Soil dehydrogenase activity and organic carbon as affected by management system

Mia BRKLJAČA¹ (⊠) Kristina KULIŠIĆ¹ C. Brannon ANDERSEN²

Summary

Tillage and agrochemicals negatively affect soil organic matter (SOM) content and microbial activity. Intense cultivation of the Nadin valley began in the 1950s after hydro melioration. Our objective was to assess the microbial activity and SOM and to propose the improvements in soil management. Samples were collected according to randomized stratified design from organic (O) and conventional (C) plowed agricultural soils, from natural grass vegetation soils (G) and from abandoned vineyard soils (A). Dehydrogenases activity (DHA) and soil moisture were analyzed at 6 cm increments and C (total, organic and inorganic) and total N were analyzed at 2 cm increments, both to the depth of 18 cm. DHA was higher in G and in A than in C or O. DHA decreased with depth from 0-6 cm to 6-12 cm and 12-18 cm depth. Soil water content was higher in O than in C or G. Soils contained 67 ± 16 mg/g total carbon (organic + carbonate), 17 ± 8.0 mg/g organic carbon, and 1.5 ± 0.49 mg/g total soil nitrogen (organic and inorganic). Soils were carbonate-rich with $42\pm13\%$ CaCO₃. C:N ratio was approximately 12:1. Soil organic carbon (SOC) and total nitrogen (TN) positively correlated to DHA, regardless of the soil management. Principal component analysis showed soil grouping based mainly on the position in landscape and not based on management. Results showed that higher inputs of organic matter in agricultural soils would increase enzymatic activity, and shallow chisel plowing conserves soil water content.

Key words

C:N, conventional management, grass vegetation, microbial activity, organic management

Corresponding author: miabrkljaca@gmail.com

Received: February 01, 2019 | Accepted: April 18, 2019

¹Department of Ecology, Agronomy and Aquaculture, University of Zadar, Trg kneza Višeslava 9, Zadar, Croatia

² Department of Earth and Environmental Sciences, Furman University, Greenville, SC USA

Introduction

The changes of soil usage from natural vegetation and pastures to agricultural soil lead to decrease in organic matter content via soil erosion and oxidation of soil organic matter (Six et al, 1999; Lal, 2004; Stockmann et al., 2013). Soil management strategy (e.g., no till farming) has been shown to prevent loss and increase soil organic matter (SOM) content in agricultural soils (Montgomery, 2007; Blanco-Canqui and Lal, 2011). Agricultural methods that increase SOM content include irrigation, conversion from cultivated land to pastures, introduction of earthworms, changes in grazing management, fertilization, sowing grasses and legumes as cover crops, and conversion native land into pastures (Conant et al., 2001), conservation tillage and crop residue incorporation (Reeves, 1997; Dikgwatlhe et al., 2014). Losses of soil organic carbon (SOC) due to cultivation depend on climate and soil physical properties (Burke et al., 1989). SOC strongly impacts soil quality, functionality and health, where soil quality is linked to what soil does and soil health treats soil as a living biological entity (Lal, 2016).

Soil enzymatic activity is an estimate of soil microbial activity. Soil enzymatic activity is often assessed because of simplicity of measurement and its quick response to management practices (Okur et al., 2009). Soil dehydrogenases activity (DHA), phosphatase, protease and urease are often determined, while DHA is used as an index of activity of soil microorganisms (García-Orenes et al., 2016).

SOC and TN strongly correlate (Cheng et al., 2016). Highest fraction of SOC and TN found inside the macroaggregates, which are the products of microbial activity that contributes to the binding microaggregates into the macroaggregates (Cambardella & Elliott, 1994).

The SOC and TN variations are correlated to C returned to the soil (Cheng et al., 2016; Havlin et al., 1990; Mazzoncini et al., 2011) and are conserved or increased with reduced tillage (Havlin et al., 1990; Mazzoncini et al., 2011) or stabilized in vegetative filter strips management system compared to no till (Veum et al., 2011).

