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Soil quality evaluation following the implementation of permanent cover crops in semi-arid vineyards. Organic matter, physical and biological soil properties

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Abstract

Changing from conventional vineyard soil management, which includes keeping bare soil through intense tilling and herbicides, to permanent grass cover (PGC) is controversial in semi-arid land because it has agronomic and environmental advantages but it can also induce negative changes in the soil physical status. The objectives of this work were (i) gaining knowledge on the effect of PGC on the soil physical and biological quality, and (ii) identifying the most suitable soil quality indicators for vineyard calcareous soils in semi-arid land. Key soil physical, organic and biological characteristics were determined in a *Cambic Calcisol* with different time under PGC (1 and 5 years), and in a conventionally managed control. Correlation analysis showed a direct positive relationship between greater aggregate stability (WSA), soil-available water capacity (AWC), microbial biomass and enzymatic activity in the topsoil under PGC. Total and labile organic C concentrations (SOC and POM-C) were also correlated to microbial parameters. Factor analysis of the studied soil attributes using principal component analysis (PCA) was done to identify the most sensitive soil quality indicators. Earthworm activity, AWC, WSA, SOC and POM-C were the soil attributes with greater loadings in the two factors determined by PCA, which means that these properties can be considered adequate soil quality indicators in this agrosystem. These results indicate that both soil physical and biological attributes are different under PGC than in conventionally-managed soils, and need therefore to be evaluated when assessing the consequences of PGC on vineyard soil quality.

Additional key words: calcareous soils; cover crops; permanent land cover; soil conservation; vineyard soil management.

Resumen

Evaluación de la calidad del suelo tras la implantación de cubiertas permanentes en viñedos de zonas semiáridas. Materia orgánica y propiedades físicas y biológicas del suelo

El establecimiento de cubiertas vegetales permanentes (PGC) en viñedos de zonas semiáridas, con manejo tradicional de suelo desnudo mediante laboreo y aplicación de herbicidas, es controvertido, porque tiene ventajas agronómicas y ambientales, pero puede inducir cambios negativos en la calidad física del suelo. Los objetivos de este trabajo fueron: (i) avanzar en el conocimiento del efecto de la implantación de PGC en la calidad física y biológica del suelo, e (ii) identificar los indicadores de calidad del suelo más apropiados para suelos calizos de viñedo en una zona semiárida. Se determinaron propiedades físicas y biológicas clave en un *Calcisol Cámbico* con PGC de diferente edad (1 y 5 años), con un control manejado convencionalmente. El análisis de correlaciones mostró una relación directa entre la estabilidad estructural (WSA), la capacidad de retención de agua útil (AWC), la biomasa microbiana y las actividades enzimáticas del suelo bajo PGC. El contenido de C orgánico total (SOC) y lábil (POM-C) estuvo también correlacionado

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Abbreviations used: AWC (plant-available water holding capacity); CLPP (community-level physiological profile); FA (factor analysis); NSU (number of substrates used); PCA (principal components analysis); PGC (permanent grass cover); POM (particulate organic matter); PR (penetration resistance); PSD (particle-size distribution); SOC (soil organic carbon); SWR (soil water retention); WSA (water-stable aggregates); ρb (bulk density).

con los parámetros microbianos. Los indicadores de calidad del suelo más sensibles se identificaron mediante análisis factorial por componentes principales (PCA). La actividad de lombrices, AWC, WSA, SOC y POM-C mostraron el mayor peso en los dos factores obtenidos con PCA, por lo que estas propiedades pueden considerarse indicadores adecuados de la calidad del suelo en este agrosistema. Estos resultados indican que tanto los atributos físicos como biológicos del suelo son diferentes bajo PGC, y necesitan ser evaluados al estudiar las consecuencias de su introducción en suelos de viñedo.

Palabras clave adicionales: conservación del suelo; cubiertas permanentes; cultivos de cobertera; manejo de suelos de viñedo; suelos calizos.

Introduction

The implementation of permanent grass cover crops (PGC) in the interrows of vineyards is not a common practice in semi-arid land, but is at present spreading in many of these areas, as it is seen as an adequate agronomic strategy to control vines vegetative growth and yield (*e.g.* Ferrini *et al.*, 1996; Monteiro & Lopes, 2007; Tesic *et al.*, 2007; Ripoche *et al.*, 2011). Such implementation implies changing the conventional vineyard soil management in most semi-arid areas, which includes intense tilling and herbicide applications to keep a weed-free bare soil surface.

