) and T test for paired values. The results are
225 summarized in an ordination diagram. PCA was carried out using the soil parameters in plots under

different silvicultural treatments using the software PAST (Hammer et al., 2001). Because the data are expressed in different units, the results are standardized with the following formula:

$$z = \frac{(x_i - \overline{x})}{SD} \quad (6)$$

where x_i is the individual value of each parameter, $\overline{x}$ is the mean and SD the standard deviation.

A MANOVA analysis was carried out for evaluating the effects of thinning, soil depth and their interaction on the set of soil parameters. Finally, since there are only two soil depths (0-15 cm and 15-30 cm), a T test was used for paired values to evaluate significant differences. This last analysis allowed us to verify if thinning affected soil properties according to depth, and if the impacts were similar. Pearson's correlations for both soil layers and all soil parameters, were carried out using PAST software (Hammer et al., 2001).

One way ANOVA and t-test was carried out to determine statistical difference for QBS-ar among the treatments. Anova, Manova models and t-test were carried out using SPSS software (IBM Corp., 2012).

3. Results

3.1. Soil chemical and biochemical features

All the soils analyzed belong to the sandy-loam textural class, with 25% silt, 13% clay and 62% sand (data not shown). Soil texture did not change over treatments (data not shown). Water content (WC) decreased in both layers (Table 1) following tree thinning intensity. In the first layer of soil (0-15 cm) WSP, OC, N, C/N decreased in T2 and much more in T3 in comparison to T1. DOC decreased only in the innovative thinning. In the underlying layer a similar trend was observed for WC (Table 1). T3 and T2 showed the lowest WSP. T2 showed the highest DOC amount and the lowest OC and N amount, while C/N ratio did not show significant change among the treatments and control (Table 1). The humification degree did not change in the 0-15 cm between the treatments and control (Table 2). Humification rate was the highest in T3 while the humification

index was the highest in T2 (Table 2). MBC decreased in T2 (-40%) and T3 (-51%) in respect to control. Ergosterol and FBC were the highest in T2 and they were almost double than T3. Regarding the enzymatic analysis, FDA in T3 was half than T1 and T2. DHA was instead greater in T3 than T1 and T2. CAT was the highest in T2 followed by T1 and T3. URE was the lowest in T3 and the highest in T1. In the 15-30 cm layer DH, HI and HR had a trend similar to those observed in 0-15 cm layer. MBC was always higher in T1 than T2 and T3. FBC and Erg were also the highest in T2. FDA was always the highest in T2. FDA was less than half in T1 in respect to the same treatment in the upper layer. DHA did not show significant variation among the treatments and control. Catalase and urease maintained the same trend in the upper and lower layers, and it was higher in T2 than T3 (Table 2).

For PCA analysis the first two components (Eigenvalues >1) have been extracted. The variance was higher at 0-15 cm (94.6 %), than at 15-30 cm (91 %). At both depths, the component 1 explained about 55 %, while the component 2 explained about 35 % of the variability in all parameters (Fig. 1). PCA diagram for 0-15 cm showed that T3 influenced only DHA, DH and HR while T1 and T2 were similar in fitting with all the other parameters. At 15-30 cm T3 influenced only HR, T2 influenced mainly the enzyme activities, Erg, FBC and DOC. All the other soil parameters were equally distributed in T2 and T1 (Fig. 1). Soil under traditional thinning, in respect to control and innovative thinning, showed a greater CAT, Erg, FBC and HI in both layers (Table 3). Still in the T2 area, FDA activity was the highest in both soil layers (Table 2). These assumptions are clearly evident in Fig. 1, where the PCA showed these values significantly correlated to T2 area and placed in the same quadrants. On the contrary, soil under innovative thinning, in both layers, (located in the quadrant with both negative components) showed only a greater humification rate. T3 positively influenced DHA in the upper layer and DH, MBC and OC in the lower layer (Fig. 1a,b). Lowest values of WSP, OC, DOC, N, C/N, Erg, MBC, FDA, CAT and URE, were detected in the upper layer under innovative thinning (located in opposite quadrants in Fig. 1a, Table 1,2). In the 15-30 cm DOC, N, DH, HR, HI, CAT and URE maintained the same trend of the upper layer for all the

277 treatments (Table 1,2). OC, C/N ratio and WSP increased in T3 in respect to its own values detected
278 in the upper layer. A strong decrease in MBC, Erg and FBC content was instead observed (Table 2).
279 In the underlying layer, FDA, URE and CAT decreased and DHA increased in T1 and T2, while all
280 these parameters decreased in T3 in respect to 0-15 cm layer (Table 2). OC in both layer and in all
281 treatments resulted positively correlated to WC, WSP, N, and MBC (Table 4).
282 FDA, C/N and URE were positively correlated to OC only in the first layer (Table 4). FBC was not
283 correlated to OC, but showed significant and positive correlation in both layer with DOC, HI, ERG,
284 CAT and NO_3^- (table 4).
285
286 *3.2. Soil ions*
287 In the surface layer, the highest values of NO_3^-, SO_4^-, Na^+, Mg^{2+} and Ca^{2+} were measured in the T2
288 area (Fig. 2a). The only one to be positively influenced by the innovative thinning was K^+ (Fig. 1a-
289 b**),** while chloride (Cl^-), NO_3^- and Na^+ decreased in T3 (0-15 cm) (Fig. 2a). In the 15-30 soil layer,
290 ions showed a general decrease in values, Na^+, Mg^{2+} and Ca^2 were higher in T1 and T2 in respect to
291 T3 (Fig. 2b). K^+ did not show significant differences between treatments and control, SO_4^- and Cl^-
292 were higher in T1 than T2 and T3, NO_3^- was the highest in T2 (Fig. 2b). The concentration of NH_4^+
293 in soil was below the detection limit.
294 PCA diagram for 0-15 cm showed that T3 influenced only K^+, while T1 and T2 similarly fitted with
295 all the other parameters. At 15-30 cm T2 influenced mainly K^+ and NO_3^- (Fig. 1b). It was also
296 evident from the PCA analysis how the innovative area was the only one with the lowest ion related
297 parameters (Fig. 1a-b). In fact, except for K^+ in the surface layer (Fig. 2a), all the parameters were
298 completely translated in the quadrants opposite to T3 (always with negative components).
299
300 *3.3. Microarthropods features*
301 QBS-ar was significantly higher in the T2 and T3 than in the control area (Fig. 3). This is certainly
302 due to microclimatic properties, solar irradiation and different presence of herbaceous vegetation

303 between the examined areas. Regarding the micro-arthropod groups a different distribution in soil
304 differently managed was found. Among the 16 species found (Table 3), in T1 9 species has been
305 observed. Chilopoda, thysanoptera, protura, diplura, lepidoptera, araneae, formicidae and isopoda
306 were totally absent (Table 3). In T2 14 species were present. The species that missed in soil under
307 traditional thinning were blattaria and diplura. In T3 13 species were present, blattaria, protura and
308 formicidae were missed. The greatest biodiversity was found in T2 in respect to all the other
309 treatments.