The potential decomposition rate of organic C and potential mineralizable N is higher in soils with high input of organic matter (Cheng et al., 2016). As the effect of climate changes SOC and total N stocks increase at 0-20 cm soil depth with the increase in total nitrogen in excess to organic carbon is potentially caused by the N fixation on clay minerals or changes in microbial populations (Schrumpf et al., 2014). Our objective was to assess how vineyard soil management affects microbial activity, soil organic carbon and total nitrogen. A better understanding of the link between management and microbial activity could lead to improvements of vineyard soil management in Mediterranean part of Croatia.

Materials and Methods

Study Area

The samples were collected in Mediterranean part of Croatia, in Nadin Valley (44.044 N, 15.49 E). All soils were classified as Calcic Anthrosol (Bogunović et al., 1998). The soil properties of the area of Vransko Basin (mean \pm standard error) are: 8.02 ± 0.055 pH in H₂O, 28.0 \pm 2.6% CaCO₃, 3.97 \pm 0.31% humus, 0.23 \pm 0.013 mg 100 g⁻¹ N, 11.6 \pm 1.0 mg 100 g⁻¹ P₂O₅ and 47.6 \pm 3.5 mg 100 g⁻¹ K₂O (Romic et al., 2009). In the past, the valley was flooded regularly during winter seasons with occasional floods through the whole year. In 1950s the valley was hydro-meliorated by building the bulwark around the valley and the network of open channels. Water was drawn by the channels to the southern part of the valley where was pumped out of the valley. The valley is subdivided into individually owned and managed (e.g., conventional, organic, fallow) plots of approximately 2 ha. Most of the valley area was planted with grapevine. Vineyards fertilized with organic or mineral fertilizers and vineyards of different age are spread irregularly in the area. The climate of Nadin is warm and temperate, classified as Cfa by Köppen and Geiger, with average temperature of 13.3°C and about 979 mm of rainfall annually (climate-data.org).

Vineyards management

Organic vineyards (O) soil was managed without vegetation cover during growing season by shallow chisel plowing at least three times a year. Fertilization consisted of yearly spring application of 2 tones/ha of dry pelleted manure with incorporation of pruned vines. No cover crops were planted during the winter. Chemical fertilizer and herbicides were not used, but copper sulfate was used as a fungicide at 1/3 the normal amounts.

Conventional vineyards (C) were once a year moldboard plowed and herbicides were applied. Fertilization was done with NPK fertilizers. Full plant protection program was applied with the use of copper sulfate and no cover crops were planted.

Soils under the natural grass (G) were not plowed, mowed nor copper sulfate was used and they were located in the vicinity of sampled vineyards. The abandoned vineyards (A) were under the conventional production but for more than 10 years without any agricultural activity.

Sampling method

Soil profiles were collected at 14 locations in May 2017 and 17 locations in May 2018 using a stratified random sampling method. The vineyards were randomly selected within sections edged with drainage channels. Each sample location was chosen by random selection of the row and the vine in a selected vineyard.

Soil samples were collected using an 18 cm soil profiler and recovered soil profiles ranged from 12 cm to 18 cm deep. The soil profile was divided in 2 cm layers for C and N analysis (6 to 9 samples per profile, n=250) and in 6 cm layers for dehydrogenases analysis (2 to 3 samples per profile, n=82). At the site of the soil profiling, a soil core was collected for determinations of bulk density, soil water content and particle size analysis. Soil samples for dehydrogenases activity analysis were put in polyethylene zip bags and immediately transferred to the refrigerator and kept at 4°C until measurement for maximum of 14 days of storage. Soil samples for C and N analysis were air dried and stored at room temperature in polyethylene zip bags until analysis.

Weather at the time of sampling was hot and dry in 2017, while it was rainy and warm in 2018. At each sample location, crop type, organic or mineral fertilizers, and state of land cover was recorded.

