



# Seasonal changes in soil carbon storage capacity and glomalin-related soil protein under different agricultural activities, abandonment, and wildfire occurrence in Mediterranean region

Mohamed Emran<sup>a,\*</sup>, Huda Naeim<sup>a</sup>, Mohamed Rashad<sup>a</sup>, Maria Gispert<sup>b</sup>

<sup>a</sup> City of Scientific Research and Technological Applications (SRTA-City), 21934 New Borg El-Arab City, Alexandria, Egypt

<sup>b</sup> Soil Science Unit, University of Girona, C/Aurèlia Capmany, 61, 17003 Girona, Spain

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## ABSTRACT

Agricultural practices can directly affect soil environmental conditions and consequently soil ecosystems and fertility. Soils under different agricultural practices like tillage and grazing were studied and compared to other soils affected by wildfire and different ages of abandonment. Atmospheric CO<sub>2</sub>, soil C-CO<sub>2</sub> loss, soil temperature, soil moisture, bulk density, soil organic carbon (SOC), total nitrogen (TN), total phosphorus (TP), Redfield ratios of C:N:P, easily extractable and total glomalin-related soil protein (GRSP<sub>EE</sub> and GRSP<sub>T</sub>), and morphology, quantity, and stability of soil aggregates (WSA) were seasonally analyzed. Statistical significance checked using the two-factor ANOVA. Intense tillage and wildfire occurrence revealed a deficiency in soil nutrients and fertility more than grazing practices. Steady soil fertility conditions occurred in soils preserved by fire and ancient abandonment. The GRSP seasonal means varied significantly among all soils because of land-use effects. GRSP<sub>T</sub>, SOC, TN, and TP increased from winter to spring and reached the highest values in summer then decreased again in autumn. GRSP was positively correlated ( $P < 0.01$ ) with atmospheric CO<sub>2</sub>, SOC, TN, and TP with high peaks in summer indicating the dependent GRSP production on plant nutrients acquisition as well as soil carbon pools. Moreover, the correlation between GRSP and emitted C-CO<sub>2</sub> may indicate the contribution of GRSP fractions to the labile as well as the stable SOC fractions. The GRSP<sub>EE</sub> showed rapidly transformed into stable forms in more fertile and better-structured soils than in less fertile soils with vulnerable soil structures. In conclusion, the stoichiometric balance of soil nutrients, aggregate stability, and stabilized GRSP pools improved relatively across the seasonal rhythms from winter to summer. Moreover, environmental soil conditions developed to preserve organic carbon in soils with long abandonment than in mid abandonment, while the worst conditions were in the vulnerable soils intensively mismanaged and devastated by fire.

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## 1. Introduction

The accumulation of soil organic matter (SOM) in the upper soil layers in the abandoned lands could be attributed to the aboveground plant residues (Cao et al., 2020). SOM deposits are generally increased due to the seasonal additions of the aboveground vegetation (Turner et al., 2015; Gispert et al., 2018; 2021). Accordingly,

land abandonment may produce beneficial effects on soil conditions by developing the stabilization mechanisms of the deposited organic materials in soil (Ovsepyan et al., 2019). This process may rebuild the soil organic matter (SOM) pools along with the soil profile when the renaturalization of the soil ecosystem resulted in the spontaneous colonization of natural plant species (Cerdà et al., 2019). In poorly structured soils with low clay contents, stable organic compounds can be considered as the most vital soil constituent for increasing soil structural stability and nourish soil health. However, the frequent wildfire occurrence together with basic agricultural practices in cultivated vines and olive groves and grazing in meadows produced soil compaction which may negatively affect the organic compounds in soil by altering the dynamics of soil microbial communities with a consequent imbalance of nutrients cycling (Emran et al., 2012a; Gispert et al., 2018).

\* Corresponding author.

E-mail address: [memran@srtacity.sci.eg](mailto:memran@srtacity.sci.eg) (M. Emran).

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Glomalin is a mycorrhizal glycoprotein (Wright and Upadhyaya, 1996) positively contributed to supply soil nutrients, with special emphasis on soil organic carbon (SOC) pools, hence, mycorrhizal symbioses provided plants with soil micro- and macro-nutrients (Wright et al., 2006; Emran et al., 2017; Gispert et al., 2018). Glomalin has been operationally recognized as a glomalin-related soil protein (GRSP) and many authors have emphasized its relevant role in the stability of SOM (Wang et al., 2018; Emran et al., 2020). The potentiality of soil carbon storage capacity increased GRSP production thus minimized the soil carbon loss in the ancient abandoned lands of Lithic Xerorthents (Gispert et al., 2017; 2021). This picture had also been observed in Haplic Calcisols under cultivated and abandoned olive groves under semi-arid Mediterranean climate (Hontoria et al., 2009).

Increasing plant nutrients acquisition occurs as a result of atmospheric CO<sub>2</sub> increase by stimulating the mycorrhizal fungi to produce more glomalin and overcome the abiotic stresses (Rillig et al., 2001; Treseder et al., 2003). Recent studies of Zhang et al. (2015) have proved that elevated atmospheric CO<sub>2</sub> significantly increased GRSP<sub>T</sub> by 35% though GRSP<sub>EE</sub> decreased probably due to the irregular symbiotic activity between Arbuscular Mycorrhizal Fungi (AMF) and their host plants. Accordingly, increasing GRSP contents as a result of soil carbon assimilation should underline its value as a mitigating agent of global climate by increasing soil carbon storage capacity (Gispert et al., 2021).

This work aimed to investigate the seasonal changes in soil carbon storage capacity and carbon loss and their relation to GRSP concentrations, soil environmental conditions, the stoichiometric balance of soil nutrients, and formation and stability of soil aggregates under changes in agricultural management practices, wildfire occurrence, and age of abandonment in the North-Eastern Iberian Peninsula.

## 2. Materials and methods

### 2.1. Study area

It is located in the Alt Empordà, Figueres, North-Eastern Iberian Peninsula (Fig. 1). It is described as a typical Mediterranean environment subjected to severe changes in land use and abandonment in the last century. The seasonal atmospheric temperature fluctuated from 33 to 36 °C in summer to –5 – –12 °C in winter. The mean seasonal precipitation recorded 13, 82, 55, and 162 mm h<sup>-1</sup> in winter, spring, summer, and autumn. Averaged atmospheric CO<sub>2</sub> concentrations resulted in 270 ppm in winter, 323 ppm in spring, 460 ppm in summer, and 448 ppm in autumn during the investigation period (<https://www.accuweather.com/en/es/figueres/304361/weather-forecast/304361>).

Seven soils were selected representing the common local succession sequence after abandonment with different land uses and abandonment ages. The changes in land use and age of abandonment may represent the temporal and spatial changes in soil organic matter dynamics (Poeplau et al., 2011). Agricultural soils under *Vitis vinifera* L. (S1) and *Olea europaea* L. (S2), where S1 received more mechanical tillage and agrochemical additions than S2. Soils under recently (20 years) abandoned stands of *Pinus halepensis* Mill trees (S3) and *Quercus suber* L. (S4) were selected and S3 was frequently affected by wildfire. Ancient (60 years) abandoned soils under *Cistus monspeliensis* L. (S5), scrub annually sheared at the beginning of summer season to prevent wildfire occurrence, and under *Erica arborea* L. (S6) scrub, more preserved by wildfire 35 years ago. The soils under *Brachypodium retusum* Pers. (S7) meadows are annually receiving cattle grazing activities mainly in winter and spring.

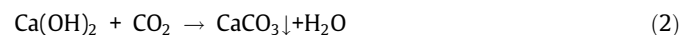
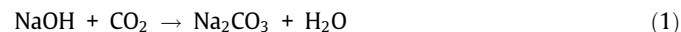
All the studied soils classified as Lithic Xerorthents (Soil Survey Staff, 2014) formed on Palaeozoic slates lithic contacts of <50 cm of the mineral soil surface. Soils were shallow with A horizon of 5–10 cm depth over a rather stony C/R horizon.

### 2.2. Experimental analysis

Soils were sampled from the upper 20 cm depth. Three soil samples were monthly collected from each site and the representative seasonal means were then presented. Samples were air-dried, sieved at 2 mm, and stored for the subsequent analyses. Soil particle analysis using Robinson's pipette (Eijkelkamp Agriresearch Equipment, Giesbeek, The Netherlands), bulk density (BD) using core method, and soil moisture (SM) using gravimetric method were analyzed. Soil organic carbon (SOC) is determined by the dichromate oxidation method, total nitrogen (TN) by the Kjeldahl method, and total phosphorus (TP) by the digestion-spectrophotometric method according to Ryan et al. (2001). The molar ratio of C:P and C:N ratios were calculated. All soil analyses were carried out in triplicates.

At each sampling location, soil temperature (ST) measured in degree Celsius (°C) was recorded using Infrared Thermometer with Laser Sighting (Mini-Temp Comark: MTSFU). Atmospheric CO<sub>2</sub> concentrations were measured at 20–30 cm above the soil surface using the infrared gas analyzer (IRGA) apparatus (Multi-GAS Monitor PGM-54, RAE Systems Inc., Sunnyvale, CA94089). Readings were picked up at midday to represent the increasing peaks of soil microbial activities. It is equipped with a pump for aspirating 300 cm<sup>3</sup> of atmospheric air per minute and detecting the CO<sub>2</sub> with the Non-Dispersive Infrared (NDIR) sensor. The atmospheric CO<sub>2</sub> concentrations were expressed in mg L<sup>-1</sup> (Emran et al., 2012b).

The CO<sub>2</sub> fluxed from the soil surface was captured by soda-lime. The soda-lime absorption method depends on the chemical adsorption of CO<sub>2</sub> and H<sub>2</sub>O that are resulting from soil respiration processes as:



The soda-lime absorption method was applied in the field using the dark cover-box technique as described in detail by Emran (2012). Incubation of soda-lime lasted for 24 h. The detected CO<sub>2</sub> concentrations were calculated using the chemically bounded CO<sub>2</sub> with soda-lime after oven-drying, then multiplied by 1.69 as a correction factor, and presented in mg g<sup>-1</sup> soil. The soil carbon emitted as C-CO<sub>2</sub> was calculated (CO<sub>2</sub> readings multiplied by 12/44: M.W. of C/CO<sub>2</sub>) and then expressed in mg g<sup>-1</sup> (Emran et al., 2012b).

The total GRSP (GRSP<sub>T</sub>) content was extracted for each soil by 50 mM trisodium citrate (pH 8.0) at 121 °C in the autoclave for 60 min. Extraction was repeated until the supernatant had a pale-yellow color announcing the absence of GRSP. The easily extractable GRSP (GRSP<sub>EE</sub>) fraction was obtained by extracting each soil with 20 mM trisodium citrate (pH 7.0) at 121 °C during one autoclave cycle of 30 min. Supernatants were centrifuged at 2,000xg for 10 min and stored at 4 °C. The GRSP fractions (mg g<sup>-1</sup>) were quantified by the Bradford method (Wright and Upadhyaya, 1996). The carbon contained in the GRSP (C-GRSP) was purified, as detailed in Emran et al. (2020), and quantified using TOC (Total Organic Carbon) Analyzer (Torch Combustion TOC Analyzer, Teledyne Tekmar, Ohio, USA), and then displayed in mg g<sup>-1</sup>.

The surface microstructure of undisturbed soil aggregates was investigated by using the Scanning Electron Microscope (SEM) (JSM-636 OLA, Jeol, Japan).



Fig. 1. Study area and sampling locations.

