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Drought differently destabilizes soil structure in a chronosequence of abandoned agricultural lands

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ABSTRACT

Climate change is causing an increase in the frequency and severity of drought, and this has a significant influence on the global C cycle as soils are the main sink for C in terrestrial ecosystems. Drought can have serious consequences for the structural stability of the soil, which influences the protection of the organic matter stored in the soils and hence its mineralization. In this work we study the effects of an extreme drought event on the structural stability of the soil in Mediterranean annual grasslands developed after the abandonment of agricultural activity. This was done by building a chronosequence of abandoned vineyards and determining the changes in the stability of the soil aggregates and organic matter by monitoring these parameters for three consecutive years in permanent plots with different ages of abandonment. This period of time included an extreme drought event in the second year with a strong positive thermal anomaly, which caused a serious structural destabilization of the soil and had a major impact on the soil C and N cycles. The breakdown of the soil aggregates left unprotected a large amount of organic substrates that were rapidly mineralized when the environmental conditions were favourable for soil biological activity. This had significant repercussions on the functioning of these abandoned ecosystems, which became sources of atmospheric CO_2, when under normal conditions they act as C sinks. Furthermore, the effects of this extreme event were more severe in the plots with longer periods of abandonment, which have the highest amount of C accumulated in the soil.

1. Introduction

Droughts, heat waves, torrential rains and other climate extremes may impact the structure, composition and functioning of terrestrial ecosystems, and hence carbon cycling and its feedbacks to the climate system (Frank et al., 2015). Since climate change is leading to a sharp increase in the frequency and severity of these extreme events (IPCC, 2022), it is expected that this will also influence the soil carbon stock. A drought event is a temporary dry period characterized by below-normal precipitation which can have important consequences on ecosystem functions such as the ability to sequester carbon (Dai, 2011). Even a small change in the frequency or severity of climate extremes could substantially reduce carbon sinks and result in considerable positive feedbacks to climate warming (Reichstein et al., 2013). Specifically, extreme droughts may have long-lasting carbon feedbacks in grasslands (Hoover and Rogers, 2016). Although Mediterranean grasslands are adapted to undergo summer drought (Clary, 2008), the consequences of additional drought episodes on the soil during the vegetative growth of grasses are not fully known.

It is important to establish the effect of soil rewetting after a drought since it influences the soil C content, and its fate (Schimel, 2018) produces changes in the composition and activity of soil microorganisms (Acosta-Martínez et al., 2014; Barnard et al., 2013; Chodak et al., 2015; De Vries et al., 2018), thus having important consequences on the functioning of nutrient cycles (Horion et al., 2019). Specifically, droughts have been linked to the loss of soil structural stability due to the destruction of macroaggregates (Zhang et al., 2019), which in turn increases bulk density and decreases porosity. The breakdown of soil aggregates has a major influence on the C cycle since this enables microbial access to the soil carbon, which then decomposes and produces CO_2 (Schimel and Schaeffer, 2012). After the drought and the subsequent rewetting of the soil, a large amount of organic matter that was protected in the soil aggregates becomes available to organisms, causing a drastic mineralization and the consequent loss of organic matter

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thanks to exoenzymes (Navarro-García et al., 2012).

The formation and stabilization of soil aggregates is determined not only by abiotic factors such as clay content and its mineralogy and organic matter content (Denef and Six, 2005), but also by biotic factors such as roots and fungal hyphae, and products exuded by the roots and soil microbiota that facilitate the formation of aggregates (Mambelli et al., 2011; Tang et al., 2011; Cotrufo et al., 2013; Degens, 1997). Soil aggregation is one of the most important abiotic factors influencing C storage in the soil and the stability of the C pool due to its physical protection which hinders the degradation of organic matter (Six et al., 2000). Soil organic matter is distributed among different-sized aggregates, and organic matter linked to microaggregates is more protected against degradation (Six et al., 2004; Davinic et al., 2012; Bach et al., 2018). The contraction of soil particles during drought causes the aggregates to break down, which means that this previously protected organic matter becomes available (Borken and Matzner, 2009). Changes in the soil structure due to the breakdown of aggregates can therefore have important consequences on the kinetics of soil organic C and on the sequestration capacity of this element (Liu et al., 2019; Yilmaz et al., 2019). Most of the previous knowledge is based either on rainfall-reduction field experiments or on drought simulation laboratory experiments. Our work, based on field monitoring in permanent plots, focuses on delimiting these consequences on Mediterranean grasslands, a system that has evolved resilience to a seasonal dry period.

Apart from the increase in extreme weather events such as droughts, other man-made changes are occurring simultaneously (Doblas-Miranda et al., 2015). Notable among these is the abandonment of agricultural activity which affects large areas of the world (Bakker et al., 2005; Rey Benayas et al., 2007; Keenleyside and Tucker, 2010), and has important effects on the functioning of ecosystems (Lozano et al., 2014; Novara et al., 2017). Colonization by vegetation after abandonment produces an increase in soil C (Novara et al., 2017; Vaquero Perea et al., 2020), and together with the rise in microbial activity and biomass (Quintana et al., 2021; Zhou et al., 2017), improves the stability of the soil aggregates which, as mentioned before, is essential for the stabilization of C in the soil. However, there is still no knowledge of how far this stabilization of aggregates and C in secondary succession is counteracted by the destabilization caused by drought events.

Our working hypotheses are that in a secondary succession drought disrupts the soil aggregates and their relationships with the organic matter dynamics, and that this disturbance is differently influenced by the time of abandonment. We test this hypothesis in a chronosequence of vineyards abandoned over the last 60 years which were sampled for three consecutive years (2016, 2017 and 2018), including a drought event. The specific objectives of this work were: (a) to determine how drought influences the organic matter distribution across aggregate classes; (b) to study the relationships between enzyme activities related to the C and N cycles and the distribution of C in the aggregate classes; and (c) to analyse whether the effects of drought on soil organic carbon dynamics differ according to the time since crop abandonment. Although seasonal dry ecosystems such as the Mediterranean are adapted to drought events, the predicted increase in the frequency and severity of these events could have an important impact on their functioning and potentially compromise their resilience. Our results could provide an insight into the feedback between the gain in soil C due to land abandonment and carbon lost due to drought events.

2. Material and methods

2.1. Study area

The study zone is located in a rural area (Navas del Rey, 709 m a.s.l.) at the western corner of the Region of Madrid, in the centre of the Iberian Peninsula. It is a slightly sloping area on the left margin of the middle valley of the Alberche river. The predominant geological materials are igneous rocks, mainly granites, and the representative soil units are Leptosols and Regosols (WRBI, 2015), which are characterised by a coarse texture and good drainage (Consorcio Sierra Oeste, 2009). The average annual temperature is 14.4 °C. The average of the maximum temperatures of the warmest month (July) is 26.7 °C and the average of the minimum temperatures of the coldest month (January) is 4.7 °C. The average annual rainfall is about 477 mm, and the rainfall regime decreases as follows: autumn > spring > winter > summer. There are two months with the lowest rainfall (July 10 mm and August 13 mm). The bioclimate in the study area is defined as Mediterranean pluviseasonal-oceanic, and it has a subhumid climate (Rivas-Martínez, 2007). The plant landscape comprises evergreen oak woodlands of *Quercus rotundifolia* which are arranged in a mosaic with shrublands of *Retama sphaerocarpa*, scrublands of *Lavandula pedunculata* and *Thymus mastichina*, and annual Mediterranean grasslands (Rivas-Martínez, 1982).

The historic landscape management of the study area has shaped the evolution of the landscape for at least the last 70 years. The area has traditionally been used for vineyards and sheep grazing in the past (Vaquero Perea et al., 2020). In the early 1950 s, the area was mainly used as vineyards, which covered 80 % of the landscape. Since the 1960s the vineyards have been progressively abandoned due to rural depopulation and the change in EU agricultural policies. Vineyards currently cover <20 % of the cultivable area.

2.2. Sampling design

Our study was based on a chronosequence of six vineyards that had been abandoned for different lengths of time (1–40 years in 2016), one active vineyard and one plot that had not been used as a vineyard in at least the last 59 years. All these plots were in close proximity. The plot size was between 1.626 and 7.869 m^2, with an average size of about 4.366 m^2. The plot slope ranged from to 2 to 7 %, with an average of 5 %. The detailed environmental features of the study plots are described in Valverde-Asenjo et al. (2020). Three permanent quadrats (1 m^2) were randomly established in each plot within the area occupied by annual grasslands in 2016. The chronosequence was studied in three consecutive years (2016–2018). The timing and variability of the climate data in the study area for the period 2015–2018 is covered in Quintana et al. (2021). This 3-year period included a drought lasting from spring to winter 2017 in which the below-normal precipitation was accompanied by an above-normal temperature. The soil was sampled in 24 grassland sites (three samplings randomly chosen per plot) in mid-May each year. The spatial distribution of the plots and sites and the soil sampling procedure is described in detail in Quintana et al. (2021). The vegetation dynamics and biodiversity in the study sites is addressed by Molina et al. (2022).