Dehydrogenases activity

Dehydrogenases activity (DHA) was determined according to the modified method reported by Wolinska et al. (2013). One g of wet soil was transferred to cuvette and incubated with 20 mg of $CaCO_3$, 0.5 ml of 1% glucose and 0.2 ml of 3% triphenyl tetrazolium chloride (TTC) at 30°C for 20 hours. The triphenyl formazan (TPF) formed from TTC by DHA was extracted by adding 5 ml of ethanol. After shaking vigorously for 30 s, the suspension was filtered and diluted to 10 ml with ethanol. TPF was measured spectrophotometrically at 485 nm. Data were expressed on an oven-dry soil basis (105°C).

Soil carbon and nitrogen content

Samples for analysis of soil carbon (organic and inorganic) and total soil nitrogen (organic and inorganic) content were sterilized at 120°C upon arrival in South Carolina. Samples were lightly disaggregated and passed through a 2 mm sieve to remove roots and rock fragments (e.g., Conant et al., 2003; Blanco-Canqui and Lal 2008; Dorji et al., 2014). The <2 mm fraction for each sample was ball-milled and dried at 105°C for 24 hours and cooled in a desiccator. Carbon and nitrogen content of dried samples were measured using a LECO TruMac Series 4000 C/N Macrodeterminator. Samples were combusted at 1350°C for analysis. C and N contents were measured in samples from 14 profiles collected in 2017 (n=122) and 17 profiles collected in 2018 (n=128). After initial analysis for total soil carbon and total soil nitrogen, samples were treated with an acid fumigation method modified from Ramnarine et al. (2011) to remove soil inorganic carbon in the form of carbonate minerals. The carbon content of acid fumigated samples were measured to determine soil organic carbon content. Carbonate carbon was different than total soil carbon and soil organic carbon.

Soil water content and soil physical properties

Soil water content was determined within 24 h of sampling. Soil was dried in ventilated drying oven for 48 h at 105°C and soil water content (w) was calculated as: w = (m - d) / d, where m is the moist soil mass prior to drying, and d is the dry mass of the same soil after drying. Clay content, particle density, bulk density and porosity was determined in all 17 samples from 2018. Clay content was determined by pipette method after aggregate disintegration with Na-pyrophosphate, particle density was determined using pycnometer, bulk density was determined using the core of 100 cm³, and porosity was determined by calculation using particle density and bulk density (Škorić, 1982).

Statistical analysis

Two-way ANOVA was used to analyze the effect of management system and soil depth as independent factors on the dehydrogenase activity and soil water content. Dehydrogenase activity data were log transformed to fulfill the homogeneity of variance assumption. The relationships between the soil variables parameters were determined by correlations (Pearson's correlation coefficient). The difference between grass and vineyard soils was determined by a t-test. The Shapiro-Wilk method was used to test for normality, and if the normality test failed the Mann-Whitney rank sum test was used to test the difference between populations. Soil carbon and nitrogen data were averaged in 6 cm increments for comparison with DHA and water content. The relations between the soil variables, soil locations and soil management were analyzed by Principal component analysis. Statistical analysis was performed with Statistica 7 (StatSoft Inc. 2004) or SigmaPlot v. 14 (Systat Software Inc. 2017).

Results

Natural vegetation soil and cultivated soils

In 2017 DHA was higher in soil in G (25.66 \pm 5.0 nmol g⁻¹ h⁻¹ TPF) than in C or O (11.2 \pm 1.0 and 9.12 \pm 0.96 nmol g⁻¹ h⁻¹ TPF, respectively) (Table 1). The DHA decreased with depth but the difference wasn't significant due to high sample variability (Table 2).

In Mediterranean climates, the conservation of soil water has the key role. Soil water content in 2017 was higher in O (23.8 \pm 1.5%) than in C or G soil (16.8 \pm 1.7 and 15.7 \pm 1.6%, respectively) (Table 1). Soil water content was higher at 6-12 cm and 12-18 cm depths (20.5 \pm 2.0% and 20.0 \pm 1.8%, respectively) than at 0-6 cm depth (14.5 \pm 1.3%) (Table 2).