The structural stability of soil aggregates (WSA) was measured using the Eijkelkamp wet-sieving apparatus (Kemper and Rosenau, 1986). The air-dried aggregates were physically separated with 2.00 mm and 0.25 mm meshes to obtain the aggregates fractions of 2.00–4.75 mm and 0.25–2.00 mm, respectively. Separated aggregates were subjected to mechanical vertical immersion-emersion cycles in deionized water ( $34 \text{ cycle min}^{-1}$ ) for three minutes. Time was pre-calibrated to avoid the total disruption of the fragile soil aggregates. Survived soil aggregates were weighed after oven-dried at  $105 \text{ }^\circ\text{C}$ . The WSA was calculated as:

$$WSA (\%) = \frac{M_A - S_A}{M_T - S_T} \times 100 \quad (3)$$

where  $M_A$  is the mass of the survived soil aggregates after oven-dried (g) and  $S_A$  represents its calculated content of sand fraction (g). The  $M_T$  is the total mass of the soil sample before water dispersion (g) and  $S_T$  represents its calculated content of sand fraction. The stability index of soil aggregates was defined as the ratio of large-sized aggregates to the small-sized aggregates and calculated as:

$$\text{Stability Index} = \frac{WSA_{2.00-4.75 \text{ mm}}}{WSA_{0.25-2.00 \text{ mm}}} \quad (4)$$

where  $WSA_{2.00-4.75 \text{ mm}}$  and  $WSA_{0.25-2.00 \text{ mm}}$  are the soil water-stable aggregates at the two studied aggregates fractions.

### 2.3. Statistical analysis

Analysis of Variance (ANOVA) and Factor Analysis were run using STATISTICA 10 of StatSoft, Inc. (Tulsa, Oklahoma, USA)

(StatSoft Inc. 2011). The two-factor ANOVA tested to analyze all soil variables with respect to the spatial distribution of soil environments and seasonal changes as the two categorical factors. The two-factor ANOVA tested to analyze the interaction between the two categorical factors to check the significant variance of all obtained data. Using STATISTICA 10, factor analysis was run using all the studied parameters to express the most effective variance among them in the first three factor structures. Factor scores for each factor structure were also checked to display the contribution of each treatment (studied soil sites) to the related factor structure (Emran et al., 2020). In addition, linear regression equations were checked between the two fractions of glomalin-related soil protein ( $GRSP_T$  and  $GRSP_{EE}$ ) and soil nutrients for each soil to display the dynamic changes of GRSP production along the seasons in the function of soil nutrients contents (C, N, P).

## 3. Results

### 3.1. Soil characteristics

Soil particle analysis showed sandy loam texture in all soils except S1 which had a loamy sand texture. The minor clay contents in all soils suggested the importance of the stabilized SOM structures to compensate for the minor clay content. Soils in S4, S6, and S7 showed better soil structure than other soils due to the high values recorded for SOC, TN, TP, C:N, and SM and lower values of BD (Table 1). Among all soils, the variations in BD, SOC, and TN contents may be attributed to agricultural management practices, age

of abandonment, type of vegetation, and wildfire occurrence (Table 1). The low severity fire in S6 may stimulate the fast recovering time of vegetation after fire towards a natural vegetation succession of *C. monspeliensis* scrub (Pardini et al., 2004; 2017). The TP values were higher in S2, S3, and S7 soils than in S1, S4, S5, and S6 soils probably due to dung deposition through the periodic passage of cattle flocks. (Table 1).

### 3.2. Stoichiometric of soil C:N:P, the “Redfield ratio”

The molar ratios among soil C, N, and P can be seen in Table 2. It can be assumed that S1-S3 soils may be classified as N and P limitations (Table 2). In general, the S4-S6 soils presented higher C:P ratios than 106 indicating low P content thus imbalance of nutrients dynamics can occur.

It can be also observed in Table 3 that all the studied soil parameters showed highly significant seasonal variability as checked by the two-factor ANOVA using SS Whole Model vs. SS Residual.

### 3.3. SEM observations and structural stability of aggregates

The selected air-dried and naturally fractured surface (0–20 cm) aggregates were examined under SEM to observe the structural micro-arrangement in the studied soils. The loamy sand texture marked the microstructure at S1 soil under frequently tilled vines which minerals compounds ( $\varnothing 90 \mu\text{m}$ ) and preferential interparticle porosity (Fig. 2a) indicating good drainage but low structural stability of aggregates. The soil in S2 under the olive grove periodically tilled showed different aggregation features (Fig. 2b) with a crumb-like microstructure and crossed by fungi filaments and root hairs. A similar trend in microstructure arrangement was observed in S3 soil under pine trees (Fig. 2c) though with dense particle pack-

ing, probably due to repeated wildfire occurrence affecting organic compounds of soil surface. By contrast, a well-structured microarray was observed in S5 soil under cork trees preserved by fire for a long time (Fig. 2d). That soil structure may improve more in soil under corks than in pines has been reported (Sardans, et al., 2006; Emran et al., 2012a). Cerdà, et al. (2017) inferred that the organic matter developed under pines was hardly mineralized with less incorporation of organic compounds in comparison with cork residues. This may be checked in the micrographs of S3 (Fig. 2c) and S4 (Fig. 2d and e) where an assemblage of organic and mineral components seems to form a feature at stable soil aggregation.

The micrographs of S5 soil (Fig. 2f) under *C. monspeliensis* scrub, more often affected by fire for its content of monoterpenes and sesquiterpenes compounds (Rivoal et al., 2010), showed some pictures of residual washed eroded material formed by burnt roots with the topped sediments. A different shape was offered by the microstructure of soil S6 under *Erica arborea* scrub, characterized by ancient abandonment out less affected by the fire. A homogeneous arrangement of organic (hair roots, fungi hyphae, and mycorrhizas) and mineral components (Fig. 2g and h) indicated favorable structural conditions even suggested by lower BD values along seasons. Surprisingly, soil S5 showed a lower yearly ratio C-CO<sub>2</sub>/C-GRSP (0.06) for soil S6 (0.09) probably suggesting a faster-growing of spontaneous vegetation renewing the plant mantle with consequent increase of fungal activity and associated glomalin formation (Reyes et al., 2019). The SEM micrograph of S7 soil under natural meadows showed a rather compacted microstructure (Fig. 2i and j) due to winter grazing (mainly cattle). However, this soil showed the lowest carbon loss (C-CO<sub>2</sub>) and the highest glomalin carbon (C-GRSP) on an early basis, with C-CO<sub>2</sub>/C-GRSP ratio decreasing by 96%, 73%, 75%, 57%, 50%, and 67% for S1, S2, S3, S4, S5, and S6 soils.

**Table 1**  
Seasonal means ( $\pm$ standard error) of the selected soil parameters.

Soil	Coordinates	Season	BD kg m <sup>-3</sup>	SM %	ST °C	SOC g kg <sup>-1</sup>	TN g kg <sup>-1</sup>	TP g kg <sup>-1</sup>
S1	42°19'9.57"N, 3°12'22.61"E	Wi	1.47 ± 0.07	7.38 ± 2.20	13.30 ± 0.41	2.16 ± 0.00	0.92 ± 0.00	0.55 ± 0.03
		Sp	1.56 ± 0.06	3.43 ± 0.97	15.83 ± 2.71	2.59 ± 0.04	0.54 ± 0.10	0.46 ± 0.01
		Su	1.59 ± 0.01	0.91 ± 0.21	40.12 ± 1.17	2.63 ± 0.41	0.77 ± 0.13	0.51 ± 0.01
		Au	2.08 ± 0.71	2.39 ± 2.66	30.50 ± 3.11	2.95 ± 1.32	0.90 ± 0.07	0.45 ± 0.02
S2	42°18'52.74"N, 3°13'13.79"E	Wi	1.34 ± 0.09	13.81 ± 4.00	22.61 ± 2.38	13.24 ± 0.00	2.26 ± 0.16	0.82 ± 0.01
		Sp	1.36 ± 0.04	7.38 ± 3.36	21.15 ± 7.28	11.17 ± 1.24	1.79 ± 0.08	0.56 ± 0.03
		Su	1.38 ± 0.03	1.57 ± 0.35	39.02 ± 0.39	16.32 ± 4.04	1.85 ± 0.93	0.75 ± 0.01
		Au	1.34 ± 0.05	6.81 ± 7.97	32.15 ± 5.44	19.03 ± 4.33	3.08 ± 0.73	0.69 ± 0.00
S3	42°18'51.40"N, 3°12'51.02"E	Wi	1.11 ± 0.06	20.50 ± 3.31	17.66 ± 3.60	25.29 ± 0.00	2.05 ± 0.16	1.06 ± 0.03
		Sp	1.21 ± 0.18	8.09 ± 3.53	16.20 ± 1.91	13.94 ± 3.50	2.31 ± 1.23	0.77 ± 0.01
		Su	1.24 ± 0.14	2.71 ± 1.38	33.52 ± 3.50	15.88 ± 7.55	2.24 ± 0.48	0.99 ± 0.02
		Au	1.18 ± 0.11	7.13 ± 7.85	26.65 ± 10.11	14.33 ± 2.53	2.32 ± 0.51	0.87 ± 0.00
S4	42°18'24.93"N, 3°15'12.05"	Wi	1.11 ± 0.07	19.24 ± 3.91	18.95 ± 4.60	31.08 ± 0.00	2.44 ± 0.02	0.85 ± 0.02
		Sp	1.12 ± 0.09	10.61 ± 4.45	17.85 ± 1.98	24.08 ± 0.71	2.09 ± 0.14	0.44 ± 0.00
		Su	1.24 ± 0.14	3.38 ± 1.45	34.90 ± 3.11	25.83 ± 7.97	2.75 ± 0.14	0.54 ± 0.01
		Au	1.09 ± 0.13	8.72 ± 9.30	29.40 ± 14.00	29.93 ± 7.07	2.83 ± 0.18	0.48 ± 0.02
S5	42°17'42.81"N, 3°13'12.08"E	Wi	1.11 ± 0.07	21.96 ± 2.97	11.25 ± 4.08	37.75 ± 0.00	3.80 ± 0.16	0.62 ± 0.02
		Sp	1.18 ± 0.13	14.35 ± 7.66	12.90 ± 1.46	25.38 ± 2.76	2.76 ± 0.14	0.40 ± 0.01
		Su	1.17 ± 0.12	3.78 ± 1.24	33.25 ± 11.67	28.46 ± 3.41	3.00 ± 0.87	0.43 ± 0.01
		Au	1.08 ± 0.12	12.31 ± 13.89	20.05 ± 8.56	28.06 ± 2.96	3.31 ± 0.48	0.46 ± 0.01
S6	42°17'2.51"N, 3°12'18.00"E	Wi	0.99 ± 0.10	22.54 ± 2.76	11.25 ± 4.23	43.53 ± 0.00	4.52 ± 0.16	0.62 ± 0.05
		Sp	0.97 ± 0.17	18.15 ± 9.57	13.81 ± 0.94	26.55 ± 4.03	2.93 ± 0.99	0.41 ± 0.02
		Su	0.99 ± 0.08	4.60 ± 1.58	31.87 ± 7.39	35.27 ± 1.03	3.45 ± 0.40	0.46 ± 0.01
		Au	0.83 ± 0.02	13.11 ± 13.25	19.22 ± 10.50	37.76 ± 5.70	4.00 ± 0.37	0.48 ± 0.02
S7	42°16'16.23"N, 3°15'3.31"E	Wi	0.88 ± 0.19	45.09 ± 16.39	10.33 ± 6.02	38.14 ± 0.00	5.13 ± 0.16	1.19 ± 0.02
		Sp	1.21 ± 0.12	14.89 ± 10.44	12.53 ± 1.68	32.58 ± 1.14	4.53 ± 1.07	0.74 ± 0.02
		Su	1.20 ± 0.05	2.54 ± 0.30	34.62 ± 3.50	35.93 ± 2.38	4.80 ± 0.40	0.87 ± 0.01
		Au	1.06 ± 0.03	11.01 ± 12.12	18.12 ± 9.72	38.81 ± 13.09	4.76 ± 0.55	0.82 ± 0.01

BD: bulk density, SM: soil moisture, ST: soil temperature, SOC: soil organic carbon, TN: total nitrogen, TP: total phosphorus, S1: soil of *Vitis vinifera* L., S2: soil of *Olea europaea* L., S3: soil of *Pinus halepensis* Mill. trees (S3) and *Quercus suber* L. trees, S5: soil of *Cistus monspeliensis* L. scrub, S6: soil of *Erica arborea* L. scrub, S7: soil of *Brachypodium retusum* Pers. Pasture, Wi: winter, Sp: spring, Su: summer, Au: autumn.