Permanent grassing is however controversial, because it appears to have both positive and negative potential consequences in the soil-plant system (*e.g.* Ruiz-Colmenero *et al.*, 2011). On the one hand, it is known that intense tillage can result in reduced physical quality (Álvaro-Fuentes *et al.*, 2008), including reduced water-holding capacity (Bescansa *et al.*, 2006), and increased erosion risks (Ramos & Martínez-Casasnovas, 2006; Quiquerez *et al.*, 2008). In contrast, permanent covering of the soil, such as achieved under no-tillage, can contribute to improve soil quality in semi-arid land (Moreno *et al.*, 1997; Bescansa *et al.*, 2006; Lampurlanés & Cantero-Martinez, 2006; Virto *et al.*, 2007; Imaz *et al.*, 2010). For instance, reduced erosion losses have been observed in vineyards where soil management techniques including reduced soil disturbance have been implemented (Shepard, 2006; Ruiz-Colmenero *et al.*, 2011). This has been associated to enhanced biological activity (Ros *et al.*, 2009), as a result of increased organic C content, of reduced evaporation, and of the suppression of tillage-induced disturbances. This seems of special importance in semi-arid lands, where soils are often poor in organic matter, as a result of historical land use, low organic inputs (Zornoza *et al.*, 2007), and a climate that favors the rapid mineralization of crop residues (Raich & Schlesinger, 1992). For instance, a recent study (Peregrina *et al.*, 2010) has demonstrated that PGC in a semi-arid vineyard resulted in enhanced organic matter stocks and quality, and greater aggregate stability. Ruiz-Colmenero *et al.* (2011) have also observed reduced erosion rates in three semi-arid rain fed vineyards in Spain. The implementation of PGC in the inter-rows in vineyards seems thus interesting from a soil and water conservation perspective (Klik *et al.*, 1998; Steenwerth & Belina, 2008a,b).

On the other hand, many producers are reluctant with regard to PGC adoption because they fear deleterious effects such as the redistribution of the vines root system (Morlat & Jacquet, 2003) due to the alteration of the water flow in the soil (Celette *et al.,* 2008), and the problems of soil compaction usually observed following the suppression of tillage (Tebrügge & Düring, 1999). Reality is that few studies have been conducted on the effect of PGC in the soil system in semi-arid land, and a gap of knowledge exists at present. In particular, information is still needed on the changes induced in the soil physical and biological status by this soil management practice, and on which soil quality indicators are the most adequate ones to evaluate such effect.

It can be hypothesized that two opposing facts affecting soil quality might interact when a permanent grass crop is sown in vineyards. First, such implementation might affect the topsoil physical quality, by increasing its compaction and by inducing changes in its pore system resulting in changes in its water-holding ability. Simultaneously, the observed gain in organic matter (Peregrina *et al.*, 2010), should favor the development of a more stable soil structure (Virto *et al.*, 2007), which could counteract these changes of the soil physical status. For instance Ruiz-Colmenero *et al.* (2011) observed that the introduction of PGC reduced soil surface sealing in comparison to the traditional tillage management.

In this work, key soil physical, organic and biological properties were studied in a calcareous vineyard soil after different times under PGC in a traditional vineyard located in a semi-arid area. The objectives were (i) gaining knowledge on the effect of PGC implementation on the soil physical and biological quality, and (ii) identifying the most suitable soil quality indicators for vineyard calcareous soils.

Material and methods

Site and vineyard management

This study was conducted in a 46 ha production vineyard near Olite (Navarra), in NE Spain, where vineyards cover more than 140,000 ha (MARM, 2009), 15,600 of which fall within the designation of origin "Navarra" (Gobierno de Navarra, 2008). According to UNESCO (1979), semi-arid regions are those with a precipitation-to-potential evapotranspiration (ETo) ratio greater than 0.2 and smaller than 0.5. The studied area, with an average rainfall of 525 mm, only 18% of which in summer (July-September), and an annual mean Penman's ETo of 1173 mm falls within this range. Climate is dry Mediterranean, according to Papadakis (1975).

Vines (3,333 plants ha^{-1}) in the study site were planted in rows (3 m wide). After 10 years of continuous conventional management of the soil, a mixture of tall fescue (*Festuca arundinacea* Schreb., 25 kg ha^{-1}) and Italian ryegrass (*Lolium multiflorum* L., 10 kg ha^{-1}) started to be progressively sown in the inter-rows of some areas. As a result, areas with different time under PGC exist, along with a control area in which the soil is kept free of vegetation with conventional management (including intense tilling and herbicide applications for weed control).

Management of the vineyard was similar in all areas. Ten to 20 kg of N ha^{-1} were applied annually for PGC fertilization after the first mowing just before springtime. In all treatments, herbicide (glyphosate) was applied for weed control in the under-vine.

No organic amendments were applied. Management of PGCs was adapted to the phenology of grapevines, and included mowing in early March (before budbreaking) and at berry-touch. After mowing, grass clippings (together with chopped pruning residues in March) were left on the ground. In the dry season (May-September) grass was let to wilt naturally.

Soil and sampling design

The soil was a *Cambic Calcisol* (FAO, 1998). In the tilled depth (0-30 cm), it contains in average 19.1% clay, 34.8% silt and 46.1% sand. Average total carbonates were 35.4% of the soil mass and the pH was 8.4.