310

311 **4. Discussion**

312 Soil quality reflects the capacity of a soil to promote biological productivity, plant and animal
313 health, and to maintain environmental quality (Doran and Parkin, 1994). In agro and forestry-
314 ecosystems the assessment of soil quality through biological indicators allows early evidence of
315 changes in soil physical and/or biological characteristics determined also due to forest management.
316 Roscoe and Buurman (2003) quantified the effects of forest management on soil estimating total
317 organic carbon, but as subsequently demonstrated forest management affect not only the organic
318 carbon storage (Lal, 2006), but also nutrient concentrations (Ashagrie et al., 2007), soil enzyme
319 activities (Kim et al., 2019), soil biodiversity (Lukac, 2017) and water retention (Resck et al., 2008).
320 Thus testing only the changes in SOM would be reductive, to assess instead SOM fractions (labile
321 and stable), and correlating them each others can give important information to evaluate the impact
322 that forest management can have in a short term on soil fertility. Our results demonstrated a
323 decrease in OC, C/N and labile fraction of organic matter in terms of MBC and WSP mainly in the
324 innovative thinning than traditional one in respect to unthinned forest. These results highlighted that
325 under thinning, the relative rate of decomposition increased mainly in T3, as demonstrated by the
326 greatest decrease in C/N ratio. The humification index, the ratio between not humified and humified
327 extracted carbon, and the percent of humification rate detected because they are considered key

328 indicators of the humification status of organic matter in soil (Gigliotti et al., 1999), evidenced that
329 T2 had a good balance between humification and mineralization process in respect to T1 and T3
330 where a shift to humification and mineralization process respectively were observed. HI was at
331 about 0.5 in T2, lower values indicate more humified extracts (Sidari et al., 2005). HR parameter
332 that is proportional to the state of humification of the soil organic matter, was similar in T2 and T1
333 as well as DH that is generally 100% when the extracted organic carbon is completely humified.
334 Pedofauna and microflora detected because soil ecosystems with large biodiversity and microbial
335 biomass improve soil carbon cycle more than soil ecosystem with reduced amount of microbiota (de
336 Graaff et al., 2015). Hooper et al. (2005) highlighted that taxonomic and functional compositions of
337 soil communities are effective drivers of SOM processes. Results evidenced that the abundance,
338 species richness of micro-arthropods, as well as fungi and bacteria colonies and soil properties
339 changed with the treatments. Under T2 we found the greatest biodiversity and the highest amount of
340 micro-arthropods. Because of their high abundance, specie richness, habitat fidelity (Andersen and
341 Majer, 2004), and high sensitivity to external perturbations, micro-arthropods are considered
342 important bio-indicators of environmental quality and can be used for monitoring short-term
343 changes in soil ecosystem. The diversity in number of species, found among the treatments, can
344 explain in part the differences in soil quality. The faeces of arthropods are the basis for the
345 formation of soil aggregates and humus, which physically stabilize the soil and increase its capacity
346 to store nutrients improving its quality and represent also a substrate for microbial decomposition
347 and to fostering the growth and dispersal of microbial populations (Culliney, 2013). Under T2, the
348 soils had the highest amount of fungi as well as ergosterol, but a low amount of MBC. Generally
349 high fungi amount correspond to higher ability of such soils to improve nutrient cycling (Hodge et
350 al., 2001; Smith & Smith, 2011). A shift toward a fungal dominance in the microbial community is
351 thought to enhance soil aggregation and soil nutrient interception and availability to plants. (Augè,
352 2004; Six et al., 2006). Ergosterol an important indicator of fungal growth on organic compounds
353 and mineralization activity was higher in T2 than T1 and T3 confirming the greatest presence of

354 fungi amount and activity in traditionally managed forest stand. For instance, fungi are thought to
355 express a broader suite of enzymes capable of transforming and stabilizing inputs; and fungal
356 biomass has greater C/N ratio which results in increased carbon use efficiency (Strickland and
357 Rousk, 2010; Waring et al., 2013). Intensively managed soils often exhibit lower fungal/microbial
358 biomass ratio. Our data apparently in contrast with data on OC, MBC and WSP that could suggest
359 in thinned forest a greater carbon loss with a decrease in fertility, evidenced a higher
360 fungi/microbial biomass ratio in managed areas and in particular in T2 than T1 and T3 as well as a
361 higher activity of FDA and catalase, enzymes fungal-produced, as demonstrated by correlation
362 analysis. Catalase activity is usually higher in high quality soils and its activity may decrease or
363 cease when soil pH, nutrients, or temperature extremes occur (Xun et al., 2015). Catalase is also an
364 important cellular antioxidant enzyme that defends soil against oxidative stress. The highest
365 catalase activity in T2 suggests that the traditional thinning positively affected soil quality. In T2,
366 the greatest amount of FDA, enzyme markers of hydrolytic soil activity, reliable estimator of fungal
367 biomass was found, suggesting that in this site a great decomposition transformation, and
368 mineralization of organic matter with a major nutrient release occurred. This data were also
369 confirmed by the increase in nutrients and DOC and by the contemporary decrease in SOM and
370 were in agreement with the findings of Bardgett et al. (2003), Lee and Jose (2003), Allison (2006),
371 De Deyn et al. (2009) and Billings et al. (2010), showing that microflora components have a key
372 role in soil nutrient and organic matter cycle. In short from this data crossing resulted that T2
373 improved soil quality even if the organic matter amount decreased resulting a sustainable forest
374 management practice.

375

376 **5. Conclusion**

377 In short, our results evidenced that organic matter, total nitrogen, C/N ratio and water content
378 cannot be considered alone or in combination indices of quality to evaluate the effect of thinning on

soils. Rather, is the data crossing of microbiota and ions with organic matter fractions (stable and labile) that can give important and accurate information on how thinning can affect soil biological properties that are strictly correlated to soil fertility and quality. It is by using specific and pertinent biological indicators in combination, that we can predict the dynamic behaviour of soil processes and the impact of management practices on soil quality allowing to determining the sustainability of forest management activities. In other words, the possibility to predict short-term variations in soil processes through the use of indicators represents a great advantage in the context of sustainable land management. Using biological indicators forest management activities will give clear and detailed information on triggering of soil fertility loss. Among the soil properties DOC, FDA, CAT, fungi and pedofauna have been identified as effective tools to evaluate performance and quality of managed soil in a short time.