**Table 2**  
Seasonal means (±standard error) of molar C:N:P ratios with CO<sub>2</sub>, C-CO<sub>2</sub>, GRSP<sub>T</sub>, and C-GRSP measurements.

Soil	Season	C:N	Molar C:P	Molar N:P	CO <sub>2</sub> mg g <sup>-1</sup>	C-CO <sub>2</sub> mg g <sup>-1</sup>	GRSP mg g <sup>-1</sup>	C-GRSP mg g <sup>-1</sup>
S1	Wi	2.57	10	3	0.26 ± 0.04	0.07	1.30 ± 0.25	0.15
	Sp	5.09	14	2	0.47 ± 0.04	0.13	1.39 ± 0.48	0.14
	Su	3.90	14	3	0.67 ± 0.05	0.18	2.14 ± 0.66	0.36
	Au	3.21	15	4	0.50 ± 0.07	0.14	0.84 ± 0.10	0.07
S2	Wi	6.45	42	6	0.36 ± 0.11	0.10	1.27 ± 0.15	0.16
	Sp	7.71	58	6	0.66 ± 0.03	0.18	3.03 ± 0.96	0.71
	Su	10.28	59	5	0.92 ± 0.01	0.25	8.76 ± 3.34	5.34
	Au	6.43	67	9	0.99 ± 0.14	0.27	2.93 ± 0.17	0.91
S3	Wi	13.55	62	4	0.55 ± 0.07	0.15	1.34 ± 0.47	0.13
	Sp	6.08	43	6	0.55 ± 0.09	0.15	3.18 ± 1.14	0.73
	Su	8.68	46	5	0.69 ± 0.03	0.19	7.47 ± 3.00	3.75
	Au	7.08	44	5	0.64 ± 0.11	0.17	2.50 ± 0.09	0.68
S4	Wi	14.03	94	6	0.44 ± 0.03	0.12	2.12 ± 0.12	0.50
	Sp	13.35	148	10	0.74 ± 0.04	0.20	4.43 ± 1.06	1.83
	Su	11.08	132	10	0.73 ± 0.02	0.20	8.46 ± 3.00	5.80
	Au	10.97	152	12	0.65 ± 0.03	0.18	3.24 ± 0.12	1.24
S5	Wi	10.93	156	12	0.43 ± 0.08	0.12	2.30 ± 0.10	0.60
	Sp	9.97	162	14	0.62 ± 0.03	0.17	4.42 ± 0.92	1.90
	Su	10.74	176	14	0.69 ± 0.06	0.19	8.85 ± 3.03	6.49
	Au	9.10	152	14	0.67 ± 0.16	0.18	3.29 ± 0.12	1.27
S6	Wi	10.60	181	15	0.48 ± 0.05	0.13	2.43 ± 0.25	0.63
	Sp	12.21	205	14	0.71 ± 0.19	0.19	6.51 ± 2.03	3.64
	Su	11.32	199	15	0.98 ± 0.14	0.27	8.03 ± 2.82	5.25
	Au	10.74	212	17	0.85 ± 0.02	0.23	3.79 ± 0.15	1.70
S7	Wi	8.17	83	9	0.55 ± 0.08	0.15	3.58 ± 0.56	1.40
	Sp	8.41	120	12	0.45 ± 0.05	0.12	6.17 ± 1.56	3.48
	Su	8.35	108	11	0.50 ± 0.04	0.14	11.35 ± 3.85	9.93
	Au	8.25	113	12	0.55 ± 0.05	0.15	4.31 ± 0.05	2.26

C:N: carbon/nitrogen ratio, Molar C:P: molar carbon/phosphorus ratio, Molar N:P: molar nitrogen/phosphorus ratio, CO<sub>2</sub>: carbon dioxide emission, C-CO<sub>2</sub>: soil carbon emitted as CO<sub>2</sub>, GRSP: glomalin-related soil protein, C-GRSP: carbon content of GRSP, S1: soil of *Vitis vinifera* L., S2: soil of *Olea europaea* L., S3: soil of *Pinus halepensis* Mill. trees (S3) and *Quercus suber* L. trees, S5: soil of *Cistus monspeliensis* L. scrub, S6: soil of *Erica arborea* L. scrub, S7: soil of *Brachypodium retusum* Pers. Pasture, Wi: winter, Sp: spring, Su: summer, Au: autumn.

**Table 3**  
ANOVA using the sum of squares (SS) of the whole Model vs. SS of the Residual.

Variables	SS model	df model	MS model	SS residual	df residual	MS residual	F-ratio	P-level
BD	1.37	9	0.15	0.29	18	0.02	9.30	0.0000
SM	1605.46	9	178.38	297.14	18	16.51	10.81	0.0000
ST	2256.06	9	250.67	104.50	18	5.81	43.18	0.0000
SOC	3918.27	9	435.36	191.99	18	10.67	40.82	0.0000
TN	41.10	9	4.57	1.97	18	0.11	41.75	0.0000
TP	1.20	9	0.13	0.08	18	0.00	31.08	0.0000
C:N	148.39	9	16.49	38.80	18	2.16	7.65	0.0001
Molar C:N	201.98	9	22.44	52.81	18	2.93	7.65	0.0001
Molar N:P	615.69	9	68.41	25.82	18	1.43	47.69	0.0000
Molar C:P	100085.44	9	11120.60	3466.41	18	192.58	57.75	0.0000
Atmospheric CO <sub>2</sub> AIR	203737.32	9	22637.48	18402.88	18	1022.38	22.14	0.0000
Soil CO <sub>2</sub> emission	1.44	9	0.16	0.56	18	0.03	5.17	0.0015
C-CO <sub>2</sub>	0.11	9	0.01	0.04	18	0.00	5.17	0.0015
GRSP <sub>EE</sub>	4.49	9	0.50	2.00	18	0.11	4.49	0.0033
GRSP <sub>T</sub>	193.19	9	21.47	22.07	18	1.23	17.51	0.0000
C-GRSP	134.19	9	14.91	26.18	18	1.45	10.25	0.0000
C-CO <sub>2</sub> /SOC	49.44	9	5.49	2.12	18	0.12	46.59	0.0000
C-GRSP/SOC	1798.79	9	199.87	296.36	18	16.46	12.14	0.0000
Aggregates <sub>0.25-2.00 mm</sub>	2478.25	9	275.36	865.31	18	48.07	5.73	0.0008
WSA <sub>0.25-2.00 mm</sub>	9426.37	9	1047.37	5015.31	18	278.63	3.76	0.0081
Aggregates <sub>2.00-4.75 mm</sub>	2926.59	9	325.18	136.77	18	7.60	42.80	0.0000
WSA <sub>2.00-4.75 mm</sub>	8408.99	9	934.33	2176.01	18	120.89	7.73	0.0001
Stability index	0.16	9	0.02	0.04	18	0.00	8.80	0.0001

SS: sum of squares, df: degree of freedom, MS: mean squares, BD: bulk density, SM: soil moisture, ST: soil temperature, SOC: soil organic carbon, TN: total nitrogen, TP: total phosphorus, C:N: carbon/nitrogen ratio, Molar C:P: molar carbon/phosphorus ratio, Molar N:P: molar nitrogen/phosphorus ratio, CO<sub>2</sub>: carbon dioxide emission, C-CO<sub>2</sub>: soil carbon emitted as CO<sub>2</sub>, GRSP<sub>EE</sub>: easily extractable glomalin-related soil protein, GRSP<sub>T</sub>: total glomalin-related soil protein, C-GRSP: carbon content of GRSP, WSA: water-stable aggregates.

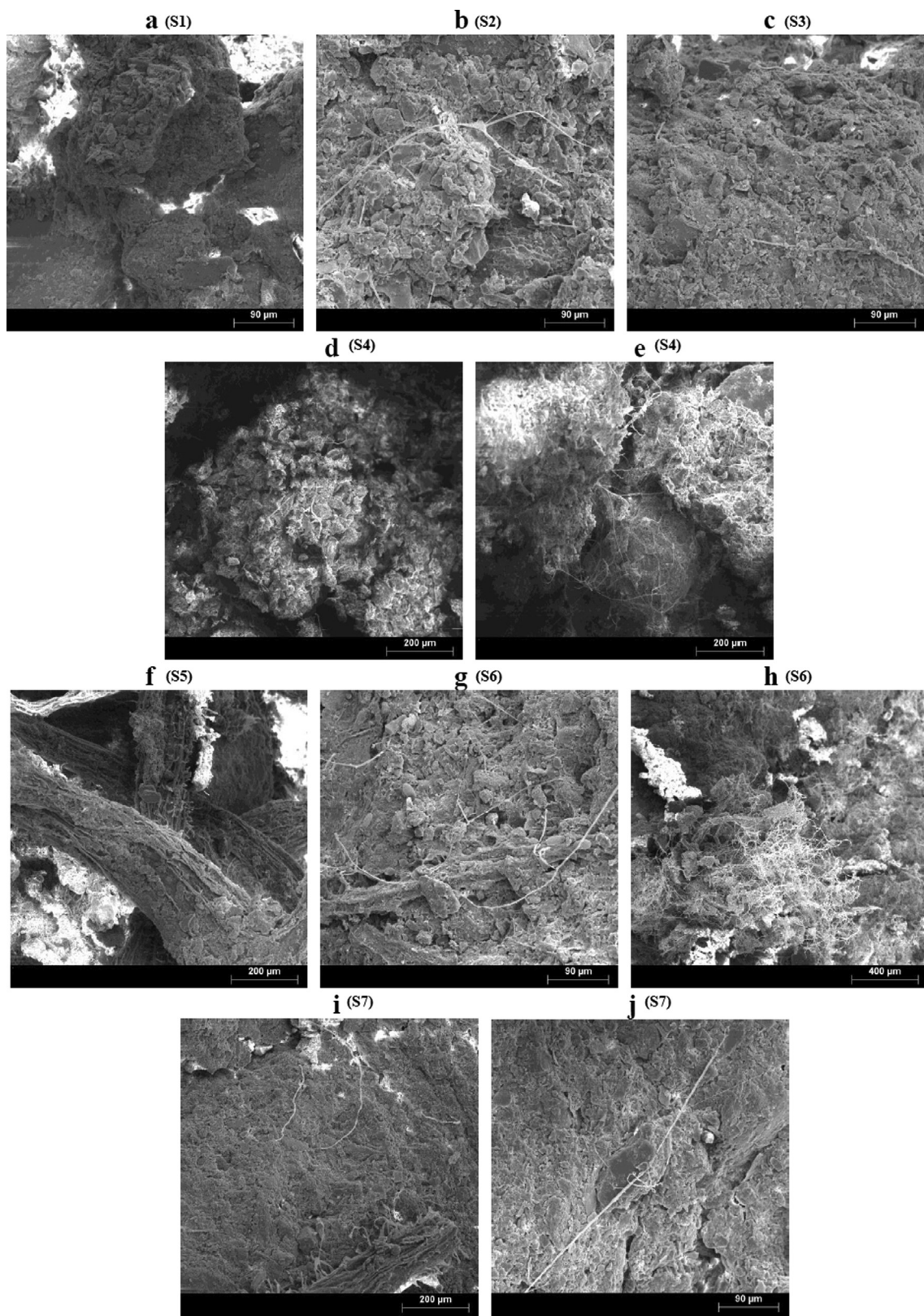
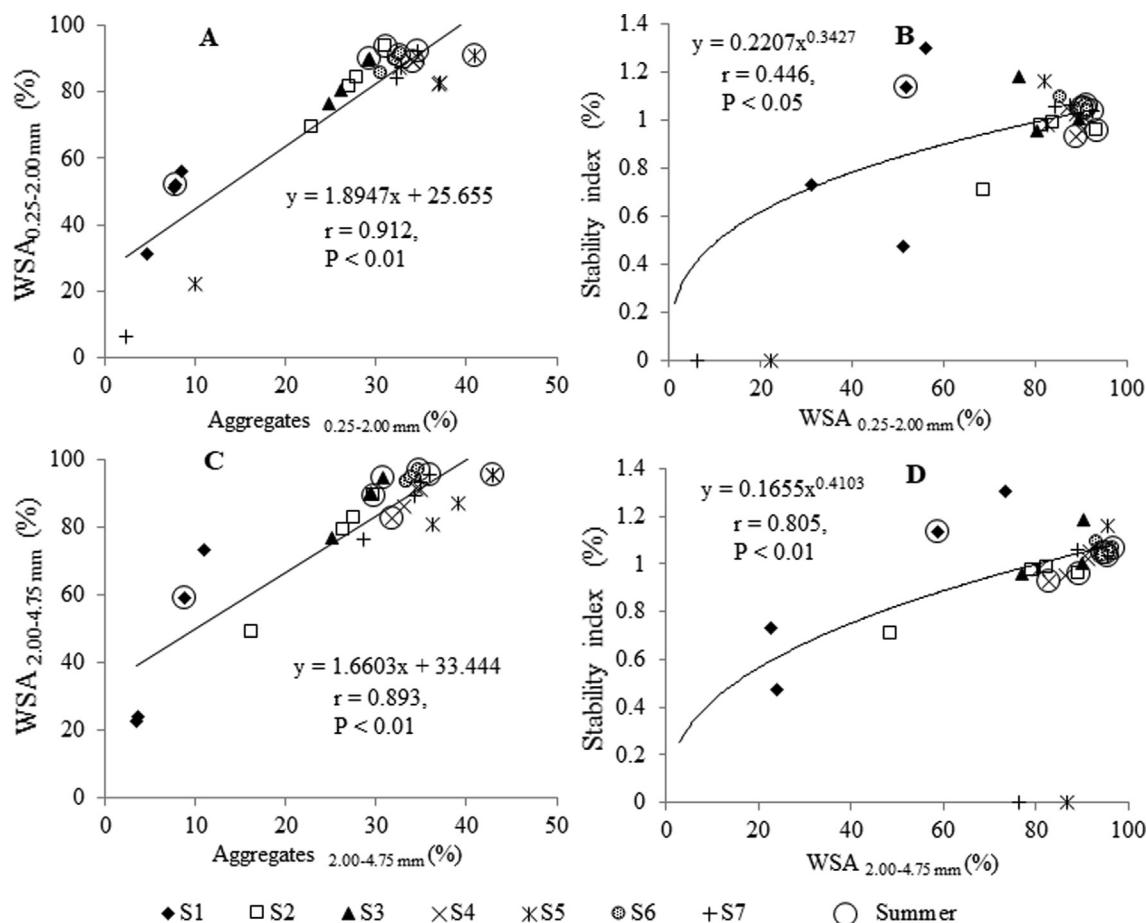