A set of three plots (125 × 75 m each) was selected for this study: one in the tilled area (TILL), used as the reference of the conventional management, and two within two contiguous areas with 1 (ONEYR), and 5 years (FIVEYR) under PGC. To exclude boundary effects, the study plots were located in the center of each area, so they were not contiguous to each other. For sampling, each plot was divided into four quadrants, and each part was considered as a field (pseudo) replicate ($n = 4$). In each quadrant, three equidistant inter-rows were selected as sample locations. The sampling points in each quadrant were set as separated as possible from those in the adjacent quadrants.

For the study, disturbed and undisturbed samples were collected at each sampling location in one single sampling campaign in spring (April) of the same farm year in the 0 to 5, 5 to 15 and 15 to 30-cm depth increments. These depth increments are noted as 0-5, 5-15 and 15-30 hereafter. Disturbed samples consisted of five subsamples randomly collected and combined to obtain a composite sample. Two portions of the 0-5 composite sample were gently pushed through a 8-mm and 2-mm sieves, respectively, and stored fresh (4°C) for some soil biological determinations (see below). The remainder of the samples was air-dried and ground to pass a 2 mm sieve. A portion of this was stored at 4°C for the enzyme analyses (see below).

Undisturbed core samples were collected using bevel-edged steel rings at 0-5, 5-15 and 15-30 to determine soil bulk density (ρ_b) and the soil water retention at field capacity. Finally, two soil blocks (20 × 20 × 20 cm) were taken at each sample location for earthworms counting and characterization.

Soil analysis

The soil static properties which are not expected to change following PGC implementation but can influence other properties that may change (particle-size distribution and equivalent carbonates content) were first measured following standard protocols (Goh *et al.*, 1993; Sheldrick & Wang, 1993). Soil dynamic properties most related to the soil physical and biological quality considering the particularities of the studied soil, were then measured.

The physical status of the soil was evaluated through its ρ_b, aggregate stability, penetration resistance (PR), and available soil-water retention capacity (AWC). Bulk density was measured by oven-drying (105°C) and weighing the undisturbed soil cores. Aggregate stability was quantified by wet sieving 20 g of air-dried soil < 2 mm on a 250-µm sieve using following sudden immersion in 150 mL of water. Samples were sieved for 3 min, the stable fraction was corrected for sand content after dispersion with 5% $(NaPO_3)_6$, and the water stable aggregates (WSA) index was calculated as the mass percentage of stable aggregates > 250 µm. This method has been already used and seen to be efficient in evaluating the stability of vineyard soils in the Mediterranean Basin (Le Bissonnais *et al.*, 2007).

Penetration resistance was measured in the field to a depth of 45 cm using a field penetrometer (Rimik CP20, Agridry Rimik Pty Ltd, Toowoomba, QLD, Australia). At each sample location, three measurements were taken along the inter-row width, which makes 36 readings per plot, recording PR at 15-mm depth intervals. This was done after an important rain episode that left the soil homogenously wet, to avoid differences in moisture content between treatments.

Soil water retention (SWR) was measured at a matric potential of –33 kPa (equivalent to field capacity) and at –1,500 kPa (equivalent to permanent wilting point), as described by Dirksen (1999). Volumetric values of SWR were calculated from the gravimetric measures using ρ_b. Soil AWC was calculated from the difference in soil moisture content at –33 kPa and –1,500 kPa.

The biological properties of the soil were evaluated through the total and labile organic fractions, microbiological characteristics (biomass, functional and metabolic diversity), and earthworms activity.

Total soil organic carbon content (SOC) was analyzed in air-dried samples. Due to the elevated carbonate content of the soil, wet oxidation (Walkley-Black) was used to analyze total oxidisable C (Tiessen & Moir, 1993), that we assumed equal to total organic C. Particulate organic matter (POM), which represents the labile fraction of SOC (Cambardella & Elliott, 1992), was isolated using the method described in Virto *et al.* (2007). Organic C in the form of POM (POM-C) was determined by wet oxidation.

Microbial soil properties were studied only in 0-5, because no differences in organic C were observed below this depth (see below), and because changes in these properties [which are known precocious soil quality indicators (Mijangos *et al.*, 2009)] related to management can be more easily detected in the surface layer (Mijangos & Garbisu, 2010). Microbial biomass C was determined in 0-5 samples by the fumigation-extraction method, which was essentially that of Vance *et al.* (1987). The functional and metabolic diversity of the soil microbial population was studied through the analysis of enzyme activities and of the community-level physiological profiles (CCPLs) in 0-5 samples. The activities of dehydrogenase and four enzymes involved in soil C (β-glucosidase), N (urease), P (alkaline phosphatase) and S (aril-sulphatase) dynamics were determined according to Dick *et al.* (1996) and Taylor *et al.* (2002), as in Epelde *et al.* (2008). For dehydrogenase activity, 1-g of field-moist soil ground to pass a 2-mm sieve and stored at 4°C was used for the analysis. Following Dick *et al.* (1996), who determined that the activity of some enzymes is more stable in air-dried than in field-moist samples, air-dried samples stored at 4°C were used in the determination of the other four enzyme activities. The C source utilization patterns observed using a Biolog Ecoplate™ microplating system (Biolog, Hayward, CA, USA) were used to determine CLPPs, as described in Epelde *et al.* (2008) and Mijangos *et al.* (2009). Ecoplates™ were designed for determining CLPPs of terrestrial communities, and comprise 31 C substrates that are major ecologically relevant compounds. The number of substrates used by the soil microbial community (NSU), equivalent to species richness (Zak *et al.,* 1994) was quantified as the number of wells showing corrected absorbance values > 0.25 (onset of the exponential microbial growth in the Biolog Ecoplate™ microplates, data not shown).