Acknowledgments

This research was supported by "Mediterranea University" of Reggio Calabria "Programmi di Ricerca Scientifica", no outside funding was received.

Author Contributions: AM conceived and designed the experiments, analyzed the data, wrote the manuscript. FR conducted fieldwork, performed the laboratory experiments, collected the data, analyzed the data, performed statistical analyses. MS critically reviewed and edited the manuscript, GS performed the laboratory experiments, CM conducted fieldwork.

Conflict of interest statement

None declared.

403 **References**

404 Adam, G., Duncan H., 2001. Development of a sensitive and rapid method for the measurement of
405 total microbial activity using fluorescein diacetate (FDA) in a range of soils. Soil Biol.
406 Biochem. 33, 943–951.

407 Allison, S.D., 2006. Soil minerals and humic acids alter enzyme stability: implications for
408 ecosystem processes. Biogeochemistry 81 (3), 361–373.

409 Andersen, A.N., Majer, J.D., 2004. Ants show the way Down Under: invertebrates as
410 bioindicators in land management. Front. Ecol. Environ. 2 (6), 291–298.

411 Ashagrie, Y., Zech, W., Guggenberger, G., Mamo, T., 2007. Soil aggregation, and total and
412 particulate organic matter following conversion of native forests to continuous cultivation in
413 Ethiopia. Soil Till. Res. 94 (1), 101–108.

414 Augé, R.M., 2004. Arbuscular mycorrhizae and soil/plant water relations. Can. J. Soil Sci. 84 (4),
415 373–381.

416 Bardgett, R.D., Streeter, T.C., Bol, R., 2003. Soil microbes compete effectively with plants for
417 organic-nitrogen inputs to temperate grasslands. Ecology 84 (5), 1277–1287.

418 Becagli, C., Puletti, N., Chiavetta, U., Cantiani, P., Salvati, L., Fabbio, G., 2013. Early impact of
419 alternative thinning approaches on structure diversity and complexity at stand level in two
420 beech forests in Italy. Ann. Silv. Res. 37, 55–63.
421 http://www.researchgate.net/publication/261027022.

422 Beck, T., 1971. Die messung der katalaseaktivitaet von Böden. Zeitschrift für Pflanzenernährung
423 und Bodenkunde, 130 (1), 68–81. (in German).

424 Billings, S.A., Lichter, J., Ziegler, S.E., Hungate, B.A., Richter, D.d.B., 2010. A call to investigate
425 drivers of soil organic matter retention vs. mineralization in a high CO_2 world. Soil Biol.
426 Biochem. 42 (4), 665–668.

427 Box, J.D., 1983. Investigation of the Folin–Ciocalteau reagent for the determination of
428 polyphenolic substances in natural waters. Water Res. 17, 511–525.
429 Brandstetter, A., Sletten, R.S., Mentler, A., Wenzel, W.W., 1996. Estimating dissolved organic
430 carbon in natural waters by UV absorbance (254 nm). J. Plant Nutr. Soil Sci. 159, 605–607.
431 https://doi.org/10.1002/jpln.1996.3581590612.
432 Chapin III, F.S., Matson, P.A., Mooney, H.A., 2002. Principles of Terrestrial Ecosystem Ecology.
433 Matson PA, Chapin III SF, Mooney HA (Eds) Springer-Verlag New York, Inc. pp. 398.
434 Ciavatta, C., Govi, M., Antisari, L.V., Sequi, P., 1990. Characterization of humified compounds
435 by extraction and fractionation on solid polyvinylpyrrolidone. J. Chromatogr. A 509 (1), 141–
436 146.
437 Conforti, M., Froio, R., Matteucci, G., Buttafuoco, G., 2015. Visible and near infrared
438 spectroscopy for predicting texture in forest soil: an application in Southern Italy. iForest 8,
439 339–347. doi: 10.3832/ifor1221-007.
440 Culliney, T., 2013. Role of Arthropods in Maintaining Soil Fertility. Agriculture 3, 629–659.
441 De Deyn, G.B., Quirk, H., Yi, Z., Oakley, S., Ostle, N.J., Bardgett, R.D., 2009. Vegetation
442 composition promotes carbon and nitrogen storage in model grassland communities of
443 contrasting soil fertility. J. Ecol. 97 (5), 864–875.
444 De Graaff, M.A., Adkins, J., Kardol, P., Throop, H., 2015. A meta-analysis of soil biodiversity
445 impacts on the carbon cycle. Soil 1 (1), 257–271.
446 Doran, J.W., Parkin, T.B., 1994. Defining and assessing soil quality. Defining soil quality for a
447 sustainable environment 1–21.
448 Elliot, W.J., Page-Dumroese, D., Robichaud, P.R., 1998. The Effects of Forest Management on
449 Erosion and Soil Productivity. Soil quality and soil erosion 12, 195.
450 Gigliotti, G., Businelli, D., Giusquiani, P., 1999. Composition changes of soil humus after massive
451 application of urban waste compost: a comparison between FT-IR spectroscopy and
452 humification parameters. Nutr. Cycl. Agroecosys. 55 (1), 23–28.

453 Gong, P., Guan, X., Witter, E., 2001. A rapid method to extract ergosterol from soil by physical
454 disruption. Appl. Soil Ecol. 17, 285–289.
455 Hammer, Ø., Harper, D.A.T., Ryan, P.D., 2001. PAST: paleontological statistics software package
456 for education and data analysis. Palaeontol. Electronica 4, 9. http://palaeo-
457 electronica.org/2001_1/past/issue1_01.htm.
458 Hodge, A., Campbell, C.D., Fitter, A.H., 2001. An arbuscular mycorrhizal fungus accelerates
459 decomposition and acquires nitrogen directly from organic material. Nature 413 (6853), 297.
460 Hooper, D.U., Chapin, F.S., Ewell, J.J., Hector, A., Inchausti, P., Lavorel, S. et al., 2005. Effects
461 of biodiversity on ecosystem functioning: a consensus of current knowledge. Ecol. Monogr. 75,
462 3–35.
463 IBM Corp. Released, 2012. IBM SPSS Statistics for Windows, Version 21.0. Armonk, NY: IBM
464 Corp.
465 Kandeler, E., Gerber, H., 1988. Short-term assay of soil urease activity using colorimetric
466 determination of ammonium. Biol. Fert. Soils 6 (1), 68–72.
467 Kim, S., Li, G., Han, S.H., Kim, C., Lee, S.T., Son, Y., 2019. Microbial biomass and enzymatic
468 responses to temperate oak and larch forest thinning: Influential factors for the site-specific
469 changes. Sci. Total Environ. 651, 2068–2079.
470 Kjeldalh, J., 1883. Neue methode zurestimmung des stickstoffs in organischen körpen. Zh. Anal.
471 Chem. 22, 366–382. (in German).
472 Köppen, W., 1936. Das geographische System der Klimate [The geographic system of climates].
473 In: “Handbuch der Klimatologie” (Köppen W, Geiger R eds). Gebrüder Borntraeger, Berlin,
474 Germany, pp. 1–44. (in German).
475 Lal, R., 2006. Enhancing crop yields in the developing countries through restoration of the soil
476 organic carbon pool in agricultural lands. Land Degrad. Dev. 17 (2), 197–209.