Fig. 2. SEM micrographs of soils aggregates features.

Linear correlations between the stability of soil aggregates and the contents of formed aggregates can be seen in Fig. 3A and C. The stability index potentially increased (power equation) with the increasing stability of soil aggregates (Fig. 3B and D)

3.4. Glomalin-related soil protein (GRSP<sub>T</sub> and GRSP<sub>EE</sub>) and soil nutrients

Significant data variability was checked by the two-factor ANOVA (Table 3). Among the studied soils GRSP<sub>EE</sub> and GRSP<sub>T</sub> varied



**Fig. 3.** Linear correlations between water-stability of aggregates (WSA) and aggregate contents (%) at the 0.25–2.00 mm (A) and 2.00–4.75 mm (C) with the power function of the stability index versus the WSA of 0.25–2.00 mm (B) and 2.00–4.75 mm (D) aggregate fractions. Summer measurements are marked with a circle.

significantly ( $P < 0.01$ ) and emphasized the impact of land use and abandonment on GRSP production. Moreover, the  $GRSP_T$  fractions varied significantly among the studied seasons in each soil ( $P < 0.001$ ), indicating its susceptibility to climatic conditions. By contrast,  $GRSP_{EE}$  did not show significant variability among the studied seasons ( $P > 0.1$ ) though resulted positively correlated with soil moisture ( $r = 0.455$ ,  $P < 0.05$ ). These findings may suggest that  $GRSP_{EE}$  production is more dependent on soil conditions rather than seasonal changes. The  $GRSP_T$  showed higher values in all soils than the  $GRSP_{EE}$  along the studied seasons supporting the assumption that  $GRSP_{EE}$  is mostly transformed into the stable  $GRSP_T$  fraction and this seems to occur substantially in summer (Fig. 4). Furthermore, the same figure indicates that  $GRSP_T$  production increases from agricultural soils to the soil in ancient abandonment and is less affected by the fire than others with intensive agricultural management practices.

The  $GRSP_{EE}$  resulted positively correlated with SOC ( $r = 0.523$ ,  $P < 0.01$ ,  $n = 28$ ) depicting that its production was mainly dependent on soil carbon content (Fig. 5). This fraction was highly transformed to the stabilized GRSP forms, mostly in the summer season as indicated by the high  $GRSP_T$  values. This mechanism was also found in soils with ancient abandonment and protected from fire.

Total nitrogen values increased significantly with  $GRSP_{EE}$  ( $r = 0.508$ ,  $P < 0.01$ ) and  $GRSP_T$  ( $r = 0.381$ ,  $P < 0.05$ ) along the studied soils from S1 to S7 showing the highest  $GRSP_T$  values in summer. The highest increase of GRSP in soils may suggest that the assimilated N may be utilized during the transformation processes of GRSP in soil. However, the  $GRSP_T$  showed negatively correlated

with TN for each soil in S5 ( $r = -0.999$ ,  $P < 0.01$ ), S6 ( $r = -0.999$ ,  $P < 0.05$ ), and S7 ( $r = -0.931$ ,  $P < 0.01$ ) (Fig. 6).

The significant positive correlation observed when plotting the seasonal means of the freshly produced glomalin fraction ( $GRSP_{EE}$ ) against the total phosphorus (TP) contents ( $r = 0.393$ ,  $P < 0.05$ ) may suggest the relevance of soil phosphorus uptake by the plant. No significant correlation was found between  $GRSP_T$  and TP indicating the unclear trend of soil phosphorus in the transformation processes of the recalcitrant GRSP molecules. Thus, the freshly produced glomalin molecules seemed to be highly correlated with the mycorrhizal metabolic activities stimulated by phosphorus dynamics in the rhizosphere zone and probably the behavior of the mycorrhizal associations in each soil may differ as a result of the plant habitats. In each soil, the  $GRSP_T$  negatively correlated with TP for S2–S7 soils (Fig. 7). The TP values increased from winter to spring and autumn, and after excluding summer values, the following regressions were obtained: S2 ( $r = -0.876$ ,  $P < 0.01$ ), S3 ( $r = -0.999$ ,  $P < 0.01$ ), S4 ( $r = -0.897$ ,  $P < 0.01$ ), S5 ( $r = -0.962$ ,  $P < 0.01$ ), S6 ( $r = -0.921$ ,  $P < 0.01$ ), and S7 ( $r = -0.817$ ,  $P < 0.01$ ) soils (Fig. 7). This may indicate that glomalin was highly produced as a result of stimulating mycorrhizal species by root plants to supply the required phosphorus from soils to plants.

### 3.5. Statistical variance and factor analysis

Factor analysis was run using all the studied soil parameters. The first three factors explained 78% of the total variance (Table 4). The 1st factor explained 42% of the total variance with high positive loadings on SOC, TN, C:N, formation and stability of soil aggre-

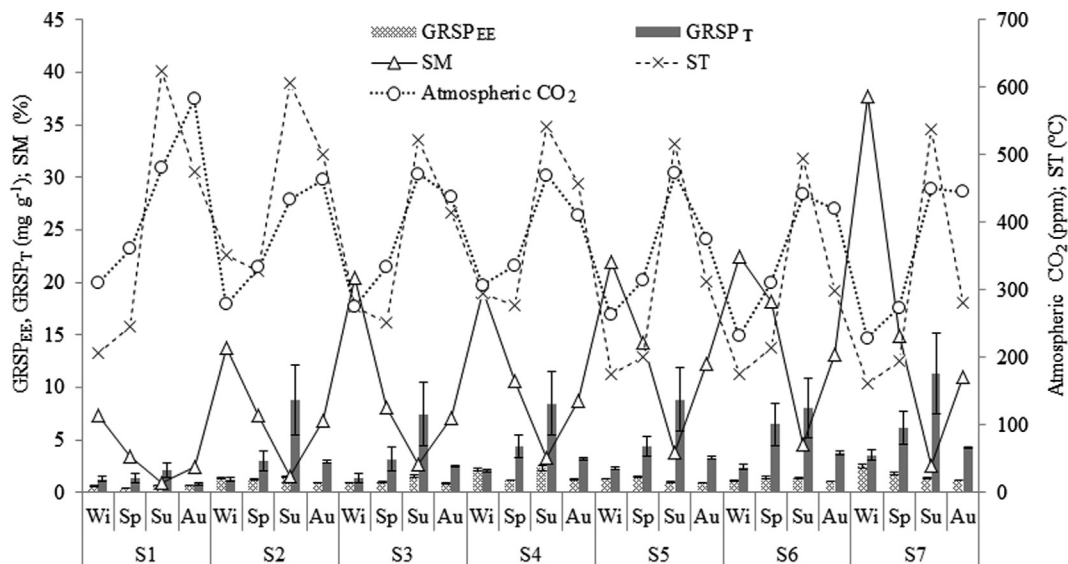


Fig. 4. Seasonal means of atmospheric CO<sub>2</sub> concentrations, soil moisture SM, soil temperature ST, easily extractable (GRSP<sub>EE</sub>) and total (GRSP<sub>T</sub>) glomalin-related soil protein fractions for the studied soils (S1-S7) in winter (Wi), spring (Sp), summer (Su), and autumn (Au).

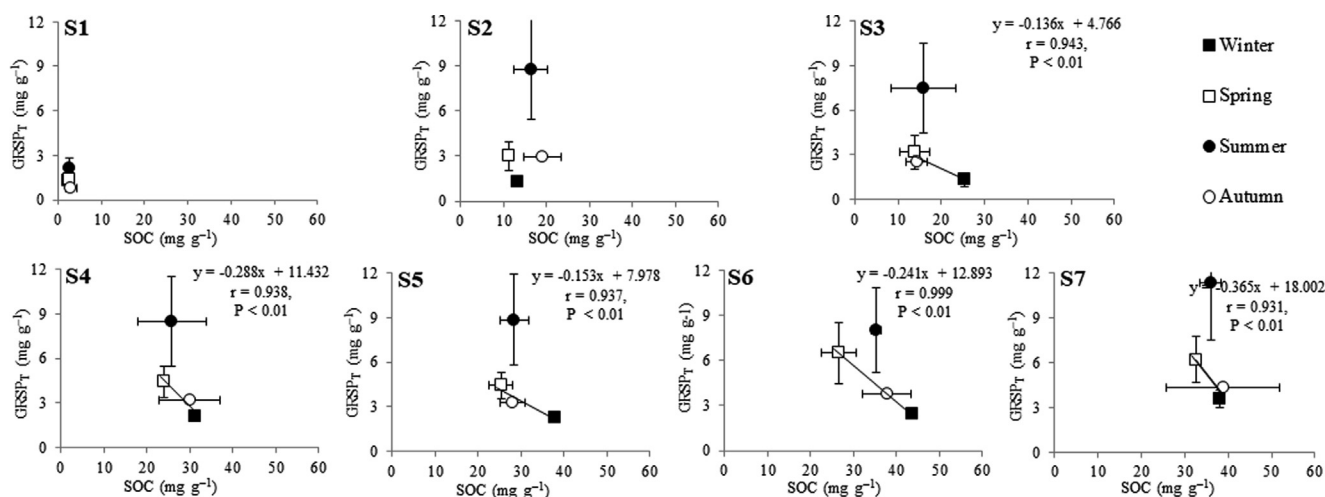


Fig. 5. Linear correlations between the seasonal means of total glomalin-related soil protein (GRSP<sub>T</sub>) and soil organic carbon (SOC) for the studied soils (S1-S7).