2.3. Laboratory methods

Total organic carbon (TOC) in soil was determined by the wet oxidation method of Walkley and Black (1934) using an automatic titrator Metrohm 888 TITRANDO and Metrohm 665 DOSIMAT. Total nitrogen (TN) was determined by combustion with a LECO CHNS-932 analyzer. Water soluble organic carbon (WSOC) and water soluble nitrogen (WSN) were extracted according to the method of Ghani et al. (2003) and quantified using a TC/TN analyzer ANALYTIKJENA MICRO N/C. Aggregate size class distribution was determined following Guidi et al. (2014). Three aggregate size fractions were obtained: i) large aggregates (LA) (2–0.5 mm), ii) medium aggregates (MA) (0.5–0.05 mm), and iii) small aggregates (SA) (<0.05 mm). In each aggregate size class, TOC was determined by the method of Walkley and Black (1934). The content of TOC in each fraction was calculated by multiplying the weight of each size class and their TOC content. The relative TOC percentage bound to each fraction was calculated by dividing their TOC content by the sum of the TOC content of the three fractions.

For the determination of enzyme activity, soil samples were refrigerated in the field and wet sieved to <2 mm in the laboratory, then stored in the refrigerator until the analyses. Activities related to the C cycle (α-glucosidase, β-glucosidase, β-galactosidase, and phenoloxidase), and to the N cycle (arylamidase, *N*-acetyl-glucosaminidase – NAG – and urease) were determined by ISO 20130 (ISO, 2018). Measurements were made on a UV–vis spectrophotometer with a TECAN NANOQUANT INFINITE M200 PRO multi-well plate reader.

2.4. Statistical analysis

Prior to the statistical analysis, the soil variables were normalized by converting them to logarithms. The Kolmogorov-Smirnov test was also used to test the normality of the variables. A mixed model ANOVA for repeated measures was performed to determine the influence of the sampling year (within-subject factor) and the age of abandonment (between-subject factor) on the soluble organic matter in soil and organic matter in aggregate fractions. Prior to the ANOVA analysis, the sphericity of the variables was tested by means of Mauchly's test so as to reduce the Type I error rate. When the variables did not fulfil the null hypothesis of sphericity, the alternatives with the highest observed power calculated by the SSPSS program were chosen. At the same observed power, the Greenhouse-Geisser test was chosen as the most conservative. An *a posteriori* Bonferroni test was then performed to test the sampling year in each age of abandonment and the age of abandonment in each sampling year. We used a threshold of $p \leq 0.05$ to assess statistical significance. The Pearson correlation coefficient was used to obtain information about the intensity and direction of the relationships between the weight of the aggregate size class, content and percentages of TOC in each class with the contents of soluble and total TOC and TN in the soil. The Pearson correlation coefficient was also used to explore the relationships between biological activity and the contents of soluble and total TOC and TN in the soil, the weight of the aggregate size class, and the content and percentages of TOC in each class. Changes in correlations may indicate disruptions in the cycles. All statistical analyses were performed using SPSS software (Statistical Package for the Social Sciences, SPSS, Inc.).

Table 1
Results of the repeated measures ANOVA mixed model calculated for soluble organic matter and organic matter in aggregate fractions considering the sampling year (Year) as a within-subjects variable and the age of abandonment (Age) as a between-subjects variable. WSOC = water soluble carbon; WSN = water soluble nitrogen; TOC = Total Organic Carbon; LA = Large aggregates (2–0.5 mm); MA = Medium aggregates (0.5–0. 05 mm); SA = Small aggregates (<0.05 mm).

Variable	Factor	SS	Sphericity	df	F	p	η_p^2
Soluble organic matter							
WSOC (mg kg^{-1})	Year	0.150	Assumed	2	2.674	0.084	0.143
	Year × Age	0.392	Assumed	14	1.000	0.476	0.304
	Error	0.896	Assumed	32			
	Age	0.582		7	2.746	0.045	0.546
WSN (mg kg^{-1})	Year	0.838	Greenhouse	1.305	13.948	0.001	0.466
	Year × Age	1.978	Greenhouse	9.137	4.702	0.002	0.673
	Error	0.962	Greenhouse	20.885			
	Age	0.763		7	14.762	0.005	0.676
Organic matter in aggregate fractions							
Weight of aggregate fractions (%)							
LA	Year	0.111	Assumed	2	37.342	<0.001	0.700
	Year × Age	0.061	Assumed	14	2.901	0.006	0.559
	Error	0.048	Assumed	32			
	Age	0.065		7	4.221	0.008	0.649
MA	Year	0.278	Assumed	2	36.381	<0.001	0.695
	Year × Age	0.170	Assumed	14	3.170	0.003	0.581
	Error	0.122	Assumed	32			
	Age	0.102		7	2.527	0.059	0.525
SA	Year	0.635	Assumed	2	34.044	<0.001	0.680
	Year × Age	0.142	Assumed	14	1.089	0.403	0.323
	Error	0.299	Assumed	32			
	Age	0.521		7	7.962	<0.001	0.777
TOC content in aggregate fractions (g kg^{-1})							
LA	Year	0.262	Assumed	2	22.886	<0.001	0.589
	Year × Age	0.075	Assumed	14	0.941	0.529	0.292
	Error	0.183	Assumed	32			
	Age	0.383		7	9.777	<0.001	0.811
MA	Year	0.013	Assumed	2	1.154	0.328	0.067
	Year × Age	0.126	Assumed	14	1.633	0.123	0.417
	Error	0.176	Assumed	32			
	Age	0.564		7	13.820	<0.001	0.858
SA	Year	0.717	Greenhouse	1.395	29.190	<0.001	0.646
	Year × Age	0.267	Greenhouse	9.765	1.550	0.188	0.404
	Error	0.393	Greenhouse	22.320			
	Age	0.468		7	5.426	0.002	0.704
TOC percentage in aggregate fractions							
LA	Year	0.134	Greenhouse	1.319	71.305	<0.001	0.817
	Year × Age	0.018	Greenhouse	9.235	1.346	0.272	0.371
	Error	0.030	Greenhouse	21.109			
	Age	0.014		7	3.266	0.024	0.588
MA	Year	0.072	Assumed	2	38.693	<0.001	0.707
	Year × Age	0.014	Assumed	14	1.119	0.380	0.329
	Error	0.030	Assumed	32			
	Age	0.006		7	2.051	0.111	0.473
SA	Year	0.035	Greenhouse	1.350	51.551	<0.001	0.763
	Year × Age	0.007	Greenhouse	9.447	1.578	0.182	0.408
	Error	0.011	Greenhouse	21.594			
	Age	0.010		7.273	7.273	0.001	0.761

3. Results

3.1. Soluble organic matter

The mixed model of repeated measures ANOVA showed that the age of abandonment had a significant influence on WSOC and WSN, which increased throughout the chronosequence (Table 1, Fig. 1a,b). The sampling year was significant for WSN (Table 1), which showed higher WSN values in the post-drought year (2018), especially in the plots that had been abandoned for longer and the control (Fig. 1b), which would explain the significant interaction between the age of abandonment and the sampling year. The significant relationship between the WSN and the interannual rate points to a change in quality in this soluble organic matter after the drought event.