Across all land covers, total carbon concentration was 65 ± 18 mg g⁻¹, organic carbon concentration was 18 ± 8.3 mg g⁻¹, and total soil nitrogen concentration was 1.4 ± 0.44 mg g⁻¹. The C:N

Table 1. Soil dehydrogenases activity expressed as TPF and soil water content under the different soil managements, Nadin Valley in 2017 and2018, mean \pm standard error. Different letters indicate statistical significance at level of 0.05 within each factor.

Factor	Level	TPF (nmol g ⁻¹ h ⁻¹)	Water content (% w/w)	Level	TPF (nmol g ⁻¹ h ⁻¹)	Water content (% w/w)
	2017	2017	2017	2018	2018	2018
Management	Natural	$25.66\pm5.0^{\rm a}$	15.7 ± 1.6^{b}	Abandoned	$34.8\pm6.9^{\rm a}$	25.1 ± 2.2^{ab}
	Conventional	$11.2\pm1.0^{\rm b}$	$16.8 \pm 1.7^{\text{b}}$	Conventional	$22.3\pm2.7^{\rm b}$	$26.8\pm0.8^{\text{a}}$
	Organic	$9.12\pm0.96^{\rm b}$	$23.8\pm1.5^{\text{a}}$	Organic	$19.4\pm2.8^{\rm b}$	$21.3\pm0.5^{\rm b}$
Significance		0.00691	0.00164		0.0127	0.0135

ratio was 14.4:1. Soils were carbonate-rich with $39\pm12\%$ CaCO₃. Correlation analysis (n=33) showed that both soil organic carbon (r=0.787, p<0.000001) and soil nitrogen (r=0.757, p<0.000001) were significantly correlated with DHA but not with soil water content (Figure 1 and Table 3).

Table 2. DHA, SOC, TN and soil water content at three soil depths at Nadin Valley in 2017 and 2018, mean \pm standard error. Different letters indicate statistical significance at level of 0.05 within each factor.

	DHA	SOC	TN	Water
Depth	2017			
0-6 cm	21.2 ± 5.7	14.3±0.55	1.33±0.096	14.5±1.3 ^b
6-12 cm	16.0 ± 2.8	13.8±0.84	1.30±0.10	20.5±2.0ª
12-18 cm	13.2 ± 3.8	13.2±0.44	1.24±0.073	20±1.6ª
		20	18	
0-6 cm	37.4±4.7ª	22.4±3.3	2.02±0.19 ^a	28±1.6
6-12 cm	21.1±3.0 ^b	16.4±2.8	1.53±0.17 ^b	25.1±0.9
12-18 cm	14.8 ± 2.4^{b}	16±3.7	1.41±0.21 ^b	22.7±0.9
Significance				
2017	0.49	0.636	0.145	0.0108
2018	0.000458	0.324	0.00728	0.0545

DHA TPF nmol g⁻¹ h⁻¹, SOC g kg⁻¹, TN g kg⁻¹, water % w/w.

Table 3. Pearson product moment correlation between SOC, TN, DHA and water for each year 2017 and 2018 and combined (2017/2018); the outliers from 2018 were excluded in correlation coefficient of DHA and SOC.

		SON	Water	DHA
SOC	2017/2018	0.829***	0.278	0.709***
	2017	0.978***	-0.030	0.857***
	2018	0.735***	0.564**	0.876***
TN	2017/2018		0.237	0.647***
	2017		-0.00767	0.896***
	2018		0.322	0.667***
Water	2017/2018			0.609***
	2017			-0.0353
	2018			0.736***

DHA TPF nmol g $^{-1}$ h $^{-1}$, SOC g kg $^{-1}$, TN g kg $^{-1}$, water % w/w.



Figure 1. Pearson product moment correlation between DHA and SOC and between DHA and TN in 2017 and 2018; the outliers from 2018 were excluded in correlation coefficient of DHA and SOC.