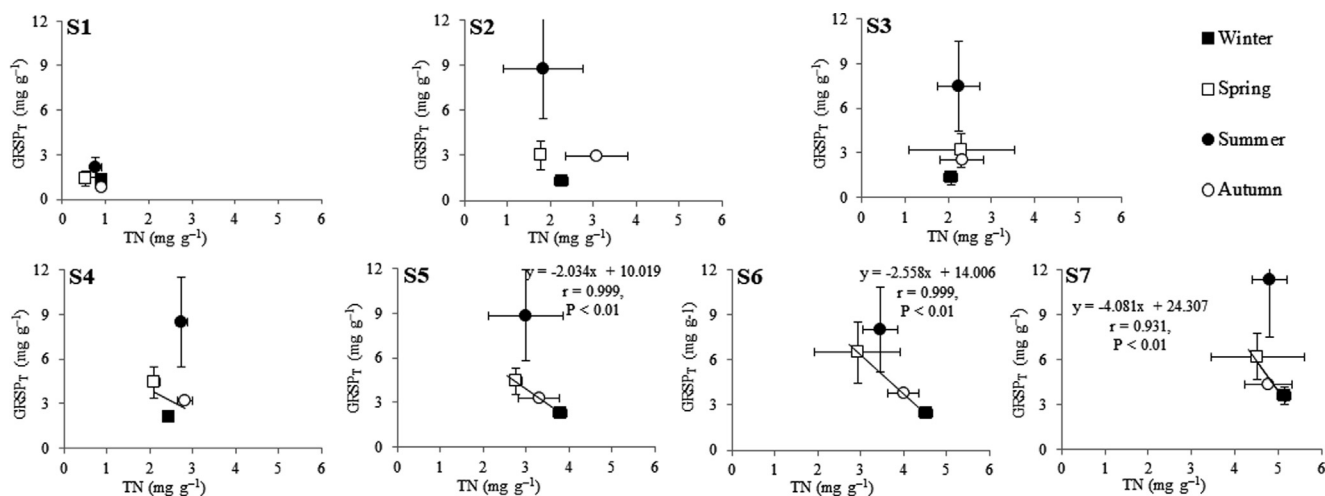


Fig. 6. Linear correlations between the seasonal means of total glomalin-related soil protein (GRSP) and total nitrogen (TN) for the studied soils (S1-S7).



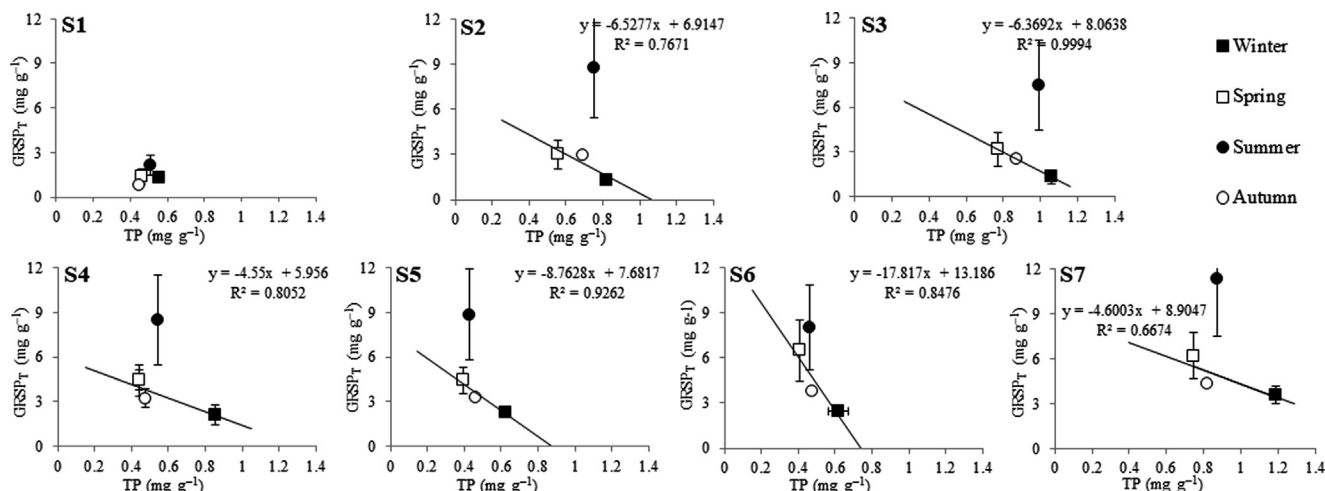


Fig. 7. Linear correlations between the seasonal means of total glomalin-related soil protein (GRSP) and total phosphorus (TP) for the studied soils (S1-S7).

gates, and Redfield ratios while negative loadings on bulk density and soil carbon loss as a proportion of soil carbon. This picture may indicate that appropriate soil structure and organic compounds as in S5-S7 soil may reduce soil carbon loss by increasing soil carbon storage capacity. Despite the lower percentage of explained variance in factor 2 (22%), negative loads were related to GRSP dynamics to contrast the CO<sub>2</sub> dynamics with positive loads as a result of the influences of soil and environmental conditions

on soil carbon loss and storage. The 3rd factor explained 14% of the total variance with a high positive contribution from soil moisture, total phosphorus, labile glomalin fraction, and stability index emphasizing the contribution of GRSP in the stability of soil aggregates.

Factor scores were obtained to assess the contribution of each variable (soil or/and season) to each factor structure. The more the factor is positive the higher contribution of this soil to each fac-

Table 4  
First three-factor structures and their factor scores of Factor analysis.

Soil variables	Factor structures				Factor scores		
	Factor 1	Factor 2	Factor 3		Factor 1	Factor 2	Factor 3
BD	-0.83	-0.18	-0.29	S1wi	-2.19	0.78	-0.09
SM	0.37	0.67	0.60	S1sp	-1.92	0.61	-0.70
ST	-0.18	-0.78	-0.22	S1su	-1.92	-0.24	-0.55
CO <sub>2</sub> AIR	-0.20	-0.61	-0.42	S1au	-1.88	0.15	-1.01
CO <sub>2</sub> emission	0.52	0.63	0.16	S2wi	-0.92	0.44	0.30
C-CO <sub>2</sub>	0.52	0.63	0.16	S2sp	-0.54	0.05	-0.35
SOC	0.89	0.08	0.32	S2su	-0.32	-2.07	0.41
TN	0.73	0.02	0.46	S2au	-0.23	-0.05	-0.44
TP	-0.07	-0.05	0.75	S3wi	0.12	0.91	0.25
C:N	0.83	0.02	0.04	S3sp	-0.46	0.15	0.00
GRSP <sub>EE</sub>	0.43	-0.21	0.67	S3su	-0.40	-1.57	0.64
GRSP <sub>T</sub>	0.42	-0.84	0.15	S3au	-0.31	0.11	-0.30
C-GRSP	0.37	-0.83	0.15	S4wi	0.54	0.40	0.38
C-CO <sub>2</sub> /SOC %	-0.84	0.18	-0.27	S4sp	0.64	0.27	-0.72
C-GRSP/SOC %	0.05	-0.92	0.08	S4su	0.28	-1.68	0.32
WSA <sub>0.25-2.00 mm</sub>	0.65	-0.49	-0.50	S4au	0.86	0.39	-0.89
Aggregates <sub>0.25-2.00 mm</sub>	0.76	-0.40	-0.34	S5wi	0.56	1.47	0.84
WSA <sub>2.00-4.75 mm</sub>	0.81	-0.29	0.02	S5sp	0.77	0.29	-0.56
Aggregates <sub>2.00-4.75 mm</sub>	0.92	-0.18	0.09	S5su	0.82	-1.35	-0.65
Stability index	0.03	0.29	0.84	S5au	0.83	0.79	-0.91
Molar N:P	0.86	0.05	-0.11	S6wi	1.06	1.08	-0.22
Molar C:P	0.92	0.07	-0.14	S6sp	0.98	0.42	-0.29
Molar C:N	0.83	0.02	0.04	S6su	0.92	-0.96	-0.42
Variance %	42	22	14	S6au	1.43	1.00	-0.93
Total variance %	42	64	78	S7wi	-0.12	1.18	4.21
				S7sp	0.40	-0.26	0.70
				S7su	0.38	-2.34	0.92
				S7au	0.64	0.02	0.07

BD: bulk density, SM: soil moisture, ST: soil temperature, SOC: soil organic carbon, TN: total nitrogen, TP: total phosphorus, C:N: carbon/nitrogen ratio, Molar C:P: molar carbon/phosphorus ratio, Molar N:P: molar nitrogen/phosphorus ratio, CO<sub>2</sub>: carbon dioxide emission, C-CO<sub>2</sub>: soil carbon emitted as CO<sub>2</sub>, GRSP<sub>EE</sub>: easily extractable glomalin-related soil protein, GRSP<sub>T</sub>: total glomalin-related soil protein, C-GRSP: carbon content of GRSP, WSA: water-stable aggregates.

S1: soil of *Vitis vinifera* L., S2: soil of *Olea europaea* L., S3: soil of *Pinus halepensis* Mill. trees (S3) and *Quercus suber* L. trees, S5: soil of *Cistus monspeliensis* L. scrub, S6: soil of *Erica arborea* L. scrub, S7: soil of *Brachypodium retusum* Pers. Pasture, Wi: winter, Sp: spring, Su: summer, Au: autumn. S1: soil of *Vitis vinifera* L., S2: soil of *Olea europaea* L., S3: soil of *Pinus halepensis* Mill. trees (S3) and *Quercus suber* L. trees, S5: soil of *Cistus monspeliensis* L. scrub, S6: soil of *Erica arborea* L. scrub, S7: soil of *Brachypodium retusum* Pers. Pasture, Wi: winter, Sp: spring, Su: summer, Au: autumn.

tor. In factor 1, soils in S4–S7 showed a high contribution in increasing soil aggregates stability due to the increasing capacity of soil nutrients and carbon storage except in winter in S7 (S7wi) due to the soil compaction resulted from grazing. The high negative factor scores were in S1 followed by S2–S3 indicating their high contribution to carbon loss because of the poor soil structure and stability caused by the low inputs of soil nutrients and organic compounds. Factor scores relevant to the 2nd factor showed negative values in summer that are in correlation to the negative loadings of soil variables in this factor from soil temperature, CO<sub>2AIR</sub>, GRSP, and C–GRSP/SOC. This is a very interesting point demonstrating the higher assimilation of CO<sub>2AIR</sub> in summer to increase the GRSP production and consequently the increase of carbon contained in GRSP (C–GRSP) as a proportion of SOC. Effectively, transformation processes of active organic compounds into stable/recalcitrant forms resulted coherently related to soil structure, nutrients stocks (mainly organic carbon pools), and climate conditions.