Finally, earthworms were sampled from the undisturbed soil blocks by hand-sorting. They were counted in the field and weighed (fresh weight basis) in the laboratory.

Statistics

Since the use of pseudo-replicates (inherent to this and any other study conducted in production fields) means that differences in the studied soil parameters cannot be attributed exclusively to treatments, data were analyzed in three steps. First, individual analysis of variance (ANOVAs) was run for each studied soil property and depth, using Bonferroni correction to test differences among means, and in post-hoc analysis. This correction implies a conservative approach to multiple comparisons, and allowed for determining the most important differences in the soil parameters among TILL, ONEYR and FIVEYR. Second, correlation analysis [Pearson correlation ($p < 0.05$)] was used to analyze the relationships among the studied soil properties at 0-5.

Finally, considering also the particularities of the experimental setup, data corresponding to variables showing significant differences among treatments were subjected to factor analysis (FA) using principal component analysis (PCA). This was done with the aim of evaluating overall soil quality and identifying the most sensitive soil quality indicators among the studied soil properties, as in Imaz *et al.* (2010). Only variables from the upper studied depth were studied, because differences among treatments in 5-15 and 15-30 were observed in few or none of the studied physical and biological parameters. PCA allows for grouping the studied variables into statistical factors based on their correlation structure (Brejda *et al.*, 2000). To eliminate the effect of different units, FA was done using the correlation matrix on the standardized values of the measured soil properties, so that each variable had mean = 0 and variance = 1 (Shukla *et al.*, 2006). Using this correlation matrix, principal components (factors) with eigenvalues > 1 were retained and subjected to varimax rotation to estimate the proportion of the variance of each soil variable explained by each selected factor (loadings). Factor scores for each sample point were calculated, and one-way ANOVAs were run in order to evaluate the significance of the differences found in such scores among the three studied plots.

All statistical analyses were performed using SPSS 17.0 software (SPSS Inc., 2010).

Results and discussion

Soil physical and biological properties

No significant differences were observed in the particle-size distribution and equivalent carbonates content among the studied areas (Table 1).

Differences in the physical status, organic matter fractions and surface biological status are summarized in Tables 2, 3 and 4, respectively. Curves of PR are represented in Fig. 1. As hypothesized, differences between the control (TILL) and the soil under PGC (ONEYR and FIVEYR) were observed in most of these parameters. These differences were however not homogeneous in depth, and among plots with PGC.

For instance, greater surface values of PR in ONEYR and FIVEYR (Fig. 1) in relation to TILL revealed some compaction of the soil associated to the presence of PGC, although the effects of this compaction were limited to the upper 7.5 cm (Fig. 1). Conversely, no significant differences were observed in this depth in ρ_b, which was greater in ONEYR in comparison to TILL and FIVEYR only in 15-30 (Table 2). Bulk density was in fact the only parameter displaying significant differences at 15-30. Similar to PR, differences in total SOC and AWR were significant only in 0-5.

In relation to the soil physical status, average topsoil PR values under PGC were similar to those found in

Table 1. Soil particle-size distribution (PSD) and total equivalent carbonates ($CaCO_3$) in the three studied plots (TILL, ONEYR and FIVEYR) (mean ± standard error)

	TILL			ONEYR			FIVEYR		
	Clay	Silt	Sand	Clay	Silt	Sand	Clay	Silt	Sand
PSD (mg g^{-1} soil)									
0-5	219.8 ± 18.5	204.7 ± 3.50	575.5 ± 21.7	190.0 ± 3.60	170.8 ± 11.9	639.2 ± 11.6	219.4 ± 8.0	190.7 ± 5.60	590.0 ± 13.3
5-15	232.5 ± 14.1	207.8 ± 5.10	559.7 ± 17.9	212.1 ± 10.1	162.0 ± 10.7	625.9 ± 19.5	256.9 ± 8.3	178.7 ± 4.70	564.4 ± 9.80
15-30	229.8 ± 10.0	220.8 ± 14.5	549.4 ± 23.3	219.2 ± 16.8	149.9 ± 11.0	630.9 ± 27.5	265.0 ± 10.7	171.2 ± 6.30	563.9 ± 16.2
$CaCO_3$ (%)									
0-5		34.1 ± 1.5			34.6 ± 2.6			30.4 ± 0.7	
5-15		33.0 ± 0.7			35.0 ± 2.9			31.3 ± 0.3	
15-30		32.5 ± 0.9			34.5 ± 2.8			31.8 ± 0.6	