Abandoned and cultivated vineyards soils

In 2018 we compared DHA of A vineyards with C and O soils (Table 1). The goal was to see how abandonment affects microbial activity and whether soil physical properties affect DHA. The C and O soils were similar in DHA (22.3 ± 2.7 and 19.4 ± 2.8 nmol g⁻¹ h⁻¹ TPF, respectively) and lower than DHA in soils of A (34.8 ± 6.9 nmol g⁻¹ h⁻¹ TPF). DHA decreased with soil depth, TPF content was higher at 0-6 cm depth (37.4 ± 4.7 nmol g⁻¹ h⁻¹) than at 6-12 cm and 12-18 cm depths (21.1 ± 3.0 and 14.8 ± 2.4 nmol g⁻¹ h⁻¹, respectively) (Table 2).

Soil water content in 2018 was higher in C soil ($26.8 \pm 0.8\%$) than in O soil ($21.3 \pm 0.5\%$) while A soil water content was similar to both ($25.1 \pm 2.2\%$) (Table 1). Soil water content decreased with depth with significance of p = 0.0545 (Table 2).

The correlations between soil bulk density, particle density, clay content, water content, porosity and DHA in 2018 were presented in Table 4. DHA was correlated only with water content (0.69). The other significant correlations were found between water content and particle density (-0.50), bulk density strongly negatively correlated with porosity and clay content (correlation coefficient of -0.99 and -0.69, respectively) and porosity correlated positively with clay content (0.66).

Principal component analysis was used to group the samples from 2018 according to the relations between the analyzed variables, the spatial soil variability and soil management practices. First three principal components accounted for 89.8% of total variance (PC1 60.5%, PC2 16.0% and PC3 13.3%). The principal component 1 showed positive loadings on bulk density and negative loadings on porosity, clay content, DHA and water content (Table 5). Principal component 2 showed positive loading on particle density. On PCA diagram the samples were labeled with subscripts for sides of the valley (Figure 2). The four samples from the southeastern part of the valley which were O or A were grouped in upper right quadrant (Figure 2).

Table 4. The significant correlations between bulk density, particle density, porosity, water content and dehydrogenase activity (DHA), Nadin Valley, 2018, p < 0.05, n = 17.

		Bulk density	Particle density	Pore	osity	Clay cont	ent	DHA	Water	r content
Bulk den	sity	1.00	0.42	-0.9	9**	-0.69**	-	-0.47		0.22
Particle of	lensity		1.00	-0.	.26	-0.44		-0.42	-0	1.50**
Porosity				1.	00	0.66**		0.43	(0.14
Clay con	tent					1.00		0.33	(0.26
DHA								1.00	0.	.69**
Water co	ntent								1	1.00
3	35									
										1
	3.0							·		
2	2.5					Os	e			
2	2.0						•]
	-						Ose	e		1
1				0						1
2%	1.0			Csw						
0.9).5				nw					
ii ,					•	Com	A	se	an a	
ctor				Cnw	Cow	Cnarw		0		1
ē -0).5	Ar	Onw Oo	1	0	C	nw			
-1	1.0						Cnw	Ose		
							0			1
0	1.9									1
-2	2.0			1						
-2	2.5							<u> </u>		
				An early services of the ser						
	-6	-5 -4	-3	2 -	1	0 1		2 3	4	5
				Facto	or 1 80	45%				

Figure 2. Principal components plot that relates soil properties to the soil position and management. A: abandoned, C: conventional, O: organic, nw: nortwest, s: south, se: south east, sw: south west.

Table 5. F	actor	loadings,	correlation	between	original	variables	and
factors.							

	Principal component 1	Principal component 2
Bulk density	0.919	
Particle density		0.673
Porosity	-0.815	
Clay content	-0.647	
DHA	-0.744	
Water content	-0.887	

The samples from the northwestern (nw), southwestern (sw) and southern (s) part of the Nadin valley managed conventionally or abandoned were positioned in the center of the coordinate system. The two samples A and C from northwest part (Anw and Cnw) contributed less to PC1 than the rest of the sample from the same group. The position of organic samples from southeast (Ose) showed high contribution to the variables with respect to other soil environments indicating lower microbial activity caused by low water, low clay content and high bulk density.