3.2. Organic matter in aggregate fractions

Aggregate classes were all significantly influenced by the sampling year, and explained a similar percentage (approx. 70 %) of the total variance (Table 1, Fig. 2). LA was the predominant aggregate fraction (>50 %) and SA was the minor fraction in all the study plots (<10 %). The drought year (2017) reduced the weight of the LA fraction – from

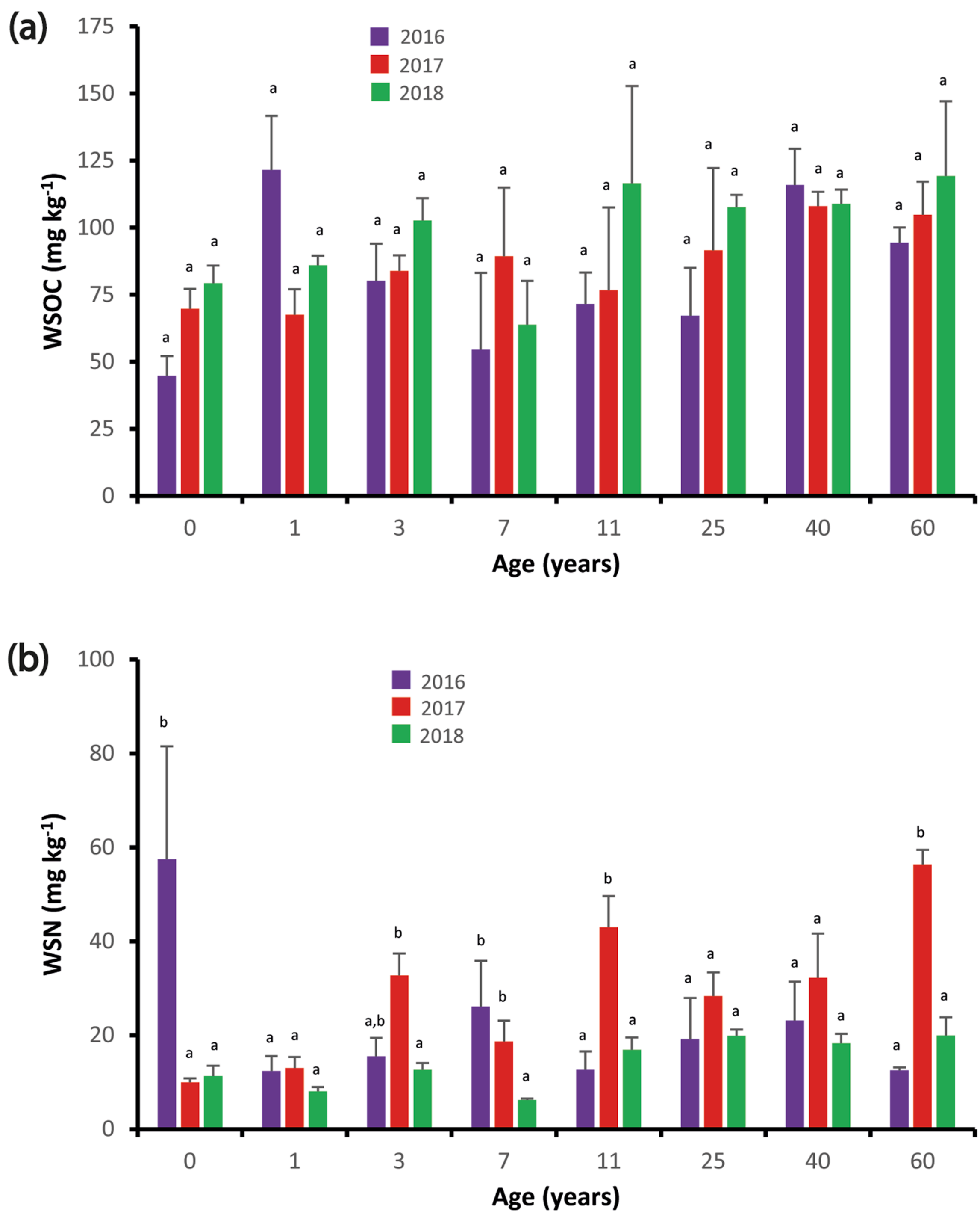

Fig. 1. Variations in the WSOC and WSN throughout the chronosequence in 2016, 2017, and 2018. (a) Water Soluble Organic Carbon, (b) Water Soluble Nitrogen. Error bars represent standard error (n = 3).

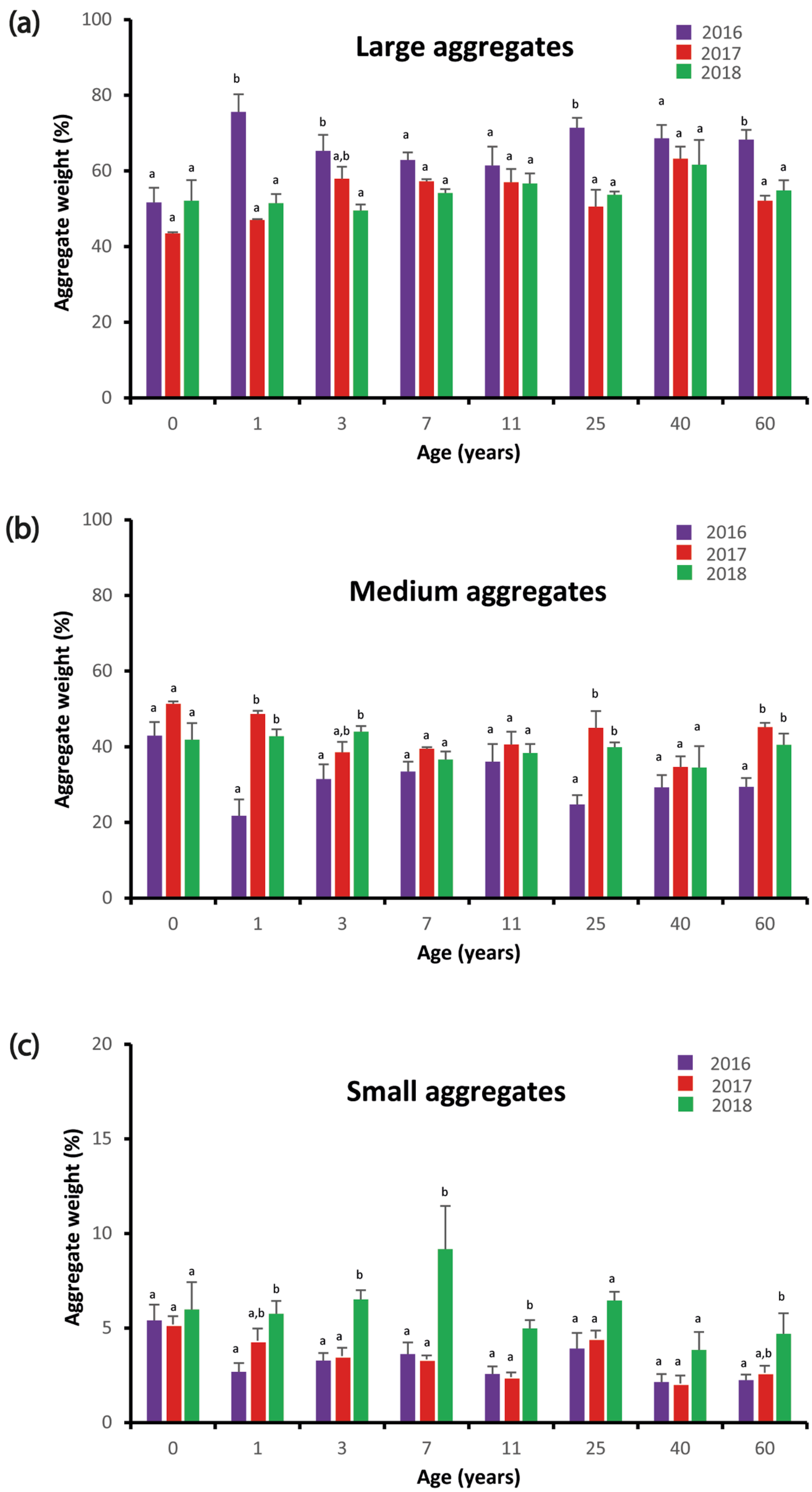

Fig. 2. Variations in aggregate weight throughout the chronosequence in 2016, 2017 and 2018. (a) Large aggregates (2–0.5 mm), (b) Medium aggregates (0.5–0.05 mm), (c) Small aggregates (<0.05 mm). Error bars represent standard error (n = 3).

$65.64 \pm 2.56_{2016}$ to $53.59 \pm 2.29_{2017}$ – and increased the MA fraction – from $31.13 \pm 2.33_{2016}$ to $42.92 \pm 1.97_{2017}$ (Fig. 2b). The SA fraction showed a sharp increase in the year after the drought – from $3.49 \pm 0.39_{2017}$ to $5.92 \pm 0.56_{2017}$ (Fig. 2c). The age of abandonment significantly influenced the LA and SA fractions (Table 1) with a slight tendency to increase the LA fraction and reduce the SA (LA decreased from $51.66 \pm 3.89_{\text{current vineyard}}$ to $68.60 \pm 3.57_{\text{40 years after abandonment in 2016}}$ and SA decreased from $5.40 \pm 0.83_{\text{current vineyard}}$ to $2.15 \pm 0.42_{\text{40 years after abandonment in 2016}}$).

The mixed model of repeated measures ANOVA showed that TOC content was significantly influenced by the sampling year in LA and SA fractions (Table 1, Fig. 3). A comparison of the pre-drought (2016) and post-drought year (2018) showed that TOC content decreased in the LA fraction from 12.36 ± 0.51 to 6.04 ± 0.46 g kg^{-1}, and increased in the SA fraction from 8.78 ± 0.58 to 22.01 ± 1.21 g kg^{-1}. The age of abandonment had a significant and positive influence on TOC content in the three aggregate fractions (Table 1). For example, in 2016 the TOC content in the LA fraction increased from 11.07 ± 0.39 g kg^{-1} $_{\text{current vineyard}}$ to 14.46 ± 1.65 g kg$^{-1}_{\text{40 years after abandonment in 2016}}$.