Table 2. Bulk density, water-stable aggregates (WSA) and available soil-water retention capacity (AWC) in TILL (no PGC), and after one (ONEYR) and five (FIVEYR) years of PGC (mean ± standard error)

	TILL	ONEYR	FIVEYR
Bulk density (ρb, Mg m^{-3})			
0-5	1.29 ± 0.1	1.38 ± 0.8	1.63 ± 0.0
5-15	1.56 ± 0.0	1.55 ± 0.7	1.66 ± 0.0
15-30	1.46 ± 0.3 a	1.72 ± 0.0 b	1.49 ± 0.0 a
Aggregate stability (WSA, %)			
0-5	5.68 ± 8.1 a	24.9 ± 1.4 ab	41.5 ± 9.5 b
5-15	11.43 ± 6.3 a	48.9 ± 3.1 b	52.9 ± 3.3 b
15-30	30.7 ± 3.8	57.4 ± 5.5	66.8 ± 1.2
Water retention (AWC, mm)			
0-5	4.20 ± 0.2 a	4.71 ± 0.1 a	5.71 ± 0.2 b
5-15	17.4 ± 0.1	15.4 ± 0.4	18.3 ± 0.4
15-30	13.1 ± 1.2	17.4 ± 2.6	14.2 ± 0.4

Within rows, different letters indicate statistically significant differences (Bonferroni test, $p < 0.05$).

compacted soils in this and other dry areas, which are usually attributed to decreased soil physical quality (*e.g.* Govaerts *et al.*, 2006; Virto *et al.*, 2007; Fernández-Ugalde *et al.*, 2009). However, correlation analyses showed that greater PR values were correlated to earthworm activity, AWC and some enzyme activities (Table 5), which means that the observed compaction under PGC was not important enough to affect the soil biological functioning by creating a different soil porosity or smaller soil AWC overall. This suggests also no major differences in the topsoil physical condition for grapevine roots, which can in fact grow as deep as 0.9 m (Morlat & Jacquet, 2003). It is also important to consider that despite PR values greater than 2 MPa being considered as limiting for root development (Carter, 2002), it is known that in non-tilled soils plant roots can grow within the newly formed bio-pores and cracks in the soil (Ehlers *et al.*, 1983).

Aggregate stability (Table 2), which is the result of the equilibrium reached within soil aggregates between the binding agents and the applied disruption forces, is a dynamic indicator of the soil physical status, and has been shown to be useful to assess soil quality in relation to management in semi-arid land (Virto *et al.*, 2007; Álvaro-Fuentes *et al.*, 2008). In the studied soil, values of WSA were significantly smaller in TILL in the topmost soil layer (0-5 and 5-15, Table 2). Similar results were observed in a study in a close area (Peregrina *et al.*, 2010), and associated to less topsoil resistance to physical disruption.

Table 3. Concentration and stock of total C (SOC), C in the particulate organic matter (POM-C) and C in the microbial biomass in TILL (no PGC), and after one (ONEYR) and five (FIVEYR) years of PGC (mean ± standard error)

	TILL	ONEYR	FIVEYR
Organic C (SOC, mg C g^{-1} soil)			
0-5	9.15 ± 0.5 a	15.7 ± 0.8 b	12.5 ± 1.1 ab
5-15	8.92 ± 0.5	11.1 ± 0.5	10.0 ± 0.2
15-30	8.81 ± 0.5	9.33 ± 0.7	9.40 ± 0.7
POM-C (mg C-POM g^{-1} soil)			
0-5	1.13 ± 0.74 a	5.17 ± 3.75 b	3.58 ± 1.49 b
5-15	0.75 ± 1.46 a	2.96 ± 3.79 b	1.62 ± 1.89 ab
15-30	0.62 ± 1.36	1.29 ± 2.85	1.17 ± 1.77
Microbial biomass C (mg C kg^{-1} soil)	57.8 ± 1.8 a	103.2 ± 3.5 b	98.0 ± 1.0 b

Within rows, different letters indicate statistically significant differences (Bonferroni test, $p < 0.05$).