## 4. Discussion

### 4.1. GRSP and soil nutrients dynamics

The high seasonal fluctuations in soil moisture (SM) and soil temperature (ST) seemed to be fundamental in these very shallow soils indicating the importance of new organic matter formation and stabilization to achieve a better aggregation of soil particles into crumbs. For instance, soils with poor aggregation may be easily subjected to water dispersion or beating action of raindrops and with low colloidal clay content, the building mechanisms between stabilized organic matter and minerals are of great relevance for maintaining a stable structure. The highest SOC and TN values were found in S7 soil followed by S6, S5, and S4 soils, respectively. Hardly mineralizable pine needles in S3 soil under pine trees may reduce soil microbial activities by decreasing the decomposition rates of SOM (Gispert et al., 2013; 2018). The repeated wildfire that occurred in S5 may have temporarily altered SOM dynamics and soil structure at the upper soil horizons as soil BD, SOC, and TN were influenced (Table 1).

Fazhu et al. (2015) reported that land-use change included abandonment may affect the stoichiometry of C, N, and P in the soil. These three major elements generally occur in relatively stable ratios in living organisms as it was primarily reported by Redfield (1958) that plankton biomass showed a C:N:P ratio of 106:16:1. In recent decades, efforts have been made to adapt the Redfield ratio to terrestrial ecosystems due to natural or anthropogenic constraints despite the heterogeneity of these biogenic elements. For instance, Cleveland and Liptzin (2007) reported a relatively stable C:N:P ratio of 186:13:1 on global scale soils. Tian et al. (2010) showed a well-constrained C:N:P of 134:9:1 for soil microbial biomass whilst Aponte et al. (2010) found that vegetation and soil properties together with the interaction among season may affect C, N, and P biochemical cycles, thus their ratios. Also, the cycling of soil nutrients can be influenced by various soil characteristics relevant to soil fertility and structure as well as environmental factors more relevant to the rhizosphere (Guo and Jiang, 2019; Jiang and Guo, 2019). Reich and Oleksyn (2004) assumed an equilibrium ecosystem by exhibiting the N:P ratio at  $14 > N:P > 16$ . Accordingly, the breakpoints of molar N:P ratio can be calculated to classify those ecosystems with N limitation at  $N:P < 14$  and P limitation at  $N:P > 16$ , indicating that N:P ratios may provide a reliable indicator of nutrient constraints in terrestrial ecosystems. As a result, all soils showed nitrogen restrictions except for S5(Su–Au) and S6 (Wi–Su). A little increase of N:P ratio of 17 in S6 in autumn can be attributed to the leaf litter debris of *E. arborea*. The C:P ratio showed an overestimation of carbon in S4–S6 and the best-fitted

ratio was found in S7 under grazing. In general, the N and P contents are extremely underestimated in S1–S3 soils leading to catalog these soils not only with poorer structure but also with nitrogen and phosphorus restrictions. The imbalance of nutrients in S4–S7 soils should present better conditions with somewhat excess nutrients based on the seasonal plant cover changes (Table 1 and 2). It can be demonstrated that better land management is of paramount importance in those soils because the ongoing restrictions of soil nutrients can affect soil ecosystem functions (Campbell et al., 2013).

Glomalin was highly produced in soils rich in their carbon content and much more when photosynthetic processes increase as in the summer season (Palmqvist, 2002). In cultivated olive groves (S2), glomalin showed a steep increase in summer compared to vines (S1) probably due to the higher SOC content and photosynthate enrichment in S2 (Fig. 5). As stated above, the SOC pool may be a source for glomalin formation when low photosynthate production occurs. Accordingly, significant negative correlations were obtained by excluding summer measurements in S3–S7 soils (Fig. 5). It can be found that glomalin production increased from winter to autumn and spring with a corresponding decrease in SOC contents. This trend may suggest the existence of a balance between building fresh glomalin molecules for increasing organic carbon pools and their consequent consumption and transformation into the mineralized carbon forms as CO<sub>2</sub>. As a result, carbon resources used for building glomalin units may have been either soil carbon pools or photo-assimilated carbon or both depending on the environmental and soil conditions at any period of the year.

Additionally, it is well known that nitrogen is considered a major nutrient in soil metabolic activity (Vasconcelos et al., 2001). Nitrogen in glomalin has been quantified to be 3–5% of total N (Lovelock et al., 2004; Nichols and Wright, 2005) because GRSP was mainly composed of N-glycosidic bonds and amino acids forming the protein molecules (Wright and Upadhyaya, 1996). Soil nitrogen is probably required to build up the fresh protein units within the glycoprotein molecules contained in the GRSP<sub>EE</sub> as indicated with the significant positive correlations between TN and GRSP<sub>EE</sub> ( $r = 0.508$ ,  $P < 0.001$ ).

Veresoglou et al. (2011) reported that in the absence of P fertilization there are extensive flows of photo-assimilated carbon into the rhizosphere that support diverse root-associated microbial communities including AMF. As reported by Bücking and Shachar-Hill (2005) an increase in carbon supply by the plant to mycorrhizal associations increased the phosphorus uptake by plants. Mycorrhizal species possess different potentials among themselves to supply plants with phosphorus (Smith et al., 2003). We can assume that when more soil nutrients become available, plants may not allocate more nutrients to their mycorrhizal symbionts. The GRSP increased negatively with TN and TP during winter, spring, and autumn as in S4–S7 soils indicating that plants may allocate more N and P to mycorrhizal fungi as these soils showed an imbalance of soil nutrients, particularly during these seasons.

### 4.2. Stability index of formed aggregates

The structural stability of soil aggregates and their stability index increased in summer in all soils probably due to the steadier soil crumb arrangement achieved by the stabilization processes of SOM at high soil temperatures. These harsh conditions have probably driven the fungi associations to resist drought conditions by excreting various metabolites. Regarding the slope values of the linear equations in Fig. 3, the stability of aggregates showed higher resistance in the 0.25–2.00 mm fraction than in the 2.00–4.75 mm fraction due to the strong linkage of minerals and organic particles in the small-sized aggregates (Gunina and Kuzyakov, 2014; Trivedi

et al., 2017). Soils with better structure and high fertility as inS4-S6 were more stable compared to other soils with poor structure and low fertility as in S1-S3. The low stability in S7 was revealed to the soil compaction by cattle especially in the winter season.

#### 4.3. Environmental and soil conditions favoring GRSP production

Treseder and Turner (2007) stated that atmospheric CO<sub>2</sub> stimulates mycorrhizal species to produce more glomalin and increase soil nutrients availability through the photosynthetic processes. Typically, 12–27% of photo-assimilated carbon was given to mycorrhizal associations (Johnson et al., 1997). It was postulated that during winter, spring, and autumn, SOC stocks were used by mycorrhizal associations to form glomalin units, but the whole soil system was also subjected to CO<sub>2</sub> loss by consuming SOC pools. We may assume that this mechanism seems to be reversed in summer, when mycorrhizae may use photo-assimilated carbon to produce more glomalin. In summer, the highest carbon loss is suspected to give a discrepancy with the high GRSP production though carbon assimilation may compensate for the soil carbon loss. Conceptually, glomalin production in low carbon soil with low vegetation may be more dependent on SOC pools, while it may be less dependent in carbon-rich soils with high vegetation when carbon assimilation is increased by plant photosynthesis.

Rillig et al. (2003b) reported low GRSP<sub>EE</sub> (0.35 mg g<sup>-1</sup>) and GRSP<sub>T</sub> (2.6 mg g<sup>-1</sup>) in Morley silt loam soils (Mesic Typic Hapludalf) under cultivated *Zea mays* and *Glycine max*. They also recorded low GRSP<sub>EE</sub> (0.3 mg g<sup>-1</sup>) and GRSP<sub>T</sub> (2.5 mg g<sup>-1</sup>) in soils under the maple, elm, and walnut trees, similar to our values in S1-S3 soils. Likewise, the GRSP in Entisols investigated by Bedini et al. (2007) with 15 mg C g<sup>-1</sup> dry soil under maize contained low GRSP<sub>EE</sub> (0.25 mg g<sup>-1</sup>) and GRSP<sub>T</sub> (1.9 mg g<sup>-1</sup>). They also reported GRSP<sub>EE</sub> (0.5–1.5 mg g<sup>-1</sup>) and GRSP<sub>T</sub> (6–9 mg g<sup>-1</sup>) in soils dominated with *Populus alba* and grassland and were similar to S4-S7 soils probably due to higher SOC contents. Hontoria et al. (2009) reported low GRSP<sub>EE</sub> (0.27 mg g<sup>-1</sup>) and GRSP<sub>T</sub> (0.77 mg g<sup>-1</sup>) in cultivated olive in Haplic Calcisol with 71% of CaCO<sub>3</sub>. Similarly, Rillig et al. (2003a) recorded low GRSP<sub>EE</sub> (0.4 mg g<sup>-1</sup>) and GRSP<sub>T</sub> (1.4 mg g<sup>-1</sup>) in Lithic Calciorthids soils under *Brachypodium retusum* and *Stipa tenacissima* steppes in south-eastern Spain with a semi-arid Mediterranean climate. GRSP concentrations were generally low in soils with high CaCO<sub>3</sub> content probably due to the detrimental effects of Ca on mycorrhizal fungi (Roldán et al., 1997). Lovelock et al. (2004) reported an increase of GRSP<sub>EE</sub> (2.0 mg g<sup>-1</sup>) and GRSP<sub>T</sub> (4.1 mg g<sup>-1</sup>) in Ultisols with low Ca content than in Inceptisols (1.3 mg g<sup>-1</sup> of GRSP<sub>EE</sub> and 3.7 mg g<sup>-1</sup> of GRSP<sub>T</sub>) with high Ca content.

Lutgen et al. (2003) reported that vegetation under an adequate climate provided an increase in the GRSP<sub>EE</sub> (1.7 mg g<sup>-1</sup>) and GRSP<sub>T</sub> (8.0 mg g<sup>-1</sup>) in sandy loam soils of 3% SOC under the dense wheat-grass communities of *Festuca idahoensis* and *Agropyron spicatum*. In general, variation in GRSP contents can be outlined by the changes in land use and vegetation, soil phosphorus, and CaCO<sub>3</sub> contents (Lutgen et al., 2003; Rillig et al., 2003a; 2003b; Lovelock et al., 2004; Nichols and Wright, 2005; Bedini et al., 2007; Hontoria et al., 2009). Accordingly, the wide range of GRSP contents in our Lithic Xerorthents soils with no CaCO<sub>3</sub> contents can be mainly attributed to many factors affecting the environmental soil conditions and ecosystems of the studied shallow soils subjected to various agricultural activities, age of abandonment, and wildfire occurrence.