Likewise, the mixed model of repeated measures ANOVA showed that the TOC percentage in each aggregate fraction was significantly influenced by the sampling year (Table 1, Fig. 4). In the pre-drought year, most of the TOC percentage was linked to the LA fraction ($70.71 \pm 1.72_{2016}$); whereas after the drought event, most of the TOC percentage became related to the MA fraction ($50.04 \pm 2.42_{2018}$). Also noteworthy was the increase in the TOC percentage in the SA fraction in the post-drought year (from $2.21 \pm 0.21_{2016}$ to $15.87 \pm 2.16_{2018}$) The age of abandonment had a significant influence on the accumulated TOC percentage in the LA fraction increase and SA fraction decrease, explaining lower percentages of variance than in the sampling year (Table 1).

In short, the drought had both an immediate and a delayed effect on aggregate class size and on TOC distribution. An immediate effect was observed in the drought year on the increase in the MA fraction and relative TOC percentage at the expense of a decrease in the LA fraction and relative TOC percentage. A delayed effect was observed in the post-drought year when there was an increase in the SA fraction and its relative percentage and content in TOC, and a decrease in the TOC content in the LA fraction.

3.3. Relationships between aggregate fractions and total and water soluble C and N

It is worth noting the significant and inverse correlation found between TOC and TN with the weight of the SA fraction over the years (Table 2). According to our results, the higher the weight of the SA fraction, the lower the content in total C and N. A negative correlation was also observed in this fraction for WSOC in the post-drought year and for WSN in both the year and the post-drought year. Also noticeable was the loss of the positive correlation between the weight of LA with WSOC in the drought year and with TOC in the post-drought year (Table 2). The correlation between LA and WSN was reversed – from a negative to positive significant correlation – in the drought year, and lost in the post-drought year. Negative correlations between MA weight and TOC and WSOC were also lost from the drought year on. As expected there was a predominant and significant positive correlation between C content in all aggregate size classes and TOC and TN, except for SA in the drought year. In the post-drought year, a positive correlation was observed between WSOC and WSN in C content in all the fractions that had not hitherto been seen. The most noteworthy finding in regard to the TOC percentage in aggregates was the negative relationship between this factor and TOC, TN and WSN from the drought year on.

3.4. Relationship between soil organic fraction and enzyme activities related to the C and N cycles

In regard to the C cycle labile enzymes, the drought year did not change the positive correlations between alpha and beta glucosidases with TOC and TN content, the percentages in LA and MA fractions, or the negative correlations with SA weight (Table 3). The drought year concentrated most of the positive correlations between beta galactosidase and soil organic fractions (TOC, TN, WSOC, WSN), as well as the C percentage in the LA and MA fraction in the years under study. The recalcitrant phenoloxidase enzyme showed negative correlations with TOC and TN in the pre-drought year – which were lost from the year of the drought on – and positive correlations with each size class weight – which were lost in the post-drought year. With reference to the N cycle enzymes, positive correlations were observed in the drought year between *N*-arylamidase and the soil organic fractions (TOC, TN, WSOC, WSN) and the C percentage in the LA fraction, and a negative correlation with the MA and SA weights (Table 4). The post-drought year had positive correlations between both *N*-acetyl-glucosaminidase and urease with soil organic fractions, and percentage of accumulated C in each aggregate size class. Immediate indicators of the impact of drought on the soil structural stability can therefore be observed in the gaining or losing relationships between soil biological activities and aggregate size and the distribution of organic fractions.

4. Discussion

Drought substantially reduces soil macroaggregates and increases microaggregates (Su et al., 2020). According to the results of our field study, drought initially provoked a decrease in the LA fraction in favour of the MA fraction, and subsequently – in the post-drought year – an increase in the SA fraction. Although Mediterranean grassland ecosystems are adapted to warm dry summers (Rundel et al., 2016), our results revealed that drought in the period of greatest activity in the community (spring) had both an immediate and prolonged negative effect on soil structural stability. It would be interesting to continue monitoring this process to determine when structural stability is restored after a drought episode, which would give us an idea of the system's resilience in the face of a probable increase in such episodes in the future. Our results also showed that the beneficial effect of increased structural stability throughout the chronosequence is counteracted after a drought event.

Changes in the precipitation regime (intensity, quantity and frequency) are known to cause important disruptions in the C cycle (Cleveland et al., 2010; Ciais et al., 2005). The combination of drought and heatwaves has a greater impact on grassland carbon cycling, namely carbon uptake and plant growth (De Boeck et al., 2011; Li et al., 2020). In terms of soil structural stability, the disintegration of macroaggregates could contribute to the change in soil organic carbon storage under a drought scenario (Zhang et al., 2019). Our results, obtained from field research, revealed that most of the carbon is normally attached to the LA fraction; however, the drought and the post-drought year led to a significant reduction in the C attached to macroaggregates. This was not only due to the decrease in the weight of the LA fraction, but also to the decline in the content of C linked to macroaggregates. The decrease in the total amount of C observed during the study period due to the drought can be explained by the macroaggregate disruption that exposes the previous physically and chemically protected organic matter, meaning that these aggregates are now available to microbiota. Laboratory studies reported that the respiration pulse after soil rewetting is essentially determined by the physical disruption of soil aggregates which leaves previously protected substrates available, when they are rapidly used by microbiota (Navarro-García et al., 2012).

Bach and Hofmockel (2014) found that most enzyme activity in soils is linked to the macroaggregate fraction, suggesting that the majority of total soil organic matter processes occur within the macroaggregates. Our results support this finding, since positive and significant

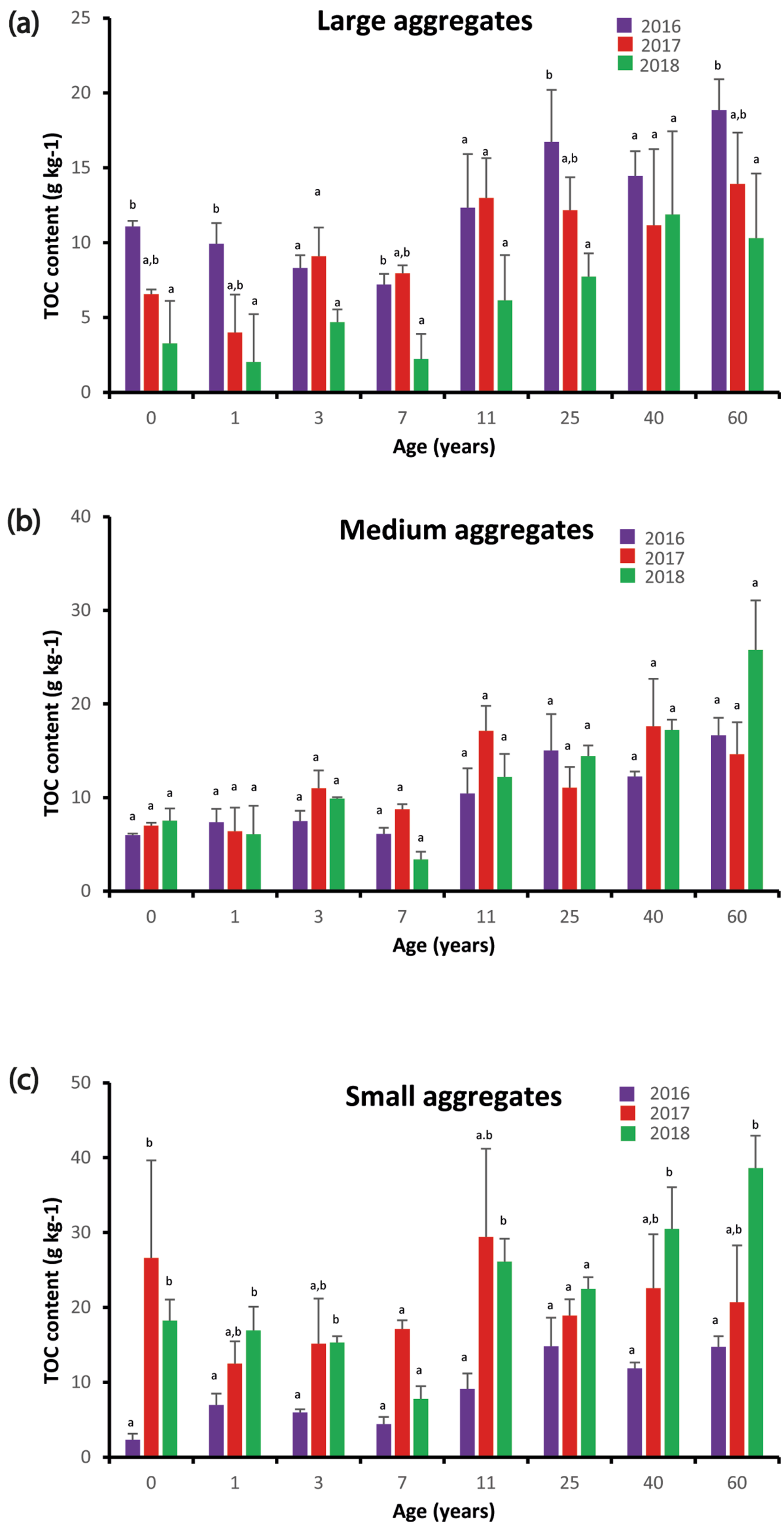

Fig. 3. Variations in TOC content (g kg^{-1}) throughout the chronosequence in 2016, 2017 and 2018. (a) Large aggregates (2–0.5 mm), (b) Medium aggregates (0.5–0.05 mm), (c) Small aggregates (<0.05 mm). Error bars represent standard error (n = 3).