Lee, K.H., Jose, S., 2003. Soil respiration, fine root production, and microbial biomass in cottonwood and loblolly pine plantations along a nitrogen fertilization gradient. For. Ecol. Manag. 185 (3), 263–273.

Lombardi, F., Lella, S.D., Altieri, V., Benedetto, S.D., Giancola, C., Lasserre, B., Kutnar, L., Tognetti, R., Marchetti, M., 2018. Early responses of biodiversity indicators to various thinning treatments in mountain beech forests. iForest 11 (5), 609.

Lukac, M., 2017. Soil biodiversity and environmental change in European forests. Central European Forestry Journal 63, 59–65.

Ma, S., Concilio, A., Oakley, B., North, M., Chen, J., 2010. Spatial variability in microclimate in a mixed-conifer forest before and after thinning and burning treatments. Forest Ecol. Manag. 259 (5), 904–915.

Marchi, E., Picchio, R., Spinelli, R., Verani, S., Venanzi, R., Certini, G., 2014. Environmental impact assessment of different logging methods in pine forests thinning. Ecol. Eng. 70, 429–436.

Masyagina, O.V., Hirano, T., Ji, D.H., Choi, D.S., Qu, L., Fujinuma, Y., Sasa, K., Matsuura, Y., Prokushkin, S.G., Koike, T., 2006. Effect of spatial variation of soil respiration rates following disturbance by timber harvesting in a larch plantation in northern Japan. Forest Sci. Technol. 2 (2), 80–91.

Ministero delle Risorse Agricole, Alimentari e Forestali, 1994. Metodi Ufficiali di Analisi Chimica del Suolo. ISMEA, Roma. (in Italian).

Montgomery, H., Monreal, C., Young, J., Seifert, K., 2000. Determination of soil fungal biomass from soil ergosterol analyses. Soil Biol. Biochem. 32 (8-9), 1207–1217.

Muscolo, A., Panuccio, M.R., Mallamaci, C., Sidari, M., 2014. Biological indicators to assess short-term soil quality changes in forest ecosystems. Ecol. Indic. 45, 416–423.

Muscolo, A., Settineri, G., Attinà, E., 2015. Early warning indicators of changes in soil ecosystem functioning. Ecol. Indic. 48, 542-549.

503 Parisi, V., Menta, C., Gardi, C., Jacomini, C., Mozzanica, E., 2005. Microarthropod communities
504 as a tool to assess soil quality and biodiversity: a new approach in Italy. Agr. Ecosyst. Environ.
505 105 (1-2), 323–333.
506 Picchio, R., Neri, F., Petrini, E., Verani, S., Marchi, E., Certini, G., 2012. Machinery-induced soil
507 compaction in thinning two pine stands in central Italy. Forest Ecol. Manag. 285, 38–43.
508 Picchio, R., Spina, R., Calienno, L., Venanzi, R., Lo Monaco, A., 2016. Forest operations for
509 implementing silvicultural treatments for multiple purposes. It. J. Agron. 11, 156–161.
510 Resck, D.V.S., Ferreira, E.A.B., Figueiredo, C.C., Zinn, Y.L., 2008. Organic matter dynamics in
511 the Cerrado = Dinâmica da matéria orgânica no Cerrado. p. 359-417. In: Santos, G.A.; Silva,
512 L.S.; Canellas, L.P.; Camargo, F.A.O., eds. Fundamentals of soil organic matter: tropical and
513 subtropical ecosystems = Fundamentos da matéria orgânica do solo: ecossistemas tropicais e
514 subtropicais. Metrópole, Porto Alegre, RS, Brazil (in Portuguese).
515 Roscoe, R., Buurman, P., 2003. Tillage effects on soil organic matter in density fractions of a
516 Cerrado Oxisol. Soil Till. Res. 70 (2), 107–119.
517 Schmidt, M.W., Torn, M.S., Abiven, S., Dittmar, T., Guggenberger, G., Janssens, I.A., Kleber,
518 M., Kögel-Knabner, I., Lehemann, J., Manning, D.A.C., Nannipieri, P., Rasse, D.P., Weiner,
519 S., Trumbore, S.E., 2011. Persistence of soil organic matter as an ecosystem property. Nature
520 478 (7367), 49.
521 Settineri, G., Mallamaci, C., Mitrović, M., Sidari, M., Muscolo, A., 2018. Effects of different
522 thinning intensities on soil carbon storage in Pinus laricio forest of Apennine South Italy. Eur.
523 J. For. Res. 137, 131–141.
524 Sidari, M., Muscolo, A., Cianci, V., Attinà, E., Vecchio, G., Zaffina, F., 2005. Evoluzione della
525 sostanza organica in suoli rappresentativi dell'Altopiano della Sila. Forest@ 2 (3), 296–305.
526 http://www.sisef.it/.
527 Six, J., Frey, S., Thiet, R., Batten, K., 2006. Bacterial and fungal contributions to carbon
528 sequestration in agroecosystems. Soil Sci. Soc. Am. J. 70 (2), 555-569.