#### 4.4. Increasing soil carbon pools

Many authors reported that glomalin may contain from 279 to 431 mg C g<sup>-1</sup> GRSP defined as C-GRSP (Rillig et al., 2003b; Nichols and Wright, 2005) and may reach 52% in organic soils (Schindler

et al., 2007). Lovelock et al. (2004) reported similar values within a range of 28%–42% (mean 36%±0.83) when working with Ultisols and Inceptisols of Costa Rica tropical wet forest. Emran et al. (2017) reported a total C-GRSP content of 29 ± 1.5% in alkaline calcareous soils of Typic Calciorthids and alluvial alkaline soils of Vertic Torrifuvents under different agricultural management. In these soils, the C-GRSP recorded 7.19 ± 4.75%, 11.26 ± 14.26%, 8.54 ± 10.29%, 8.95 ± 9.34%, 9.11 ± 9.44%, 8.63 ± 6.67%, and 11.96 ± 10.87% for S1-S7 soils, respectively. The high data variability indicated by the standard deviation values can be explained by the highest C-GRSP values in summer varied within 14–32% among the studied soils. The C-GRSP in soil S7 was 66%, 6%, 40%, 34%, 31%, and 39% higher than in S1, S2, S3, S4, S5, and S6 soils, respectively. This indicates that carbon enrichment occurs with soil development along with land use and abandonment. The buildup of stable organic compounds in the developed soil was balanced by the continuous input of residues and their subsequent loss by the metabolic activity of microbial biomass (Liang et al., 2019). The C-GRSP amount found positively correlated with GRSP contents ( $y = 0.842x - 1.410$ ,  $r = 0.976$ ,  $P < 0.01$ ,  $n = 28$ ). On the one hand, when plotting C-GRSP/SOC to GRSP<sub>T</sub> a significant increase was found along the studied soil sequence showing high susceptibility to seasonal changes as indicated by the low values in winter, intermediate values in spring, and autumn, and the highest values in summer (Fig. 8A). On the other hand, the proportion of C-CO<sub>2</sub>/SOC decreased significantly with SOC ( $r = -0.924$ ,  $P < 0.01$ ) (Fig. 8B). The lowest proportion was in summer pointing that the fast transformation of labile SOC fractions (i.e. GRSP<sub>EE</sub>) may occur into stable

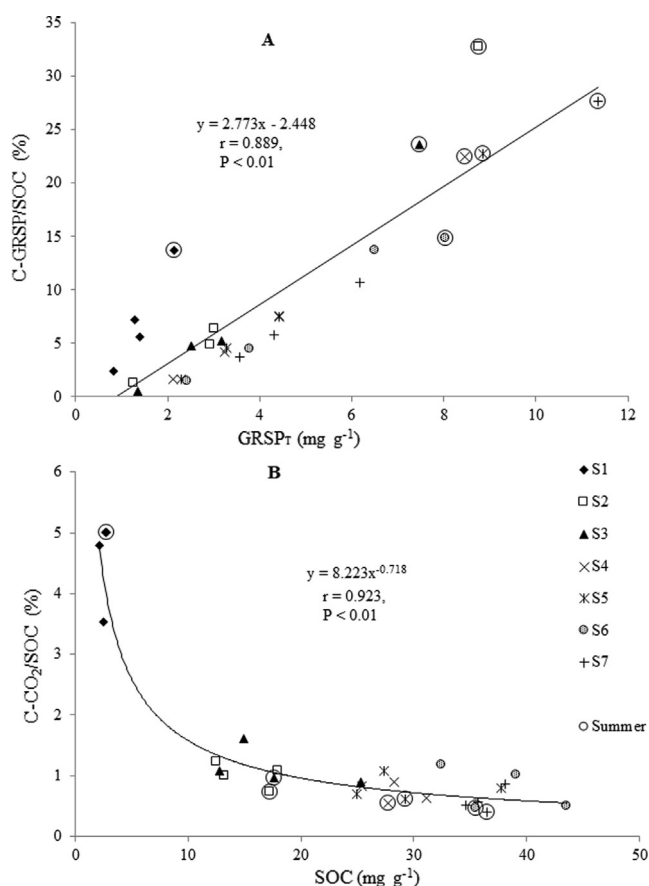


Fig. 8. Positive linear correlation between the seasonal means of C-GRSP/SOC and GRSP (A) and the negative power equation between the seasonal means of C-CO<sub>2</sub>/SOC and SOC (B) for the studied soils. Summer measurements are marked with a circle.

SOC fractions in this season. This pattern may indicate a dual trend in carbon dynamics (simultaneous loss and storage) depending on the changes in soil and environmental conditions along the four seasons. Preger et al. (2007) reported that GRSP content was not reduced below a certain steady-state level under prolonged arable cropping in sandy soils despite the potentially negative management effects. Our results showed that soil carbon is lost in comparatively low amounts in carbon-rich soils. The two carbon forms of C-GRSP and C-CO<sub>2</sub> in the studied soils suggested that C-GRSP in S1 soil may be prevalently formed by the labile GRSP fraction and most easily mineralized, thus higher C-CO<sub>2</sub> losses by soil respiration should be expected compared to other soils. Therefore amounts of C-CO<sub>2</sub> would decrease drastically from S1 to S7 due to the aging process of labile SOC fraction and increase of the recalcitrant GRSP fraction. Also, these processes of carbon storage and loss dynamics would have more relevance in summer than in spring, autumn, and winter. Further studies are needed to deepen into the relationships between the labile and stable fractions of SOC and GRSP, especially in abandoned and fire-affected soils, where complex biophysical processes occur in soils with organic structure contents.

The overview of these results may reveal that both assimilated C and SOC pools have significant impacts on GRSP concentrations. Plants may allocate excess nutrients even in N or P limited soils for improving nutrients acquisition by GRSP formation. Particularly, GRSP production increase based on the assimilated C was mainly occurring in summer either because plants are more N or P limited, photosynthate is more readily available, or both.

## 5. Conclusions

The GRSP in these shallow soils with very low clay contents should be considered a valuable resource of stable organic compounds for its contribution to soil nutrients acquisition, thus increasing aggregates stability and soil fertility. The frequent wildfire in S5, deposition of hardly decomposable pine needles in S3, and intense agricultural practices in S1-S2 soils decreased the soil nutrients and GRSP contents. Generally, more fertile and better-structured soils were observed in those soils with low wildfire occurrence and ancient abandonment. The labile GRSP fractions were supposed to be rapidly transformed into stable forms in these soils due to more suitable soil conditions, favoring the metabolic activities of soil microorganisms at the upper soil horizons. In addition, the GRSP contributed positively to increase soil carbon sequestration capacity.

## Declaration of Competing Interest

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

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## References

Aponte, C., Marañón, T., García, L.V., 2010. Microbial C, N and P in soils of Mediterranean oak forests: influence of season, canopy cover and soil depth. *Biogeochemistry* 101 (1–3), 77–92. <https://doi.org/10.1007/s10533-010-9418-5>.  
 Bedini, S., Avio, L., Argese, E., Giovannetti, M., 2007. Effects of long-term land use on arbuscular mycorrhizal fungi and glomalin-related soil protein. *Agric. Ecosyst. Environ.* 120 (2–4), 463–466. <https://doi.org/10.1016/j.agee.2006.09.010>.  
 Bücking, H., Shachar-Hill, Y., 2005. Phosphate uptake, transport and transfer by the arbuscular mycorrhizal fungus *Glomus intraradices* is stimulated by increased

carbohydrate availability. *New Phytol.* 165, 899–912. <https://doi.org/10.1016/j.agee.2006.09.010>.  
 Campbell E, J, Lobell B, D, Genova C, R, Zumkehr, A, Field B, C, 2013. Seasonal energy storage using bioenergy production from abandoned croplands. *Environ. Res. Lett.* 8, 035012. <https://doi.org/10.1088/1748-9326/8/3/035012>.  
 Cao, J., Wei, C., Adamowski, J.F., Zhou, J., Liu, C., Zhu, G., Dong, X., Zhang, X., Zhao, H., Feng, Q.i., 2020. Could arid and semi-arid abandoned lands prove ecologically or economically valuable if they afford greater soil organic carbon storage than afforested lands in China's Loess Plateau? *Land Use Policy* 99, 105027. <https://doi.org/10.1016/j.landusepol.2020.105027>.  
 Cerdà, A., Ackermann, O., Terol, E., Rodrigo-Comino, J., 2019. Impact of farmland abandonment on water resources and soil conservation in citrus plantations in eastern Spain. *Water* 11(4), 824. <https://www.mdpi.com/2073-4441/11/4/824>.  
 Cerdà, A., Borja, M.E.L., Úbeda, X., Martínez-Murillo, J.F., Keesstra, S., 2017. *Pinus halepensis* M. versus *Quercus ilex* subsp. *Rotundifolia* L. runoff and soil erosion at pedon scale under natural rainfall in Eastern Spain three decades after a forest fire. *For. Ecol. Manag.* 400, 447–456. <https://doi.org/10.1016/j.foreco.2017.06.038>.  
 Cleveland, C.C., Liptzin, D., 2007. C:N: P stoichiometry in soil: is there a “Redfield ratio” for the microbial biomass? *Biogeochemistry* 85 (3), 235–252. <https://doi.org/10.1007/s10533-007-9132-0>.  
 Emran, M., Gispert, M., Pardini, G., 2012a. Patterns of soil organic carbon, glomalin, and structural stability in Mediterranean abandoned terraced lands. *Eur. J. Soil Sci.* 63, 637–649. <https://doi.org/10.1111/j.1365-2389.2012.01493.x>.  
 Emran, M., Gispert, M., Pardini, G., 2012b. Comparing measurements methods of carbon dioxide fluxes in a soil sequence under land use and cover change in North Eastern Spain. *Geoderma* 170, 176–185. <https://doi.org/10.1016/j.geoderma.2011.11.013>.  
 Emran, M., Rashad, M., Gispert, M., Pardini, G., 2017. Increasing Soil Nutrients Availability and Sustainability by Glomalin in Alkaline Soils. *International Journal of Agricultural and Biosystems Engineering* 2, 74–84. <http://www.aasci.org/journal/ijabe>.  
 Emran, M., Doni, S., Macci, C., Masciandaro, G., Rashad, M., Gispert, M., 2020. Susceptible soil organic matter, SOM, fractions to agricultural management practices in salt-affected soils. *Geoderma* 366, 114257. <https://doi.org/10.1016/j.geoderma.2020.114257>.  
 Gispert, M., Emran, M., Pardini, G., Doni, D., Ceccanti, B., 2013. The impact of land management and abandonment on enzymatic activity, glomalin content, and aggregate stability. *Geoderma* 202–203, 51–61. <https://doi.org/10.1016/j.geoderma.2013.03.012>.  
 Gispert, M., Pardini, G., Coldecarrera, M., Emran, M., Doni, S., 2017. Water erosion and soil properties patterns along selected rainfall events in cultivated and abandoned terraced fields under renaturalisation. *Catena* 155, 114–126. <https://doi.org/10.1016/j.catena.2017.03.010>.  
 Gispert, M., Pardini, G., Emran, M., Doni, S., Masciandaro, G., 2018. Seasonal evolution of soil organic matter, glomalin and enzymes and potential for C storage after land abandonment and renaturalization processes in soils of NE Spain. *Catena* 162, 402–413. <https://doi.org/10.1016/j.catena.2017.10.019>.  
 Gispert, M., Kulush, T., Dyachenko, L., Kharytonov, M., Emran, M., Verdager, D., Llorens, L., Carrasco-Barea, L., 2021. Appraising soil carbon storage potential under perennial and annual Chenopodiaceae in salt marsh of NE Spain. *Estuarine, Coastal and Shelf Science* 252, 107240. <https://doi.org/10.1016/j.ecss.2021.107240>.  
 Gunina, A., Kuzyakov, Y., 2014. Pathways of litter C by formation of aggregates and SOM density fractions: implications from <sup>13</sup>C natural abundance. *Soil Biol. Biochem.* 71, 95–104. <https://doi.org/10.1016/j.soilbio.2014.01.011>.  
 Guo, X.i., Jiang, Y., 2019. Spatial characteristics of ecological stoichiometry and their driving factors in farmland soils in Poyang Lake Plain. Southeast China. *J. Soil Sediment.* 19 (1), 263–274. <https://doi.org/10.1007/s11368-018-2047-7>.  
 Hontoria, C., Velásquez, R., Benito, M., Almorox, J., Moliner, A., 2009. Bradford-reactive soil proteins and aggregate stability under abandoned versus tilled olive groves in a semi-arid calcisol. *Soil Biol. Biochem.* 41 (7), 1583–1585. <https://doi.org/10.1016/j.soilbio.2009.04.025>.  
 Jiang, Y., Guo, X.i., 2019. Stoichiometric patterns of soil carbon, nitrogen, and phosphorus in farmland of the Poyang Lake region in Southern China. *J. Soil Sediment.* 19 (10), 3476–3488. <https://doi.org/10.1007/s11368-019-02317-3>.  
 Johnson, D.L., Ambrose, S.H., Bassett, T.J., Bowen, M.L., Crummey, D.E., Isaacson, J.S., Johnson, D.N., Lamb, P., Saul, M., Winter-Nelson, A.E., 1997. Meanings of environmental terms. *J. Environ. Qual.* 26 (3), 581–589. <https://doi.org/10.2134/jeq1997.00472425002600030002x>.  
 Kemper D, W, Rosenau C, R, 1986. Aggregate stability and size distribution. *Methods of soil analysis, Part 1. Agronomy Monograph 9..* Madison, Wisconsin, pp. 425–442.  
 Liang, C., Amelung, W., Lehmann, J., Kästner, M., 2019. Quantitative assessment of microbial necromass contribution to soil organic matter. *Glob. Chang. Biol.* 25 (11), 3578–3590. <https://doi.org/10.1111/gcb.v25.1110.1111/gcb.14781>.  
 Lovelock, C.E., Wright, S.F., Clark, D.A., Ruess, R.W., 2004. Soil stocks of glomalin produced by arbuscular mycorrhizal fungi across a tropical rain forest landscape. *J. Ecol.* 92, 278–287. <https://doi.org/10.1111/j.0022-0477.2004.00855.x>.  
 Lutgen, E.R., Muir-Clairmont, D., Graham, J., Rillig, M.C., 2003. Seasonality of arbuscular mycorrhizal hyphae and glomalin in a western Montana grassland. *Plant Soil* 257 (1), 71–83. <https://doi.org/10.1023/A:1026224209597>.  
 Nichols, K.A., Wright, S.F., 2005. Comparison of glomalin and humic acid in eight native U.S. Soils. *Soil Sci.* 170 (12), 985–997. <https://doi.org/10.1097/01.ss.0000198618.06975.3c>.