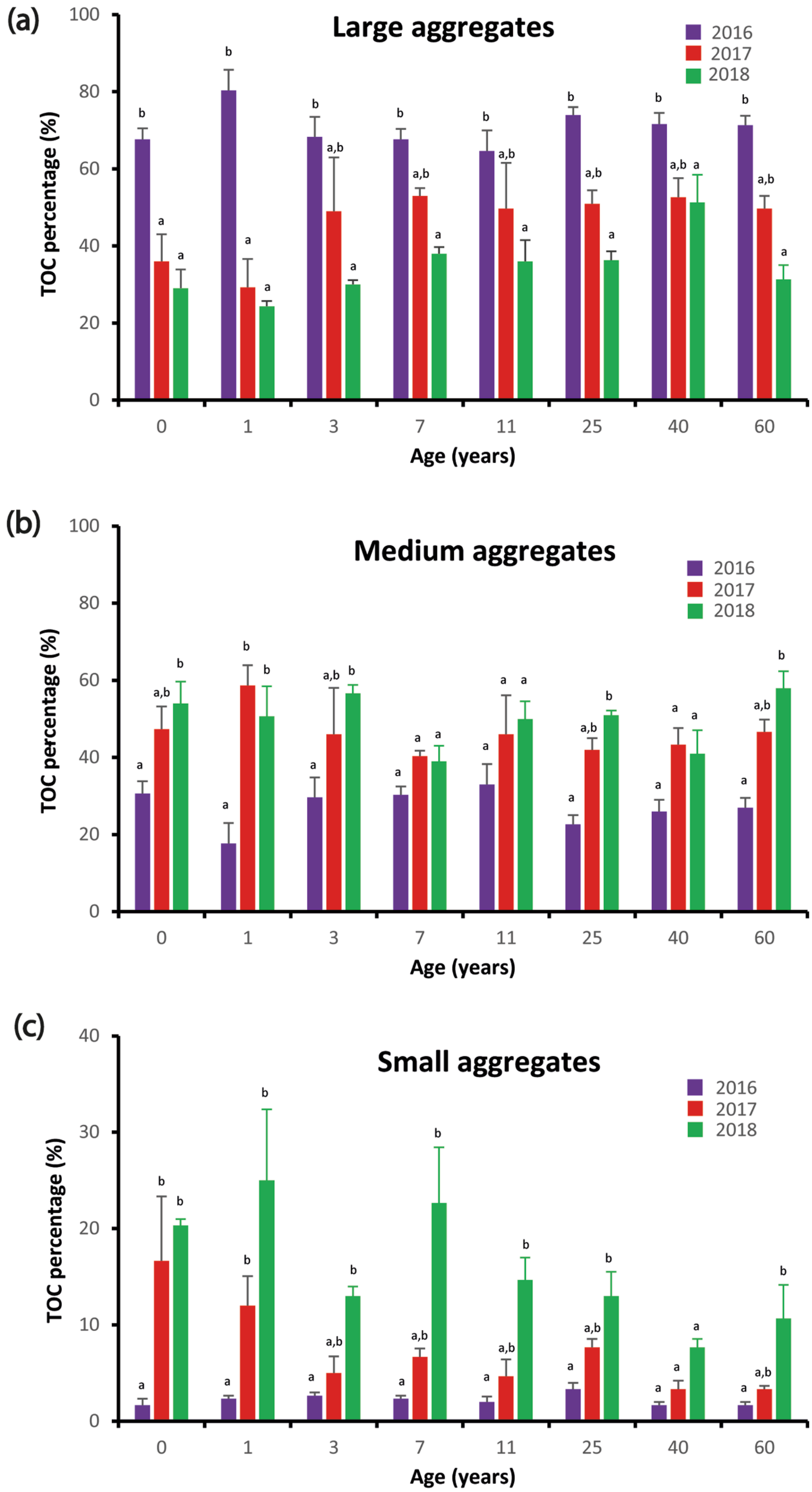

Fig. 4. Variations in TOC percentage throughout the chronosequence in 2016, 2017 and 2018. (a) Large aggregates (2–0.5 mm), (b) Medium aggregates (0.5–0.05 mm), (c) Small aggregates (<0.05 mm). Error bars represent standard error (n = 3).

Table 2
Linear correlation values/relationships (r Pearson) between aggregate fractions and total and water soluble Carbon and Nitrogen (n = 24). Total Organic carbon = TOC; Total Nitrogen = TN; Water Soluble Organic Carbon = WSOC; Water Soluble Nitrogen = WSN; LA = Large aggregates (2–0.5 mm); MA = Medium aggregates (0.5–0. 05 mm); SA = Small aggregates (<0.05 mm).

		Aggregate weight percentage			TOC content in aggregates (g kg^{-1})			TOC percentage in aggregates		
		LA	MA	SA	LA	MA	SA	LA	MA	SA
TOC	2016	**0.572****	**−0.450***	**−0.545****	**0.744*****	**0.853*****	**0.572****	0.331	−0.3	−0.143
	2017	**0.433***	−0.353	**−0.666*****	**0.608****	**0.658*****	0.168	0.385	−0.161	**−0.613****
	2018	0.258	−0.076	**−0.486***	**0.736*****	**0.783*****	**0.771*****	0.245	0.221	**−0.541****
TN	2016	0.297	−0.184	**−0.459***	**0.792*****	**0.875*****	**0.834*****	0.093	−0.071	−0.223
	2017	0.301	−0.232	**−0.587****	**0.451***	**0.593****	0.116	0.228	−0.014	**−0.510***
	2018	0.181	0.014	**−0.439***	**0.746*****	**0.824*****	**0.705*****	0.147	0.297	**−0.559****
WSOC	2016	**0.499***	**−0.461***	−0.391	0.302	0.345	0.355	**0.431***	−0.394	−0.196
	2017	0.155	−0.154	−0.262	0.318	0.133	−0.074	0.271	−0.213	0.237
	2018	0.188	−0.515	**−0.618****	**0.698*****	**0.710*****	**0.684*****	0.216	0.278	**−0.617****
WSN	2016	**−0.492***	**0.472***	0.265	−0.121	−0.206	−0.275	−0.276	0.314	−0.209
	2017	**0.483***	**−0.421***	**−0.596****	**0.523***	**0.508***	0.005	**0.442***	−0.244	**−0.611****
	2018	0.267	−0.045	**−0.601****	**0.827*****	**0.875*****	**0.807*****	0.273	0.354	**−0.733*****

*p < .05; **p < .01; ***p < .001.

Table 3
Linear correlation values/relationships (r Pearson) between soil organic fraction and enzyme activities related to the C cycle. Total Organic carbon = TOC: Total Nitrogen = TN, Water Soluble Organic Carbon = WSOC; Water Soluble Nitrogen = WSN; (n = 24). LA = Large aggregates (2–0.5 mm); MA = Medium aggregates (0.5–0.05 mm); SA = Small aggregates (<0.05 mm).