529 Smith, S.E., Smith F.A., 2011. Roles of arbuscular mycorrhizas in plant nutrition and growth: new
530 paradigms from cellular to ecosystem scales. Annu. Rev. Plant Biol. 62, 227–250.
531 Smolander, A., Kitunen, V., Kukkola, M., Tamminen, P., 2013. Response of soil organic layer
532 characteristics to logging residues in three Scots pine thinning stands. Soil Biol. Biochem. 66,
533 51–59.
534 Soil Survey Staff, 2010. Keys to soil taxonomy (11th edn). Natural Resources Conservation
535 Service, USDA, Washington, DC, USA, p. 338.
536 http://www.nrcs.usda.gov/Internet/FSE_DOCUMENTS/nrcs142p2_050915.pdf.
537 Springer, U., Klee, J., 1954. Prüfung der Leistungsfähigkeit von einigen wichtigeren Verfahren
538 zur Bestimmung des Kohlenstoffs mittels Chromschwefelsäure sowie Vorschlag einer neuen
539 Schnellmethode. Zeitschrift für Pflanzenernährung, Düngung, Bodenkunde 64(1), 1-26. (in
540 German).
541 Strickland, M.S., Rousk, J., 2010. Considering fungal: bacterial dominance in soils–methods,
542 controls, and ecosystem implications. Soil Biol. Biochem. 42 (9), 1385-1395.
543 Trentini, C.P., Campanello, P.I., Villagra, M., Ritter, L., Ares, A., Goldstein, G., 2017. Thinning
544 of loblolly pine plantations in subtropical Argentina: Impact on microclimate and understory
545 vegetation. Forest Ecol. Manag. 384, 236–247.
546 Vance, E.D., Brookes, P.C., Jenkinson, D.S., 1987. An extraction method for measuring soil
547 microbial biomass C. Soil Biol. Biochem. 19, 703–707.
548 von Mersi, W., Schinner, F., 1991. An improved and accurate method for determining the
549 dehydrogenase activity of soils with iodonitrotetrazolium chloride. Biol. Fertil. Soils 11, 216–
550 220.
551 Wagg, C., Bender, S.F., Widmer, F., van der Heijden, M.G., 2014. Soil biodiversity and soil
552 community composition determine ecosystem multifunctionality. Proc. Natl. Acad. Sci. 111
553 (14), 5266–5270.

554 Walkley, A., Black, I.A., 1934. An examination of the Degtjareff method for determining soil
555 organic matter and a proposed modification of the chromic acid titration method. Soil Sci. 37,
556 29–38.
557 Wang, Q., Wang, S., 2011. Response of labile soil organic matter to changes in forest vegetation
558 in subtropical regions. Appl. Soil Ecol. 47 (3), 210–216.
559 Wang, Z., Gao, M., Wang, Z., She, Z., Hu, B., Wang, Y., Zhao, C., 2013. Comparison of
560 physicochemical parameters during the forced-aeration composting of sewage sludge and
561 maize straw at different initial C/N ratios. J. Air Waste Manage. 63 (10), 1130–1136.
562 Waring, B.G., Averill, C., Hawkes, C.V., 2013. Differences in fungal and bacterial physiology
563 alter soil carbon and nitrogen cycling: insights from meta-analysis and theoretical models. Ecol.
564 16 (7), 887–894.
565 Zagal, E., Muñoz, C., Quiroz, M., Córdova, C., 2009. Sensitivity of early indicators for evaluating
566 quality changes in soil organic matter. Geoderma 151 (3/4), 191–198.
567 Zhang, X., Guan, D., Li, W., Sun, D., Jin, C., Yuan, F., Wang, A., Wu, J., 2018. The effects of
568 forest thinning on soil carbon stocks and dynamics: A meta-analysis. Forest Ecol. Manag. 429,
569 36–43.
570 Xun, W., Huang, T., Zhao, J., Ran, W., Wang, B., Shen, Q., Zhang, R., 2015. Environmental
571 conditions rather than microbial inoculum composition determine the bacterial composition,
572 microbial biomass, and enzymatic activity of reconstructed soil microbical communities. Soil
573 Biol. Biochem. 90, 10–18.
574

575 **Figure Captions**

576 **Fig. 1** Principal component analysis diagram (PCA) in area with different forest management:
577 reference areas (T1), traditional thinning (T2), and innovative thinning (T3), at 0-15 cm (a) and 15-
578 30 cm (b) soil depths.

579 **Fig. 2** Distribution of mean values and standard deviations (bars) of soil ions (mg/Kg-1 dry soil),
580 collected at 0–15 cm (a) and 15–30 cm (b) depths; referred to different management: control (T1),
581 traditional thinning (T2), innovative thinning (T3). For each parameter the sample size was n = 3
582 for treatment. Bars followed by the same letter are not statistically different at $p \leq 0.05$ (Tukey test)

583 **Fig. 3** Distribution of mean values and standard deviations (bars) of QBS-ar index for micro-
584 arthropods, collected at 0–10 cm depths; referred to different management: control (T1), traditional
585 thinning (T2), innovative thinning (T3). For each parameter the sample size was n = 3 for treatment.
586 Bars and columns followed by the same letter are not statistically different at $p \leq 0.05$ (Tukey test)

587 **Fig. 4** Pearson's correlations (r) between the soil parameters at 0–15 cm (a) and 15–30 cm (b)
588 depths. The boxed dots show the significant correlations between values, the magnitude shows the
589 level (small boxed dots $p < 0.05$, large boxed dots $p < 0.01$). The red dots the negative ones, the blue
590 ones the positive ones (see the bars on the right of the figure)

591

592

Table 1 Mean values and standard deviations of soil parameters referred to two layers of soil (0-15; 15-30 cm): water content (WC, %); water soluble phenols (WSP, µg TAE g^{-1} dry soil); organic carbon (OC, %); dissolved organic carbon (ml DOC L^{-1}); total nitrogen (N, %); C/N ratio. For each parameter the sample size was n = 3 for treatment (T1- Control; T2- Traditional thinning; T3- Innovative thinning). Means in the same column followed by the same letter are not statistically different at $p \leq 0.05$ (Tukey test)

	0-15 cm			15 - 30 cm		
	T1	**T2**	**T3**	**T1**	**T2**	**T3**
WC	$41^{a} \pm 2$	$20.7^{b} \pm 3$	$18.5^{b} \pm 2$	$40.6^{a} \pm 4$	$16.8^{b} \pm 2$	$12^{b} \pm 3$
WSP	$259^{a} \pm 6$	$155^{b} \pm 7$	$67^{c} \pm 5$	$313^{a} \pm 6$	$163^{b} \pm 10$	$156^{b} \pm 16$
OC	$11.5^{a} \pm 0.4$	$6.5^{b} \pm 0.2$	$3.1^{c} \pm 0.1$	$5.4^{a} \pm 0.02$	$4.2^{c} \pm 0.02$	$4.9^{b} \pm 0.03$
DOC	$14.1^{a} \pm 0.1$	$14.5^{a} \pm 0.4$	$12.9^{b} \pm 0.2$	$11.1^{b} \pm 0.4$	$12.4^{a} \pm 0.6$	$10.3^{b} \pm 0.4$
N	$0.57^{a} \pm 0.01$	$0.43^{b} \pm 0.01$	$0.23^{c} \pm 0.01$	$0.34^{a} \pm 0.01$	$0.26^{c} \pm 0.01$	$0.31^{b} \pm 0.01$
C/N	$20.4^{a} \pm 0.6$	$15.1^{b} \pm 0.4$	$13.1^{c} \pm 0.3$	$15.7^{a} \pm 0.1$	$15.8^{a} \pm 0.1$	$15.9^{a} \pm 0.1$

Table 2 Mean values and standard deviations of soil parameters referred to two layers of soil (0-15; 15-30 cm): humification degree (DH, %); humification rates (HR, %); humification index (HI); microbial biomass (MBC, µg C g^{-1} f.s.); Fungal biomass-carbon (FBC, µg g^{-1}); ergosterol fungal biomarker (ERG, µg g $soil^{-1}$); fluorescein released (FDA, µg g^{-1} dry soil); dehydrogenase (DHA, µg INTF g^{-1} dry soil h^{-1}); catalase activity (CAT, O2%/3min/g dry soil $^{-1}$); urease activity (URE, mg NH_4^+–N g^{-1} dry soil $2h^{-1}$). For each parameter the sample size was n = 3 for treatment (T1- Control; T2- Traditional thinning; T3- Innovative thinning). Means in the same column followed by the same letter are not statistically different at $p \leq 0.05$ (Tukey test).