Across both land uses, total carbon concentration was 69 ± 14 mg g⁻¹, organic carbon concentration was 16 ± 7.7 mg g⁻¹, and total soil nitrogen concentration was 1.5 ± 0.55 mg g⁻¹. Soils were carbonate-rich with $45\pm14\%$ CaCO₃. Carbonate content variation in 2018 was higher because of a single sample location with very low carbonate content ($10.3\pm0.908\%$). Most likely, this sample did not have all carbonate removed during fumigation. If the low carbonate sample is removed, soil organic carbon was 15 ± 5.7 mg g⁻¹, total soil nitrogen was 1.5 ± 0.56 mg g⁻¹, and carbonate content was $47\pm12\%$. The C:N ratio was 11.5:1 excluding the outlier sample location. Nine sample locations were analyzed for both soil carbon and nitrogen and DHA.

Correlation analysis (n=40) showed that both soil organic carbon (r=0.510, p<0.001) and soil nitrogen (r=0.668, p<0.001) were significantly correlated with DHA (Figure 1) and soil water content significantly correlated with organic carbon (0.541, p<0.001) and DHA (0.674, p<0.001) (Table 3). If the sample with low carbonate is removed from the analysis, correlation coefficients for between DHA and soil organic carbon and soil nitrogen were 0.862 and 0.668, respectively (p<0.001). DHA, soil organic carbon, and soil nitrogen decreased with depth. Abandoned land had significantly greater total soil nitrogen than cultivated land (p<0.001). Although the mean soil organic carbon was greater for abandoned land, the difference was not significant. However, sample size is small.

Discussion

Higher DHA observed in the soils under natural grass than in the vineyard soils managed conventionally and organically (by 2.3 and 2.8 times, respectively) was consistent with the literature. In Island, DHA was higher 1.2 and 1.3 times in uncultivated soil ($4.4 \ \mu g \ g^{-1} \ h^{-1}$) than in two cultivated soils (3.8 and 3.4 $\mu g \ g^{-1} \ h^{-1}$) (Guicharnaud et al., 2010). Likewise, Błońska et al. (2017a) found DHA was 4 to 7 times lower in tilled soil than in soils under natural vegetation. This is consistent with Steenwerth and Belina (2008) who showed that cover crops added soil organic matter to the soil and improved microbial activity in the vineyard soils in California. The DHA in both organic and conventional vineyards soils were similar, which was different than the previous studies. Okur et al. (2009) found higher DHA, protease, urease and alkyl phosphatase in organic than in conventional vineyard. Similarly, García-Orenes et al. (2016) found the soil enzymatic activity, organic C, total N and available P were higher in organic vineyard soils than in conventional vineyard soils. The soil treatments that include organic inputs improve SOC, dehydrogenase activity and nutrients availability (Adak et al., 2014). The soil enzymatic activity correlate to labile organic C rather than to SOC (Shao et al., 2015). The correlation of SOC and TN to DHA indicates that the large part of the soil organic matter consisted of easily decomposable organic material, consistent with recent interpretations of the composition of soil organic matter (Stockman et al., 2013). Our results confirm the relationship between DHA and SOC, but organic and conventional soil managements were not a factor. This could be the result of relatively low organic matter input into the soils of dry pelleted manure in O, along with both O and C being tilled compared to the inputs of 30 mg/ha farmyard manure or 10 mg/ha farmyard manure and green manure in the study of Okur et al. (2009) and the inputs of pruned material with 20 Mg/ ha sheep manure or pruned material with vetch as cover crops in the study of García-Orenes et al. (2016). Additionally, the differences in DHA between G and O were possibly a function of the differences in the quality of inputs between grass and pelleted composted manure. For similar material, Gale et al. (2006) found cumulative decomposition in first 20 to 30 days for composted dry manure of approximately 10% of total C and for fresh yard trimmings 20-30% of total C while after 70-days decomposition two organic materials were decomposed to the similar degree (29% and 24%, respectively). As in soils under the natural vegetation the abandoned vineyards were rich in grasses that probably decompose readily compared to the dry pelleted manure.