	α-Glucosidase			β-Glucosidase			β-Galactosidase			Phenoloxidase		
	2016	2017	2018	2016	2017	2018	2016	2017	2018	2016	2017	2018
TOC	**0.702*****	**0.905*****	**0.735****	**0.784*****	**0.857*****	**0.783*****	**0.477***	**0.850*****	0.404	**−0.549****	0.067	0.349
TN	**0.565****	**0.915*****	**0.813*****	**0.659*****	**0.780*****	**0.836*****	0.317	**0.723*****	**0.475***	−0.321	0.202	0.298
WSOC	0.270	0.334	**0.645****	0.226	0.339	**0.593****	0.134	**0.532****	0.255	**−0.405***	−0.221	0.188
WSN	−0.1555	**0.852*****	**0.763*****	−0.136	**0.815*****	**0.808*****	−0.128	**0.707*****	0.290	0.358	0.061	0.158
LA weight	**0.405***	**0.442***	0.272	**0.433***	**0.468***	0.249	0.294	**0.626****	−0.335	**0.609****	**0.422***	−0.083
MA weight	−0.275	**−0.412***	−0.049	−0.280	**−0.453***	−0.044	−0.205	**−0.609****	0.404	**0.488***	**−0.417***	0.091
SA weight	**−0.451***	**−0.701*****	**−0.631****	**−0.477***	**−0.744*****	**−0.549****	−0.092	**−0.782*****	−0.043	**0.456***	−0.227	0.186
LA content	0.172	0.312	0.124	0.177	0.269	0.103	0.183	**0.519****	**−0.461***	−0.166	0.209	0.050
MA content	−0.166	−0.029	0.308	−0.172	−0.033	0.340	−0.163	−0.310	**0.510***	0.217	−0.101	−0.091
SA content	−0.028	**−0.710*****	**−0.576****	−0.055	**−0.652****	**−0.584****	−0.035	**−0.698*****	−0.021	−0.228	−0.329	0.056
LA percentage	**0.522****	**0.498****	**0.682*****	**0.628****	**0.422***	**0.667****	0.401	**0.590****	0.086	−0.177	0.192	0.012
MA percentage	**0.628****	**0.699*****	**0.794*****	**0.740*****	**0.693*****	**0.800*****	0.443*	**0.670*****	0.301	**−0.418***	0.393	−0.042
SA percentage	**0.655****	0.159	**0.802*****	**0.773*****	0.220	**0.773*****	0.377	0.288	0.291	**−0.588****	0.023	−0.098

*p < .05; **p < .01; ***p < .001.

Table 4
Linear correlation values/relationships (r Pearson) between soil organic fraction and enzyme activities related to the N cycle. Total Organic carbon = TOC: Total Nitrogen = TN, Water Soluble Organic Carbon = WSOC; Water Soluble Nitrogen = WSN; (n = 24). LA = Large aggregates (2–0.5 mm); MA = Medium aggregates (0.5–0.05 mm); SA = Small aggregates (<0.05 mm).

	N-Arylamidase			*N*-Acetyl-Glucosaminidase			Urease		
	2016	2017	2018	2016	2017	2018	2016	2017	2018
TOC	**0.570****	**0.896*****	0.250	0.366	**0.829*****	**0.593****	**0.472***	**0.667*****	**0.573****
TN	0.390	**0.856*****	0233	**0.417***	**0.776*****	**0.751*****	0.101	**0.622****	**0.785*****
WSOC	**0.455***	**0.411***	**0.421***	−0.168	0.098	**0.548****	**0.572****	0.251	**0.679****
WSN	−0.014	**0.745*****	**0.467***	0.095	**0.671*****	**0.586****	−0.342	**0.503***	**0.676****
LA weight	0.157	**0.456***	0.380	0.261	0.395	0.119	**0.554****	0.510*	0.252
MA weight	−0.122	**−0.428***	−0.203	−0.160	−0.403	0.053	**−0.516****	**−0.534****	−0.033
SA weight	−0.090	**−0.680*****	**−0.690*****	−0.259	**−0.663*****	**−0.421***	−0.385	**−0.441***	**−0.685*****
LA content	0.063	0.235	0.264	−0.018	0.238	0.068	0.395	0.317	0.088
MA content	−0.049	0.034	0.117	0.002	−0.034	0.271	−0.390	−0.139	0.337
SA content	0.097	**−0.703*****	**−0.490***	0.131	**−0.602***	**−0.437***	0.093	**−0.542****	**−0.591****
LA percentage	**0.444***	**0.427***	**0.432***	0.041	0.299	**0.527****	0.237	0.351	**0.658*****
MA percentage	**0.504***	**0.695*****	**0.440***	0.236	**0.535****	**0.582****	0.333	**0.574****	**0.775*****
SA percentage	**0.448***	0.137	**0.550****	0.347	0.049	**0.486***	**0.410***	0.125	**0.755*****

*p < .05; **p < .01; ***p < .001.

correlations were found between the weight of the macroaggregates and the activity of the enzymes related to the C and N cycles before the drought period. Drought events cause a decrease in the biological activity of soils (Sheik et al., 2011; Schimel, 2018) which could have important consequences for the structural stability of the soil. We found significant structural destabilization of the soil in the post-drought year leading to the loss of the relationship between macroaggregates and enzyme activity. Changes in the soil structure affect soil microbial activity and greenhouse gas emissions (Smith et al., 2017). Soil shrinkage also affects the size and stability of the pores, and was most pronounced in soils with a large proportion of macropores, such as the soils in this study. This structural destabilization of the soil causes a disruption in the carbon cycle, which switches from being a sink to a source of C, as demonstrated by the decrease in the soil TOC content (Quintana et al., 2021).

The drought produced a significant increase in WSN, followed by a sharp decline in the post-drought year. The drought year showed a positive relationship between WSN and *N*-cycle-related enzymes, and also with enzyme activities related to the labile C-cycle, which were maintained in the following year. The increase in N is likely due to the breakdown of macroaggregates, causing the organic matter, which is rich in nitrogen compounds, to become available during drought events. Nitrogen mineralization often increases when dry soils are rewetted (Leitner et al., 2017; Saetre and Stark, 2005). The material mobilized by rewetting is rich enough in nitrogen to provide surplus nitrogen for microbes as they regrow, so they can mineralize nitrogen. Potential nitrogen-rich substrates that could fuel such a flush include bacterial osmolytes, microbial necromass (Liang et al., 2017), and nitrogen-rich but clay-protected small molecules (Kleber et al., 2007).

Aggregate formation is promoted by the aggregation of particles such as clay, Fe and Mn oxides, exchangeable divalent cations and organic matter (Rowley et al., 2018; Pronk et al., 2013). Land abandonment leads to small increments in these components that have important effects on the stabilization of TOC and TN (Valverde-Asenjo et al., 2020). Our results show that this increase occurred in all aggregate size classes, although most of the TOC was linked to the LA fraction. A long period of abandonment produces an increase in the percentage of LA, which is precisely the fraction most affected by drought. Thus, the longer the time since the land abandonment, the more changes in soil organic matter in the event of extreme drought. The loss of protection for the organic matter caused the soil aggregates to break down when the drought was interrupted, and, together with the higher enzyme activity and higher TOC content in the plots that had been abandoned for longest (Quintana et al., 2021), meant that these lands became more exposed to the effects of the extreme drought event.

5. Conclusions

Although there are many laboratory and experimental studies on the effect of drought on soils, our work represents a contribution to the research in field conditions. The results of our study supported our initial hypothesis. An extreme drought event had a major impact on the functionality of the soils since it produced a strong structural destabilization that had the most severe effect on the largest aggregates. The decomposition of the aggregates exposed the organic matter that had previously been protected, and combined with the enhanced activity of the enzymes responsible for their mineralization, caused a serious disturbance to soil functionality. When the environmental conditions were favourable, unprotected organic matter was rapidly consumed by soil microorganisms. This implies that under normal conditions ecosystems that function as carbon sinks become sources of CO_2 to the atmosphere, in a positive feedback mechanism for climate change. The time since the abandonment of agricultural activity is related to an increase in TOC and the large aggregate fraction, so the plots abandoned for longest suffered the most severe effects on the destabilization of the C and N cycles.

Declaration of Competing Interest

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

Data availability

Data will be made available on request.

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References

Acosta-Martínez, V., Cotton, J., Gardner, T., Moore-Kucera, J., Zak, J., Wester, D., Cox, S., 2014. Predominant bacterial and fungal assemblages in agricultural soils during a record drought/heat wave and linkages to enzyme activities of biogeochemical cycling. Appl. Soil Ecol. 84, 69–82. https://doi.org/10.1016/j.apsoil.2014.06.005.

Bach, E.M., Hofmockel, K.S., 2014. Soil aggregate isolation method affects measures of intra-aggregate extracellular enzyme activity. Soil Biol. Biochem. 69, 54–62. https://doi.org/10.1016/j.soilbio.2013.10.033.

Bach, E.M., Williams, R.J., Hargreaves, S.K., Yang, F., Hofmockel, K.S., 2018. Greatest soil microbial diversity found in micro-habitats. Soil Biol. Biochem. 118, 217–226. https://doi.org/10.1016/j.soilbio.2017.12.018.

Bakker, M.M., Govers, G., Kosmas, C., Vanacker, V., Van Oost, K., Rounsevell, M., 2005. Soil erosion as a driver of land-use change. Agric. Ecosyst. Environ. 105, 467–481. https://doi.org/10.1016/j.agee.2004.07.009.