- Ovsepyan, L., Kurganova, I., Gerenyu, V.L., Kuzyakov, Y., 2019. Recovery of organic matter and microbial biomass after abandonment of degraded agricultural soils: the influence of climate. *Land Degrad. Dev.* 30 (15), 1861–1874. <https://doi.org/10.1002/ldr.3387>.
- Palmqvist, K., 2002. Cyanolichens: Carbon Metabolism. In: Rai, A.N., Bergman, B., Rasmussen, U. (Eds.), *Cyanobacteria in Symbiosis*. Kluwer Academic Publishers, Printed in the Netherlands, pp. 73–96.
- Pardini, G., Gispert, M., Dunjo, G., 2004. Relative influence of wildfire on soil properties and erosion processes in different Mediterranean environments in NE Spain. *Scie. Total Environ.* 328 (1–3), 237–246. <https://doi.org/10.1016/j.scitotenv.2004.01.026>.
- Pardini, G., Gispert, M., Emran, M., Doni, S., 2017. Rainfall/runoff/erosion relationships and soil properties survey in abandoned shallow soils of NE Spain. *J. Soil Sediment.* 17 (2), 499–514. <https://doi.org/10.1007/s11368-016-1532-0>.
- Poepplau, C., Don, A., Vesterdal, L., Leifeld, J., Van Wesemael, B.A.S., Schumacher, J., Gensior, A., 2011. Temporal dynamics of soil organic carbon after land-use change in the temperate zone—carbon response functions as a model approach. *Global Change Biology* 17 (7), 2415–2427.
- Preger, A.C., Rillig, M.C., Johns, A.R., Du Preez, C.C., Lobe, I., Amelung, W., 2007. Losses of glomalin-related soil protein under prolonged arable cropping: a chronosequence study in sandy soils of the South African Highveld. *Soil Biol. Biochem.* 39 (2), 445–453. <https://doi.org/10.1016/j.soilbio.2006.08.014>.
- Redfield, A.C., 1958. The biological control of chemical factors in the environment. *Am. Sci.* 46, 205–219. <https://www.jstor.org/stable/27827150>
- Reich, P.B., Oleksyn, J., 2004. Global patterns of plant leaf N and P in relation to temperature and latitude. *Proc. Natl. Acad. Sci. USA* 101 (30), 11001–11006. <https://doi.org/10.1073/pnas.0403588101>.
- Reyes, H.A., Ferreira, P.F.A., Silva, L.C., da Costa, M.G., Nobre, C.P., Gehring, C., 2019. Arbuscular mycorrhizal fungi along secondary forest succession at the eastern periphery of Amazonia: Seasonal variability and impacts of soil fertility. *Appl. Soil Ecol.* 136, 1–10. <https://doi.org/10.1016/j.apsoil.2018.12.013>.
- Rillig, M.C., Maestre, F.T., Lamit, L.J., 2003a. Microsite differences in fungal hyphal length, glomalin, and soil aggregate stability in semiarid Mediterranean steppes. *Soil Biol. Biochem.* 35 (9), 1257–1260. [https://doi.org/10.1016/S0038-0717\(03\)00185-8](https://doi.org/10.1016/S0038-0717(03)00185-8).
- Rillig, M.C., Ramsey, P.W., Morris, S., Paul, E.A., 2003b. Glomalin, an arbuscular-mycorrhizal fungal soil protein responds to land use change. *Plant Soil* 253, 293–299. <https://doi.org/10.1023/A:1024807820579>.
- Rillig, M.C., Wright, S.F., Kimball, B.A., Pinter, P.J., Wall, G.W., Ottman, M.J., Leavitt, S.W., 2001. Elevated carbon dioxide and irrigation effects on water stable aggregates in a sorghum field: A possible role for arbuscular mycorrhizal fungi. *Glob. Chang. Biol.* 7, 333–337. <https://doi.org/10.1046/j.1365-2486.2001.00404.x>.
- Rivoal, A., Fernandez, C., Lavoit, A.V., Olivier, R., Lecareux, C., Greff, S., Roche, P., Vila, B., 2010. Environmental control of terpene emissions from *Cistus monspeliensis* L. in natural Mediterranean shrublands. *Chemosphere* 78 (8), 942–949. <https://doi.org/10.1016/j.chemosphere.2009.12.047>.
- Roldan, A., Garcia, C., Albaladejo, J., 1997. AM fungal abundance and activity in a chronosequence of abandoned fields in a semiarid Mediterranean site. *Arid Soil. Rehab.* 11 (3), 211–220. <https://doi.org/10.1080/15324989709381474>.
- Ryan, J., Estefan, G., Rashid, A., 2001. Soil and plant analysis laboratory manual. (second ed.), Jointly published by the International Center for agricultural Research in the dry Areas (ICARDA) and the National agricultural Research Center (NARC), Aleppo, Syria, pp. 46–48.
- Sardans, J., Rodà, F., Peñuelas, J., 2006. Effects of a nutrient pulse supply on nutrient status of the Mediterranean trees *Quercus ilex* subsp. *ballota* and *Pinus halepensis* on different soils and under different competitive pressure. *Trees* 20 (5), 619–632. <https://doi.org/10.1007/s00468-006-0077-z>.
- Schindler, F.V., Mercer, E.J., Rice, J.A., 2007. Chemical characteristics of glomalin-related soil protein (GRSP) extracted from soils of varying organic matter content. *Soil Biol. Biochem.* 39 (1), 320–329. <https://doi.org/10.1016/j.soilbio.2006.08.017>.
- Soil Survey Staff., 2014. Keys to soil taxonomy. 12<sup>th</sup> ed. USDA-NRCS, Washington, DC, pp. 141, 310.
- Smith, S., Smith, A., Jakobsen, I., 2003. Mycorrhizal fungi can dominate phosphate supply to plants irrespective of growth responses. *Plant Physiol.* 133 (1), 16–20. <https://doi.org/10.1104/pp.103.024380>.
- StatSoft Inc. 2011. STATISTICA (data analysis software system), version 10. www.statsoft.com.
- Tian, H.Q., Chen, G.S., Zhang, C., Melillo, J.M., Hall, C.A.S., 2010. Pattern and variation of C:N: P ratios in China's soils: a synthesis of observational data. *Biogeochemistry* 98, 139–151. <https://doi.org/10.1007/s10533-009-9382-0>.
- Treseder, K., Turner, K.M., 2007. Glomalin in Ecosystems. *Soil Sci. Soc. Am. J.* 71, 1257–1266. <https://doi.org/10.2136/sssaj2006.0377>
- Treseder, K., Egerton-Warburton, L.M., Allen, M.F., Cheng, Y.F., Oechel, W.C., 2003. Alteration of soil carbon pools and communities of mycorrhizal fungi in chaparral exposed to elevated carbon dioxide. *Ecosystems* 6, 786–796. <https://doi.org/10.1007/s10021-003-0182-4>
- Trivedi, P., Delgado-Baquerizo, M., Jeffries, T.C., Trivedi, C., Anderson, I.C., Lai, K., McNee, M., Flower, K., Pal Singh, B., Minkey, D., Singh, B.K., 2017. Soil aggregation and associated microbial communities modify the impact of agricultural management on carbon content. *Environ. Microbiol.* 19(8), 3070–3086. <https://doi.org/10.1111/1462-2920.13779>
- Turner, B.L., Yavitt, J.B., Harms, K.E., Garcia, M.N., Wright, S.J., 2015. Seasonal changes in soil organic matter after a decade of nutrient addition in a lowland tropical forest. *Biogeochemistry* 123 (1–2), 221–235.
- Vasconcelos, M.C., Keller, M., Kummer, M., 2001. Soil nitrogen utilisation for growth and gas exchange by grapevines in response to nitrogen supply and rootstock. *Aust. J. Grape Wine R.* 7, 2–11. <https://doi.org/10.1111/j.1755-0238.2001.tb00187.x>.
- Veresoglou, S.D., Mamolos, A.P., Thornton, B., Voulgari, O.K., Sen, R., Veresoglou, D.S., 2011. Medium-term fertilization of grassland plant communities masks plant species-linked effects on soil microbial community structure. *Plant Soil* 344 (1–2), 187–196. <https://doi.org/10.1007/s11104-011-0739-5>.
- Wang, Q., Lu, H., Chen, J., Hong, H., Liu, J., Li, J., Yan, C., 2018. Spatial distribution of glomalin-related soil protein and its relationship with sediment carbon sequestration across a mangrove forest. *Sci. Total Environ.* 613, 548–556. <https://doi.org/10.1016/j.scitotenv.2017.09.140>.
- Wright, S.F., Nichols, K.A., Schmidt, W.F., 2006. Comparison of efficacy of three extractants to solubilize glomalin on hyphae and in soil. *Chemosphere* 64 (7), 1219–1224. <https://doi.org/10.1016/j.chemosphere.2005.11.041>.
- Wright, S.F., Upadhyaya, A., 1996. Extraction of an abundant and unusual protein from soil and comparison with hyphal protein of arbuscular mycorrhizal fungi. *Soil Sci.* 161 (9), 575–586. <https://doi.org/10.1097/00010694-199609000-00003>.
- Zhang, J., Tang, X., He, X., Lin, J., 2015. Glomalin related soil protein responses to elevated CO<sub>2</sub> and nitrogen addition in a subtropical forest. Potential consequences for soil carbon accumulation. *Soil Biol. Biochem.* 33, 142–149. <https://doi.org/10.1016/j.soilbio.2015.01.023>.
- Fazhu, Z., Jiao, S., Chengjie, R., Di, K., Jian, D., Xinhui, H., Gaihe, Y., Yongzhong, F., Guangxin, R., 2015. Land use change influences soil C, N, and P stoichiometry under 'Grain-to-Green Program' in China. *Sci. Rep.* 5, 10195. <https://doi.org/10.1038/srep10195>.