Table 4. Enzyme activities and metabolic diversity in the topsoil (0-5 cm) and earthworms activity (0-20 cm) in TILL (no PGC), and after one (ONEYR) and five (FIVEYR) years of PGC (mean ± standard error)

	TILL	ONEYR	FIVEYR
Enzyme activities (0-5 cm)			
Dehydrogenase (μg INTF g^{-1} soil h^{-1})	20 ± 1.2 a	47 ± 3.5 b	49 ± 3.0 b
β-glucosidase (μg 4-NP g^{-1} soil h^{-1})	71 ± 2.1 a	208 ± 2.0 b	273 ± 2.3 b
Urease (μg N-NH_4^+ g^{-1} soil h^{-1})	33 ± 2.6	61 ± 7.4	45 ± 7.0
Alkaline phosphatase (μg 4-NP g^{-1} soil h^{-1})	108 ± 4.0 a	302 ± 2.1 b	291 ± 11.0 b
Arylsulphatase (μg 4-NP g^{-1} soil h^{-1})	13 ± 0.9 a	41 ± 4.9 b	52 ± 6.0 b
Metabolic diversity (0-5 cm)			
Number of substrates used (NSU)[1]	16 ± 2.0	15.5 ± 0.5	24.5 ± 2.5
Earthworms (0-20 cm)			
Individuals m^{-2}	0.00 ± 0.0 a	50.0 ± 0.0 a	150.0 ± 34 b

Within rows, different letters indicate statistically significant differences (Bonferroni test, $p < 0.05$).
[1] NSU calculated from absorbance data after 54 h of incubation in a Biolog Ecoplate™ (Zak *et al.*, 1994).

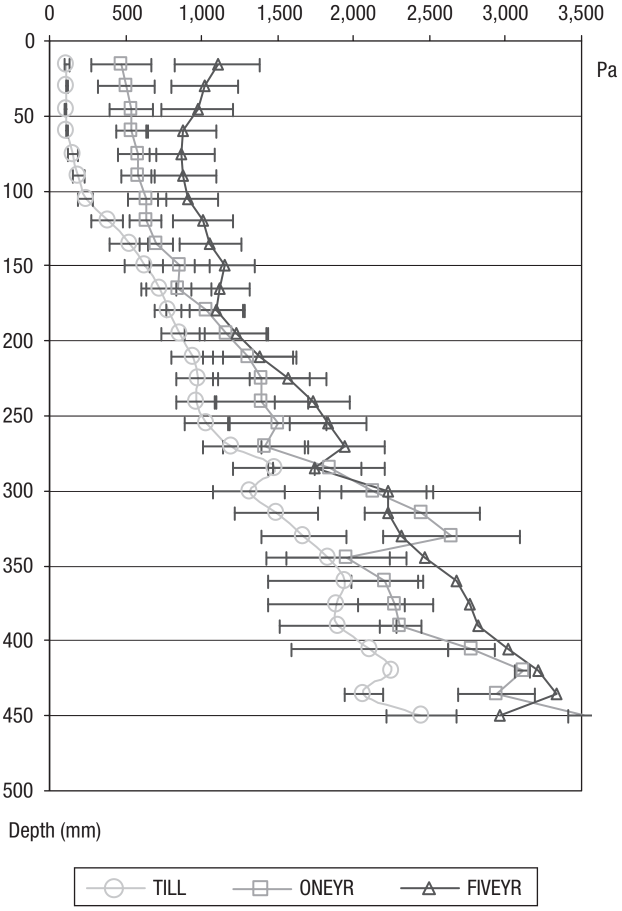

Figure 1. Penetration resistance profiles in TILL (no PGC), and after one (ONEYR) and five (FIVEYR) years of PGC (mean ± standard error). For each depth**,** asterisks indicate significant differences (Bonferroni test, $p < 0.05$).

The study of the correlations between WSA and the other studied soil parameters in 0-5 (Table 5) allows understanding the relationship between the studied physical and biological soil properties in TILL and in ONEYR and FIVEYR.

In this soil, aggregate stability was not correlated with SOC or POM-C, as observed in similar studies and other soil types. This can be explained considering two factors: First, the fact that differences in WSA did not coincide with differences in SOC and POM-C (while the latter was greater in ONEYR and FIVEYR than in TILL, WSA was different form TILL only in FIVEYR, Tables 1 and 2), which suggests a different pace of changes in both parameters. Second, in carbonate-rich soils, as the one in this study, it is known that the dependency of aggregation on organic matter is smaller than in soils without carbonates (Fernández-Ugalde *et al.*, 2011).

Aggregate stability was however correlated to other physical and biological parameters, such as AWC, microbial parameters (biomass, diversity and most enzyme activities), and earthworms density. This means that the existence of a more stable structure with PGC in this soil was related to the major differences observed in the soil under PGC in comparison to TILL.

The correlation of WSA with the studied microbiological parameters suggests a direct interaction between the microbial populations of the soil and the development of a stable structure in surface, and vice-versa (a well developed structure can favour the conditions for