Barnard, R.L., Osborne, C.A., Firestone, M.K., 2013. Responses of soil bacterial and fungal communities to extreme desiccation and rewetting. ISME J 7, 2229–2241. https://doi.org/10.1038/ismej.2013.104.

Borken, W., Matzner, E., 2009. Reappraisal of drying and wetting effects on C and N mineralization and fluxes in soils. Glob. Change Biol. 15, 808–824. https://doi.org/10.1111/j.1365-2486.2008.01681.x.

Chodak, M., Gołebiewski, M., Morawska-Płoskonka, J., Kuduk, K., Niklińska, M., 2015. Soil chemical properties affect the reaction of forest soil bacteria to drought and rewetting stress. Ann. Microbiol. 65, 1627–1637. https://doi.org/10.1007/s13213-014-1002-0.

Ciais, P.h., Reichstein, M., Viovy, N., Granier, A., Ogée, J., Allard, V., Aubinet, M., Buchmann, N., Bernhofer, C., Carrara, A., Chevallier, F., De Noblet, N., Friend, A.D., Friedlingstein, P., Grünwald, T., Heinesch, B., Keronen, P., Knohl, A., Krinner, G., Loustau, D., Manca, G., Matteucci, G., Miglietta, F., Ourcival, J.M., Papale, D., Pilegaard, K., Rambal, S., Seufert, G., Soussana, J.F., Sanz, M.J., Schulze, E.D., Vesala, T., Valentini, R., 2005. Europe-wide reduction in primary productivity caused by the heat and drought in 2003. Nature 437, 529–533. https://doi.org/10.1038/nature03972.

Clary, J., 2008. Rainfall seasonality determines annual/perennial grass balance in vegetation of Mediterranean Iberian. Plant Ecol. 195, 13–20. https://doi.org/10.1007/s11258-007-9294-9.

Cleveland, C.C., Wieder, W.R., Reed, S.C., Townsend, A.R., 2010. Experimental drought in a tropical rain forest increases soil carbon dioxide losses to the atmosphere. Ecology 91, 2313–2323. https://doi.org/10.1890/09-1582.1.

Consorcio Sierra Oeste, 2009. Análisis del subsistema físico natural. In: Estrategia de Desarrollo Comarcal de la Sierra Oeste de Madrid, Madrid, Spain, pp. 2007–2013.

Cotrufo, M.F., Wallenstein, M.D., Boot, C.M., Denef, K., Paul, E., 2013. The Microbial Efficiency-Matrix Stabilization (MEMS) framework integrates plant litter decomposition with soil organic matter stabilization: do labile plant inputs form stable soil organic matter? Glob. Change Biol. 19, 988–995. https://doi.org/10.1111/gcb.12113.

Dai, A., 2011. Drought under global warming: a review. WIREs Clim. Change 2, 45–65. https://doi.org/10.1002/wcc.81.

Davinic, M., Fultz, L.M., Acosta-Martinez, V., Calderón, F.J., Cox, S.B., Dowd, S.E., Allen, V.G., Zak, J.C., Moore-Kucera, J., 2012. Pyrosequencing and mid-infrared spectroscopy reveal distinct aggregate stratification of soil bacterial communities and organic matter composition. Soil Biol. Biochem. 46, 63–72. https://doi.org/10.1016/j.soilbio.2011.11.012.

De Boeck, H.J., Dreesen, F.E., Janssens, I.A., Nijs, I., 2011. Whole-system responses of experimental plant communities to climate extremes imposed in different seasons. New Phytol. 189, 806–817. https://doi.org/10.1111/j.1469-8137.2010.03515.x.

De Vries, F.T., Griffiths, R.I., Bailey, M., et al., 2018. Soil bacterial networks are less stable under drought than fungal networks. Nat. Commun. 9, 3033. https://doi.org/10.1038/s41467-018-05516-7.

Degens, B.P., 1997. Macro-aggregation of soils by biological bonding and binding mechanisms and the factors affecting these: a review. Soil Res. 35, 431–460. https://doi.org/10.1071/S96016.

Denef, K., Six, J., 2005. Clay mineralogy determines the importance of biological versus abiotic processes for macroaggregate formation and stabilization. Eur. J. Soil Sci. 56, 469–479. https://doi.org/10.1111/j.1365-2389.2004.00682.x.

Doblas-Miranda, E., Martínez-Vilalta, J., Lloret, F., Álvarez, A., Ávila, A., Bonet, F.J., Brotons, L., Castro, J., Curiel Yuste, J., Díaz, M., Ferrandis, P., García-Hurtado, E., Iriondo, J.M., Keenan, T.F., Latron, J., Llusià, J., Loepfe, L., Mayol, M., Moré, G., Moya, D., Peñuelas, J., Pons, X., Poyatos, R., Sardans, J., Sus, O., Vallejo, V.R., Vayreda, J., Retana, J., 2015. Reassessing global change researchpriorities in mediterranean terrestrialecosystems: how far have we come andwhere do we go from here? Glob. Ecol. Biogeogr. 24, 25–43. https://doi.org/10.1111/geb.12224.

Frank, D., Reichstein, M., Bahn, M., Thonicke, K., Frank, D., Mahecha, M.D., Smith, P., van der Velde, M., Vicca, S., Babst, F., Beer, C., Buchmann, N., Canadell, J.G., Ciais, P., Cramer, W., Ibrom, A., Miglietta, F., Poulter, B., Rammig, A., Seneviratne, S.I., Walz, A., Wattenbach, M., Zavala, M.A., Zscheischler, J., 2015. Effects of climate extremes on the terrestrial carbon cycle: concepts, processes and potential future impacts. Glob. Change Biol. 21, 2861–2880. https://doi.org/10.1111/gcb.12916.

Ghani, A., Dexter, M., Perrott, K.W., 2003. Hot-water extractable carbon in soils: A sensitive measurement for determining impacts of fertilisation, grazing and cultivation. Soil Biol. Biochem. 35, 1231–1243. https://doi.org/10.1016/S0038-0717(03)00186-X.

Guidi, C., Magid, J., Rodeghiero, M., Gianelle, D., Vesterdal, L., 2014. Effects of forest expansion on mountain grassland: changes within soil organic carbon fractions. Plant Soil. 385, 373–387. https://doi.org/10.1007/s11104-014-2315-2.

Hoover, D.L., Rogers, B.M., 2016. Not all droughts are created equal: the impacts of interannual drought pattern and magnitude on grassland carbon cycling. Glob. Change Biol. 22, 1809–1820. https://doi.org/10.1111/gcb.13161.

Horion, S., Ivits, E., De Keersmaecker, W., Tagesson, T., Vogt, J., Fensholt, R., 2019. Mapping European ecosystem change types in response to land-use change, extreme climate events, and land degradation. Land Degrad. Dev. 30, 951–963. https://doi.org/10.1002/ldr.3282.

IPCC, 2022. Climate change 2022: impacts, adaptation and vulnerability. In: Pörtner, H.-O., Roberts, D.C., Tignor, M., Poloczanska, E.S., Mintenbeck, K., Alegría, A., Craig, M., Langsdorf, S., Löschke, S., Möller, V., Okem, A., Rama, B. (Eds.), Contribution of Working Group II to the Sixth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge University Press. Cambridge University Press, Cambridge, UK and New York, NY, USA, 3056 pp. doi: 10.1017/9781009325844.

ISO, 2018. ISO 20130 Soil quality-Measurement of enzyme activity patterns in soil samples using colorimetric substrates in micro-wells plates.

Keenleyside, C., Tucker, G., 2010. Farmland Abandonment in the EU: An Assessment of Trends and Prospects; Report prepared for WWF. Institute for European Environmental Policy, London, UK.

Kleber, M., Sollins, P., Sutton, R.A., 2007. Conceptual model of organo-mineral interactions in soils: self-assembly of organic molecular fragments into zonal structures on mineral surfaces. Biogeochemistry 85, 9–24. https://doi.org/10.1007/s10533-007-9103-5.

Leitner, S., Homyak, P.M., Blankinship, J.C., Eberwein, J., Jenerette, G.D., Zechmeister-Boltenstern, S., Schimel, J.P., 2017. Linking NO and N2O emission pulses with the mobilization of mineral and organic N upon rewetting dry soils. Soil Biol. Biochem. 115, 461–466. https://doi.org/10.1016/j.soilbio.2017.09.005.

Li, L., Zheng, Z., Biederman, J.A., Qian, R., Ran, Q., Zhang, B., Xu, C., Wang, F., Zhou, S., Che, R., Dong, J., Xu, Z., Cui, X., Hao, Y., Yanfen Wang, Y., 2020. Drought and heat wave impacts on grassland carbon cycling across hierarchical levels. Plant Cell Environ. 2021 (44), 2402–2413. https://doi.org/10.1111/pce.13767.