	0-15 cm			15 - 30 cm		
	T1	**T2**	**T3**	**T1**	**T2**	**T3**
DH	$88.7^a \pm 0.6$	$88.5^a \pm 0.5$	$89.7^a \pm 1.9$	$87.1^a \pm 0.7$	$83.4^b \pm 0.2$	$85.6^a \pm 0.9$
HR	$62.8^b \pm 0.4$	$61.1^c \pm 0.4$	$67.2^a \pm 0.7$	$61.7^b \pm 0.5$	$57.6^c \pm 0.1$	$64.1^a \pm 0.1$
HI	$0.33^c \pm 0.01$	$0.53^a \pm 0.01$	$0.42^b \pm 0.03$	$0.44^b \pm 0.02$	$0.58^a \pm 0.01$	$0.45^b \pm 0.01$
MBC	$1529^a \pm 7$	$910^b \pm 6$	$742^c \pm 3$	$612^a \pm 3$	$487^c \pm 6$	$593^b \pm 6$
FBC	$1051^b \pm 95$	$1778^a \pm 87$	$911^b \pm 94$	$167^b \pm 4$	$373^a \pm 9$	$140^b \pm 17$
Erg	$6.1^b \pm 0.5$	$10.3^a \pm 0.5$	$5.3^b \pm 0.5$	$0.96^b \pm 0.02$	$2.15^a \pm 0.05$	$0.08^c \pm 0.02$
FDA	$55.2^a \pm 5.1$	$56.9^a \pm 4.9$	$27.4^b \pm 0.3$	$21.9^b \pm 3.8$	$37.4^a \pm 2.9$	$25.6^b \pm 1.7$
DHA	$1.28^c \pm 0.02$	$1.93^b \pm 0.03$	$2.96^a \pm 0.04$	$1.62^a \pm 0.02$	$1.87^a \pm 0.28$	$1.51^b \pm 0.01$
CAT	$2.41^b \pm 0.05$	$3.08^a \pm 0.09$	$1.56^c \pm 0.08$	$1.01^b \pm 0.11$	$1.75^a \pm 0.06$	$1.16^b \pm 0.19$
URE	$87.5^a \pm 1.1$	$76.2^b \pm 2.2$	$68.1^c \pm 2.7$	$39.8^a \pm 1.7$	$36.5^a \pm 1.8$	$28.3^b \pm 1.1$

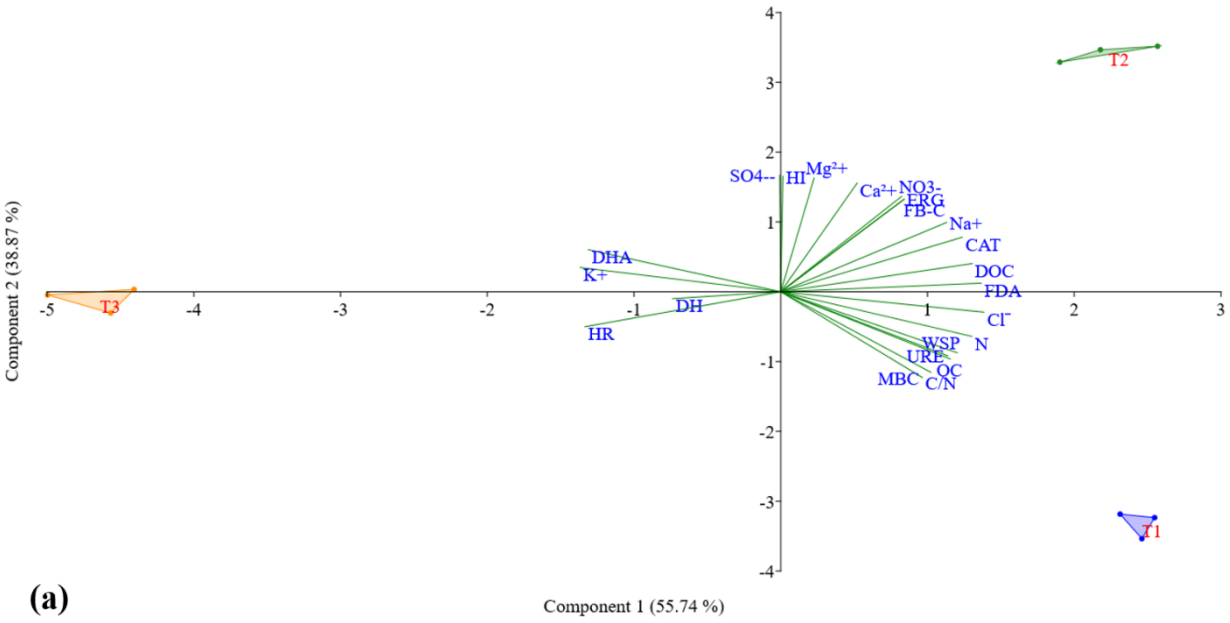
623 **Table 3** Micro-arthropod groups found in soils under different management: control (T1),
624 traditional thinning (T2), innovative thinning (T3). QBs index is obtained from the sum of the
625 highest values of EMI of all the collected groups.