Table 5. Correlation among the studied parameters in 0-5 cm

	SOC	POM-C	ρb	WSA	PR	AWC	Mic. C	E-worms	NSU	DH-ase	β-gluc.	Urease	P-ase	Aryl-S
SOC	1													
POM-C	0.93*	1												
ρb	0.27	0.45	1											
WSA	0.56	0.55	0.62	1										
PR	0.53	0.48	0.40	0.79*	1									
AWC	0.40	0.46	0.67*	0.91*	0.67*	1								
Microbial-C	0.68*	0.79*	0.41	0.62*	0.40	0.56	1							
Earthworms	0.46	0.40	0.58	0.91*	0.85*	0.85*	0.30	1						
NSU	0.04	0.11	0.56	0.79*	0.60	0.89*	0.24	0.84*	1					
Dehydrogenase	0.83*	0.85*	0.70*	0.83*	0.73*	0.77*	0.71*	0.77*	0.51	1				
β-glucosidase	0.64	0.67*	0.65	0.72*	0.83*	0.75*	0.59*	0.73*	0.57	0.90*	1			
Urease	0.30	0.76*	0.38	0.11	–0.04	0.11	0.72*	–0.10	–0.18	0.53	0.41	1		
Phosphatase	0.84*	0.89*	0.56	0.71*	0.77*	0.66	0.75*	0.64	0.38	0.94*	0.93*	0.57	1	
Arylsulpahtase	0.58	0.66*	0.68*	0.77*	0.83*	0.82*	0.61*	0.77*	0.65	0.89*	0.99*	0.37	0.91*	1

Values marked with * are significantly correlated (Pearson's correlation, $p < 0.05$).

microbial growth). Greater microbial densities under PGC (Table 3) can be in this sense attributed to greater SOC availability, and to reduced moisture stress, as reported by Whitelaw-Weckert *et al.* (2007) for different vineyard soils.

The study of the soil biological properties supported this statement. Dehydrogenase, β-glucosidase, alkaline phosphatase and arylsulphatase were 140, 239, 174 and 258% greater in average, respectively, in ONEYR and FIVEYR than in TILL (Table 4). All enzyme activities were significantly and positively correlated to microbial biomass and POM-C (Table 5). In the case of dehydrogenase, which is found only in living cells, its activity in soils is considered the expression of the total oxidative activity of the soil microflora and therefore of the metabolic activity of soil (Nannipieri *et al.*, 2002; Shaw & Burns, 2008). This enzyme has been determined to be the most suitable indicator to predict organic C accumulation in soils under organic management in a Mediterranean environment (Lagomarsino *et al.*, 2009b). In this study, it was in fact correlated to all organic, physical and biological indicators except NSU and urease activity (Table 5). This means that it can be associated to active organic matter metabolization, as observed also in other Mediterranean soils following the implementation of permanent soil cover (Lagomarsino *et al.*, 2009a; Moreno *et al.*, 2009). Increased β-glucosidase activity, which was correlated to POM-C, aggregation, AWC, and earthworm density (Table 5), has also been reported as associated to PGC in olive grooves in semi-arid SE Spain (Moreno *et al.*, 2009).

The correlation between earthworm density, WSA, AWC and biological diversity and enzymatic activity also indicates an association between earthworms and a stable soil structure. This is in line with the observation of Peres *et al.* (1998), who related improved soil structure in vineyard soils to higher levels of earthworm species diversity and distribution and to increased organic inputs. The possible role of earthworms in moving microorganisms up to the surface from deeper soil layers, and their potential contribution to increased microbial activity in the topsoil in agricultural soils, as observed in this study, has also been suggested by Amador & Görres (2007).

Finally, the soil available water capacity (AWC) was correlated to aggregation (WSA) and to the biological soil parameters (Table 5), in agreement with observations in other studies in vineyard soils. For instance, Steenwerth & Belina (2008a,b) found that permanent plant cover in California resulted in more available water in the soil after nine years. Morlat & Jacquet (2003) also observed greater AWC in a loam clay soil in the Loire Valley (France) following 17 years of permanent grass cover compared to bare soil. Steenwerth & Belina (2008a,b) associated the observed greater volumes of plant-available water with PGC to increased microbial activity and C and N dynamics, in agreement with the results in this study. The fact that similar studies (Klik *et al.*, 1998) in vineyards in more humid areas like Vienna (Austria) also found better physical soil conditions under PGC than with bare soil, but not significant differences in the microbial biomass, suggests the existence of a stronger relationship between more abundant and diverse microbial populations and improved soil structure and more available water in semi-arid soils than in other conditions.

Soil quality evaluation and indicators

One of the aims of this study was to assess the validity of the selected physical and biological attributes as soil quality indicators, in order to identify the most adequate soil attributes for the evaluation of vineyard soil management in semi-arid land. Ideal soil quality indicators must relate to ecosystem functions, be sensitive to variations in management, and integrate soil physical, chemical and biological properties and processes (Doran & Parkin, 1994). In this study, PCA in the upper soil layer was used with the aim of identifying (i) those soil attributes showing the greatest sensitivity to changes in soil functioning (Andrews *et al.*, 2004), and (ii) those explaining the highest proportion of the observed variability in the soil system in surface as a consequence of the implementation of PGC (Andrews *et al.*, 2002). Principal component analysis was run using data on aggregate stability, AWC, SOC, POM-C, microbial biomass C, enzyme activities except urease, and earthworms abundance in 0-5, as these were the soil properties displaying differences among treatments in 0-5. PCA resulted in only two factors (PCA-F1 and PCA-F2) with eigenvalue > 1. Table 6 summarizes the loading of each selected soil attribute in each factor, and the average scores of PCA-F1 and PCA-F2 for TILL, ONEYR and FIVEYR. PCA-F1 received the highest positive loadings from earthworm density and the physical attributes included for the PCA (WSA, AWC), followed by β-glucosidase and arylsulfatase. PCA-F2 received the highest loadings from the soil organic attributes (SOC, POM-C, microbial biomass) and from the activity of dehydrogenase and alkaline phosphatase.