Liang, C., Schimel, J., Jastrow, J., 2017. The importance of anabolism in microbial control over soil carbon storage. Nat. Microbiol. 2, 17105. https://doi.org/10.1038/nmicrobiol.2017.105.

Liu, M., Guilin, H., Zhang, Q., 2019. Effects of soil aggregate stability on soil organic carbon and nitrogen under land use change in an erodible region in southwest China. Int. J. Environ. Res. Public Health 16 (20), 3809. https://doi.org/10.3390/ijerph16203809.

Lozano, Y.M., Hortal, S., Armas, C., Pugnaire, F.I., 2014. Interactions among soil, plants, and microorganisms drive secondary succession in a dry environment. Soil Biol. Biochem. 78, 298–306. https://doi.org/10.1016/j.soilbio.2014.08.007.

Mambelli, S., Bird, J.A., Gleixner, G., Dawson, T.E., Torn, M.S., 2011. Relative contribution of foliar and fine root pine litter to the molecular composition of soil organic matter after in situ degradation. Org. Geochem. 42, 1099–1108. https://doi.org/10.1016/j.soilbio.2021.108347.

Molina, J.A., Martín-Sanz, J.P., Valverde-Asenjo, I., et al., 2022. Mediterranean grassland succession as an indicator of changes in ecosystem biodiversity and functionality. Biodivers. Conserv. https://doi.org/10.1007/s10531-022-02481-y.

Navarro-García, F., Casermeiro, M.A., Schimel, J.P., 2012. When structure means conservation: effect of aggregate structure in controllingmicrobial responses to rewetting events. Soil Biol. Biochem. 44, 1–8. https://doi.org/10.1016/j.soilbio.2011.09.019.

Novara, A., Gristina, L., Sala, G., Galati, A., Crescimanno, M., Cerdà, A., Badalamenti, E., La Mantia, T., 2017. Agricultural land abandonment in Mediterranean environment provides ecosystem services via soil carbon sequestration. Sci. Total Environ. 576, 420–429. https://doi.org/10.1016/j.scitotenv.2016.10.123.

Pronk, G.J., Heister, K., Kögel-Knabner, I., 2013. Is turnover and development of organic matter controlled by mineral composition? Soil Biol. Biochem. 67, 235–244. https://doi.org/10.1016/j.soilbio.2013.09.006.

Quintana, J.R., Molina, J.A., Diéguez-Antón, A., Valverde-Asenjo, I., 2021. Interannual climate variability determines the efficiency of functional recovery in dry Mediterranean abandoned vineyards. Land Degrad. Dev. 2020, 1–18. https://doi.org/10.1002/ldr.3843.

Reichstein, M., Bahn, M., Ciais, P., Frank, D., Mahecha, M.D., Seneviratne, S.I., Zscheischler, J., Beer, C., Buchmann, N., Frank, D.C., Papale, D., Rammig, A., Smith, P., Thonicke, K., van der Velde, M., Vicca, S., Walz, A., Wattenbach, M., 2013. Climate extremes and the carbon cycle. Nature, 500, 7462, 287–95. doi: 10.1038/nature12350. PMID: 23955228.

Rey Benayas, J.M., Martins, A., Nicolau, J.M., Schulz, J., 2007. Abandonment of agricultural land: An overview of drivers and consequences. CAB Rev.: Perspect. Agric. Vet. Sci. Nutr. Nat. Resour. 2, 57. https://doi.org/10.1079/PAVSNNR20072057.

Rivas-Martínez, S., 1982. Mapas de las series de vegetación de Madrid. Diputación Provincial de Madrid Mapa 1 (200), 000.

Rivas-Martínez, S., 2007. Mapa de series, geoseries y geopermaseries de vegetación de España (Memoria del Mapa de Vegetación Potencial de España. Parte 1). Itinera Geobot. 17, 5–436.

Rowley, M.C., Grand, S., Verrecchia, É.P., 2018. Calcium-mediated stabilisation of soil organic carbon. Biogeochemistry 137, 27–49. https://doi.org/10.1007/s10533-017-0410-1.

Rundel, P.W., Arroyo, M.T., Cowling, R.M., Keeley, J.E., Lamont, B.B., Vargas, P., 2016. Mediterranean biomes: evolution of their vegetation, floras, and climate. Annu. Rev. Ecol. Evol. Syst. 47, 383–407. https://doi.org/10.1146/annurev-ecolsys-121415-032330.

Saetre, P., Stark, J.M., 2005. Microbial dynamics and carbon and nitrogen cycling following re-wetting of soils beneath two semi-arid plant species. Oecologia 142, 247–260. https://doi.org/10.1007/s00442-004-1718-9.

Schimel, J.P., 2018. Life in dry soils: effects of drought on soil microbial communities and processes. Annu. Rev. Ecol. Evol. Syst. 49, 409–432. https://doi.org/10.1146/annurev-ecolsys-110617-062614.

Schimel, J.P., Schaeffer, S.M., 2012. Microbial control over carbon cycling in soil. Front. Microbiol. 3, 348. https://doi.org/10.3389/fmicb.2012.00348.

Sheik, C., Beasley, W., Elshahed, M., et al., 2011. Effect of warming and drought on grassland microbial communities. ISME J. 5, 1692–1700. https://doi.org/10.1038/ismej.2011.32.

Six, J., Paustian, K., Elliott, E.T., Combrink, C., 2000. Soil structure and soil organic matter: I. Distribution of aggregate size classes and aggregate associated carbon. Soil Sci. Soc. Am. J. 64, 681–689. https://doi.org/10.2136/sssaj2000.642681x.

Six, J., Bossuyt, H., Degryze, S., Denef, K., 2004. A history of research on the link between (micro)aggregates, soil biota, and soil organic matter dynamics. Soil Tillage Res. 79, 7–31. https://doi.org/10.1016/j.still.2004.03.008.

Smith, A., Bond-Lamberty, B., Benscoter, B.W., Tfaily, M.M., Hinkle, C.R., Liu, C., Bailey, V.L., 2017. Shifts in pore connectivity from precipitation versus groundwater rewetting increases soil carbon loss after drought. Nat. Commun. 8, 1335. https://doi.org/10.1038/s41467-017-01320-x.

Su, X., Su, X., Zhou, G., Du, Z., Yang, S., Ni, M., Qin, H., Huang, Z., Zhou, X., Deng, J., 2020. Drought accelerated recalcitrant carbon loss by changing soil aggregation and microbial communities in a subtropical forest. Soil Biol. Biochem. 148 (2020), 107898 https://doi.org/10.1016/j.soilbio.2020.107898.

Tang, J., Mo, Y., Zhang, J., Zhang, R., 2011. Influence of biological aggregating agents associated with microbial population on soil aggregate stability. Appl. Soil Ecol. 47, 153–159. https://doi.org/10.1016/j.apsoil.2011.01.001.

Valverde-Asenjo, I., Diéguez-Antón, A., Martín-Sanz, J.P., Molina, J.A., Quintana, J.R., 2020. Soil and vegetation dynamics in a chronosequence of abandoned vineyards. Agric. Ecosyst. Environ. 301, 107049 https://doi.org/10.1016/j.agee.2020.107049.

Vaquero Perea, C., Valverde-Asenjo, I., Vázquez de la Cueva, A., Martín-Sanz, J.P., Molina, J.A., Quintana, J.R., 2020. Colonizing vegetation type drives evolution of organic matter in secondary succession in abandoned vineyards. Plant Ecol. 221, 1143–1158. https://doi.org/10.1007/s11258-020-01069-2.

Walkley, A., Black, I.A., 1934. An examination of the Degtjareff method for determining soil organic matter and proposed modification of chromic acid tritation. Soil Sci. 37, 317–327.

WRBI, 2015. Reference base for soil resources 2014, update 2015. International soil classification system for naming soils and creating legends for soil maps. No. 106. FAO, Rome, Italy.

Yilmaz, E., Çanakci, M., Topakci, M., Sönmez, S., Ağsaran, B., Alagöz, Z., Çitak, S., Uras, D.S., 2019. Effect of vineyard pruning residue application on soil aggregate formation, aggregate stability and carbon content in different aggregate sizes. Catena 183, 104219. https://doi.org/10.1016/j.catena.2019.104219.

Zhang, Q., Shao, M., Jia, X., Wei, X., 2019. Changes in soil physical and chemical properties after short drought stress in semi-humid forests. Geoderma 338, 170–177. https://doi.org/10.1016/j.geoderma.2018.11.051.

Zhou, Z., Wang, C., Jiang, L., Luo, Y., 2017. Trends in soil microbial communities during secondary succession. Soil Biol. Biochem. 115, 92e99. https://doi.org/10.1016/j.soilbio.2017.08.014.