Species	**T1**	**T2**	**T3**
Acari	*	*	*
Collembola	*	*	*
Orthoptera	*	*	*
Hemiptera	*	*	*
Diptera (larvae)	*	*	*
Coleoptera	*	*	*
Chilopoda		*	*
Blattaria	*		
Symphyla	*	*	*
Thysanoptera		*	*
Protura		*	
Diplura			*
Lepidoptera (larvae)		*	*
Araneae	*	*	*
Formicidae		*	
Isopoda		*	*

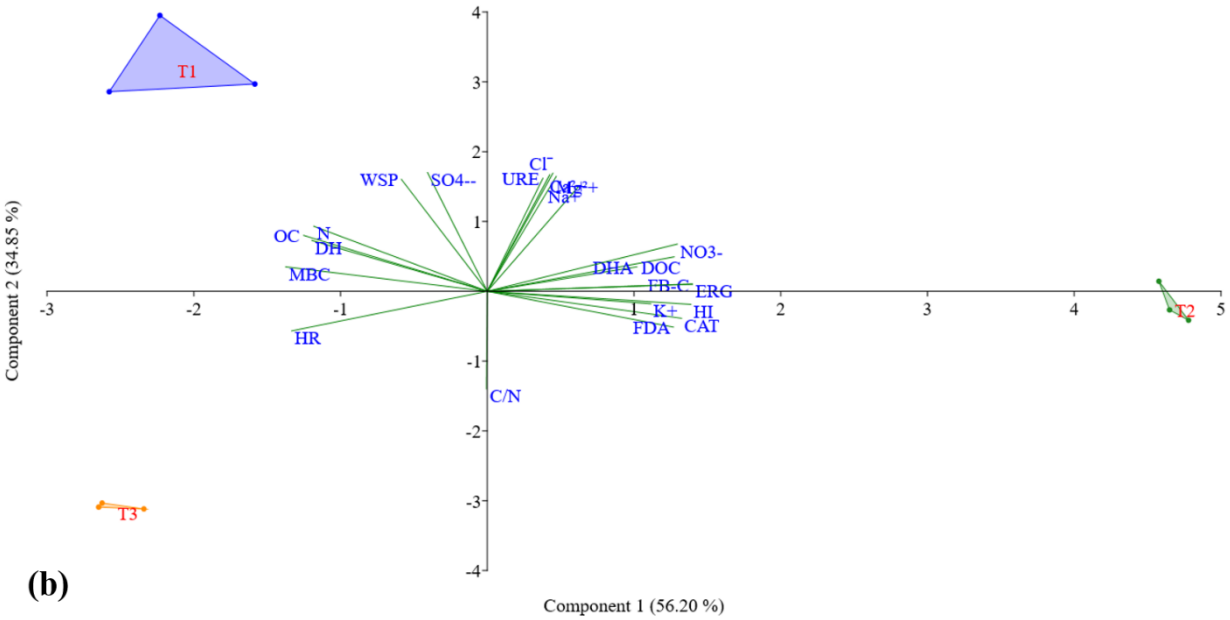
626

627

628



(a)



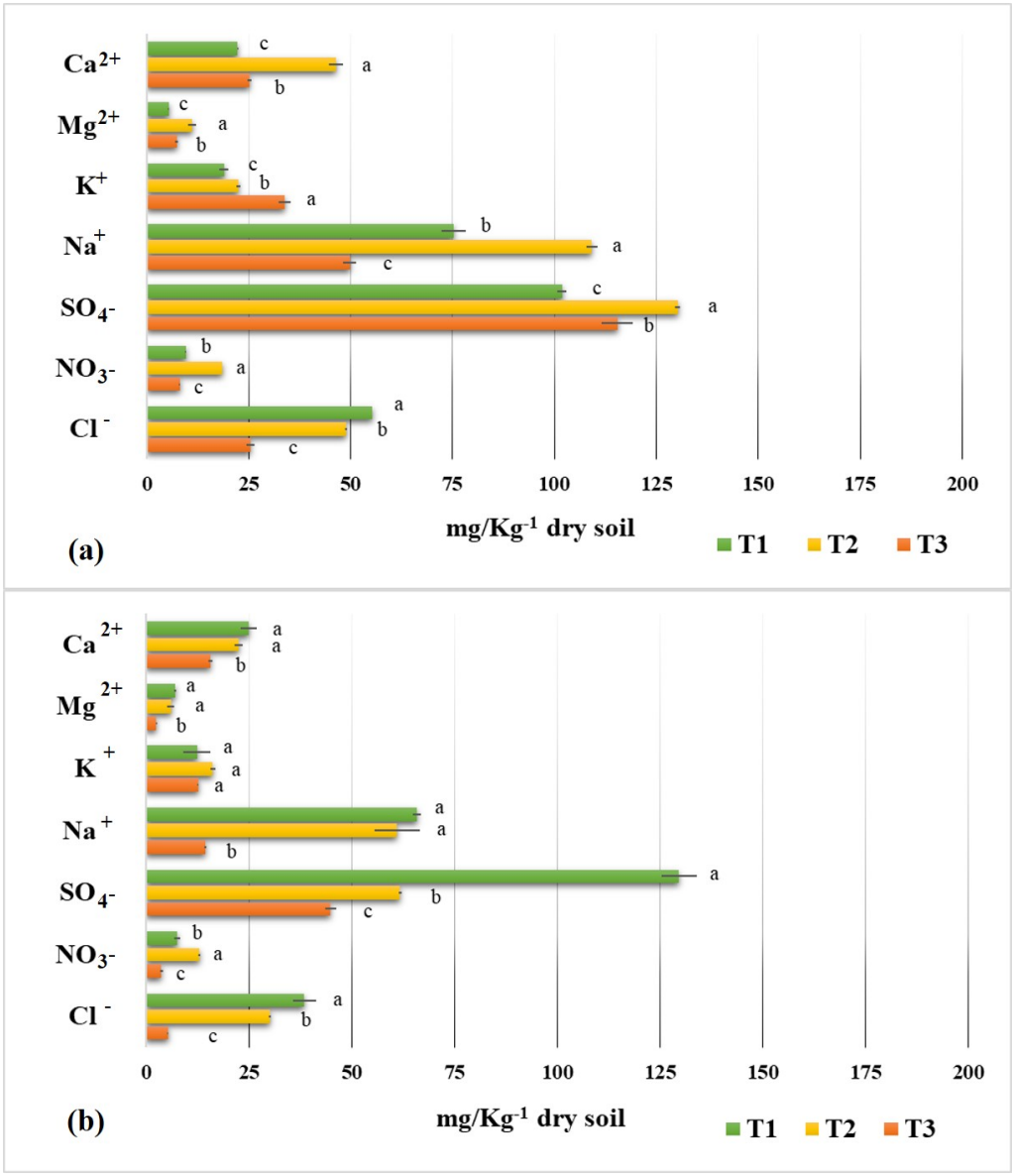
(b)