Selection of soil quality indicators

Among the soil properties included in the PCA, those displaying the greatest loadings for PCA-F1 and PCA-F2 can be considered the ones better explaining the variability induced by PGC implementation in this soil (Andrews & Carroll, 2001; Andrews *et al.*, 2002). These properties were earthworm density and POM-C, for PCA-F1 and PCA-F2, respectively (Table 6). Following the approach of Andrews & Carroll (2001), the variables with loadings within 10% of those with the highest loadings can be considered as the ones best representing the system attributes, and would be selected as the most sensitive topsoil quality indicators for the studied soil. Such variables were WSA and AWC in PCA-F1, and SOC in PCA-F2 (Table 6). These results are in agreement with those in previous studies in the area in non-tilled soils (Bescansa *et al.*, 2006; Virto *et al.*, 2007; Imaz *et al.*, 2010) and soils under PGCs (Peregrina *et al.*, 2010), and are a good example of the need for evaluating both the soil physical and biological attributes to assess overall soil quality in vineyards in relation to changes in soil management.

Table 6. Proportion of variance explained using varimax rotation for each of the factors with eigenvalue > 1 (PCA-F1 and PCA-F2) in the 0-5 cm depth, and scores of PCA-F1 and PCA-F2 for TILL (no PGC), ONEYR and FIVEYR

	PCA-F1	PCA-F2
Eigenvalue	**7.565**	**1.352**
Loadings		
WSA	**0.868**	0.360
AWC	**0.914**	0.246
SOC	0.229	**0.903**
POM-C	0.235	**0.953**
Microbial biomass	0.278	0.800
Dehydrogenase	0.668	0.719
β-glucosidase	0.720	0.584
Alkaline phosphatase	0.546	0.813
Arylsulphatase	0.781	0.530
Earthworms	**0.943**	0.166
Scores		
TILL	–0.871 a	–0.986 a
ONEYR	–0.288 a	1.120 c
FIVEYR	1.160 b	–0.134 b

For scores, different letters indicate statistically significant differences (Bonferroni test, $p < 0.05$), for each factor. Bold figures indicate the greatest loadings for each factor.

Sensitivity to changes

Assuming initial homogeneity of the soil properties in the three plots, so that the situation of the soil in ONEYR and FIVEYR before PGC seeding would have been similar to the present situation in TILL, the effect of the implementation of PGC on the top layer (0-5) can be analyzed in relation to the time since PGC were sown.

Following this approach, differences observed among treatments in the scores for PCA-F1 (receiving greater loadings from physical properties) and PCA-F2 (receiving greater loadings from organic and biological soil attributes, Table 6) can be used to evaluate the sensitivity of these two types of soil attributes to changes when PGC are introduced, as in Imaz *et al.* (2010) for no-tilled soils. The scores for PCA-F1 were similar for TILL and ONEYR and significantly greater for FIVEYR (Table 6). TILL had also the lowest score for PCA-F2, followed by FIVEYR and then ONEYR, which displayed the maximum score for this factor (Table 6). This indicates that, in general, changes in the topsoil physical status appeared only after five years under PGC (FIVEYR), while biological indicators overall were different from TILL already in ONEYR. Differences were however observed among biological parameters. For instance, the soil enzyme activities and microbial biomass increased already in ONEYR in relation to TILL, but were similar in FIVEYR than in ONEYR (Tables 3 and 4). Earthworm abundance, which was grouped with the soil physical attributes in PCA-F1, and thus more sensitive to the soil physical status than to organic or biological soil characteristics, was similar in ONEYR and TILL, and changed in FIVEYR (Table 4). This is another evidence of the need for evaluating both the soil physical and biological attributes when studying changes in vineyard soil as a consequence of changes in management, as indicated by Doran & Parkin (1994) and Karlen *et al.* (2003).

Finally, if one considers soil quality an adequate tool for the evaluation of the sustainability of land management (Herrick, 2000), and accepts that its assessment must be approached considering both the ecosystem characteristics and the primary purpose for which the evaluation is being made (Karlen & Stott, 1994), grape yields and quality should also be considered to determine the net gain in soil quality of vineyard soils when PGC is implemented. The study of the relationship between soil quality, vineyard yield and grapes characteristics in semi-arid land is beyond the objectives of this study, but the tradeoffs between soil conservation and production need to be accounted for, as recently indicated by Ruiz-Colmenero *et al.* (2011) for semi-arid vineyards in steep land.

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