629

630

631 **Fig. 1**

632



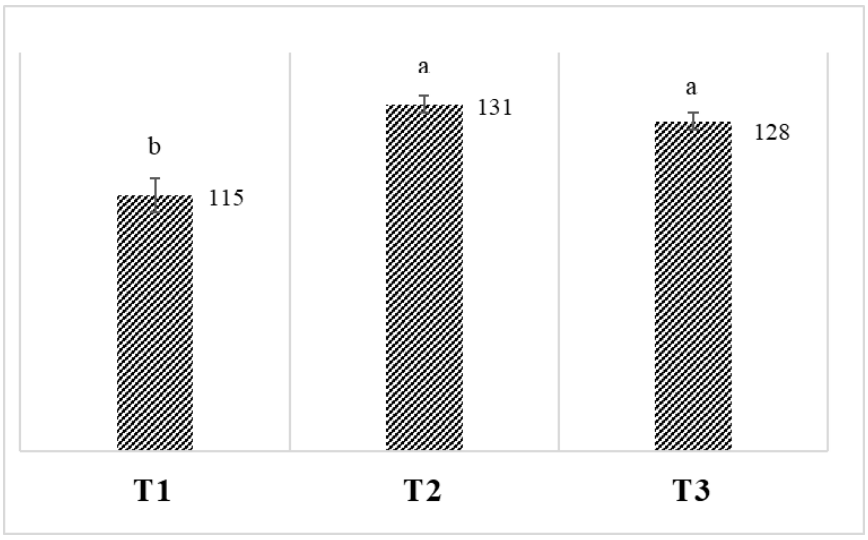
633

634

635 **Fig. 2**

636

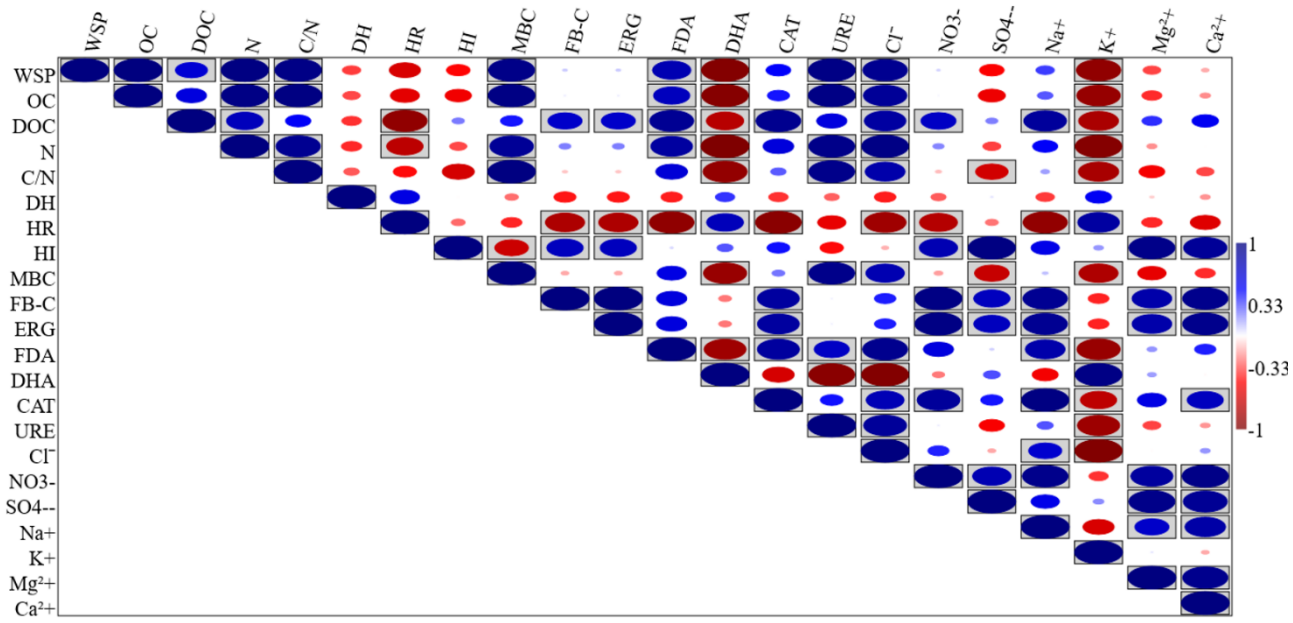
637



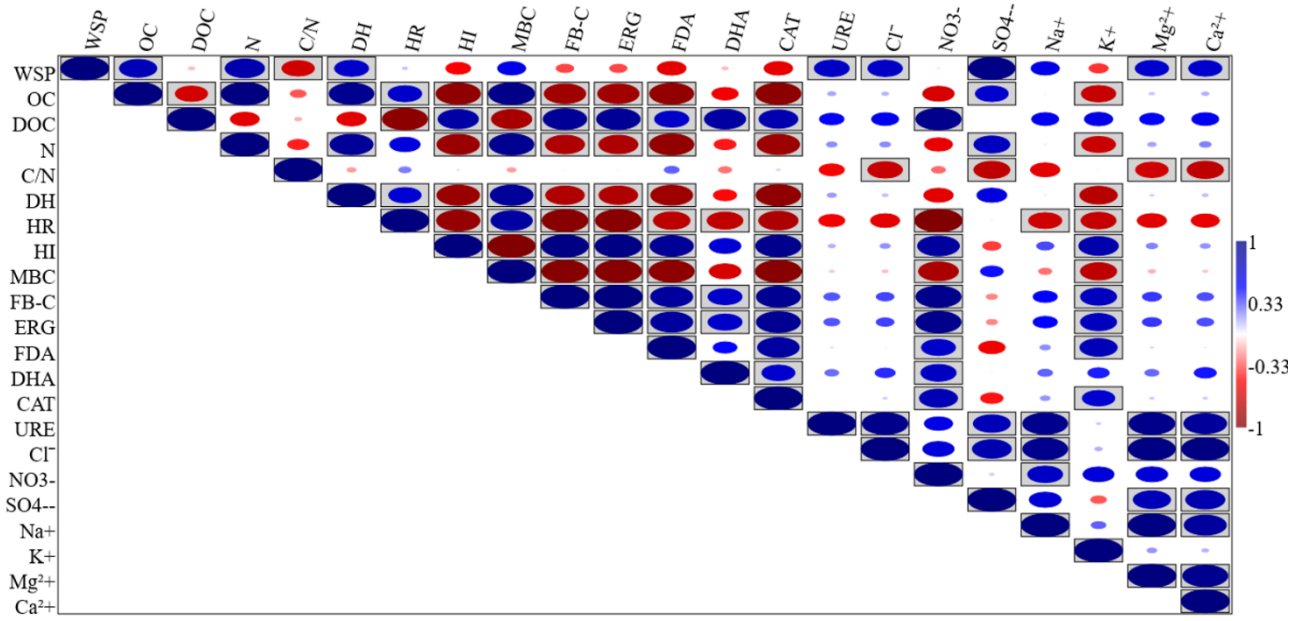
638

639

640 **Fig. 3**



(a)



(b)

641

642

643 **Fig. 4**