Similar, Bhatt et al. (2016) found the significance of the organic material quality for microbial activity in the long-term experiment with cropping rice-wheat. The enzymatic activity, populations of microorganisms and microbial biomass were the highest under NPK fertilization improved with farmyard manure of 15 t ha⁻¹ than under the mineral fertilization or control.

The results indicate that inputs of organic matter in the organic management system should be increased. Root turnover is the biggest source of organic matter in soils (Stockman et al., 2013), the vineyards are tilled, and cover crops are not planted, though some rows have natural grasses and weeds.

Abandoned plots have higher DHA, but there is no difference between O and C (though both do use copper sulfate). Abandoned plots do have higher soil nitrogen. The results showed that the soil recovered DHA after the abandoning agricultural management. Similar found Gispert et al. (2013) for abandoned soils, which had higher glucosidase, protease and urease activity than vineyard soils. But copper is retained in the soil and negatively affect DHA (Fernández-Calviño et al., 2010) though we don't know how much. The use of copper sulfate may inhibit development of fungi in the soil (e.g., boreal forests, Clemmenson et al. 2013) and thereby affect the ability to sequester soil organic carbon. Additionally, copper may interfere with TTC and result in lower TPF (Chander and Brookes, 1991). This would suggest that some combination of reduced fungicide/pesticide and chemical fertilizer use and lack of tilling allows soil microbial activity to recover. Since both conventional and organic use copper sulfate and are tilled, some combination of those two diminish microbial activity.

The observation of higher water content at 6-12 cm and 12-18 cm depth than at surface depth of 0-6 cm in dry 2017 but similar at three depths in wet 2018 was expected since the soil sampling occurred in May 2017, after the intercept of the dry season. The soil water content higher in organic vineyard than in conventional vineyard was attributed to the shallow tillage, a kind of conservation tillage, which broke the surface soil cracks. Similarly, according to Busari et al. (2015) soils under minimum tillage or zero tillage have higher water flow than soils under conventional tillage. The observed lower soil water content at the G than at C was attributed to the formation of soil cracks and absence of tillage. That was in accordance with Cucci et al. (2016) who found that in hot dry climate the water holding capacity was higher in tilled than in no-till soil. Higher DHA and lower water content of the soil at G than at O and C supports the findings of Paradelo and Barral (2009) that the microbial enzymatic activity is 40% of maximum value even in air-dry soil.

G soils have slightly, but not significantly, higher SOC and significantly higher TN. It might be that lack of disturbance was the cause which would suggest that no-till approaches with groundcover might be better soil management. The vegetation is an important factor in soil enzymatic activity (Błońska et al., 2017b) and in the spatial-temporal variability of the soil labile organic C content (Shao et al., 2015).

Ratios of DHA and SOC content in surface layer to deeper layer of both years showed that both SOC and DHA were the highest in upper 6 cm of soil even in plowed soils. That was explained by the position of the sampling spot being in the line with vines that is not plowed.

Principal component analysis was able to differentiate soils with different cropping and sampling times (Gispert et al., 2013). In this study the soils were grouped based mainly on the position in landscape and not based on management system.

Conclusion

DHA was higher in soils under the natural grass or in abandoned vineyards soils than in soils under organic (no use of herbicides and chemical fertilizers, but use of dry manure pellets and reduced copper sulfate) or conventional (use of herbicides, chemical fertilizers and pesticides) management systems. Dry pelleted manure may have been less decomposable compared to grass root inputs in soils under the natural grass and abandoned vineyards. Strong positive correlations of DHA with SOC and TN indicated that great part of soil organic matter was easily decomposable. In vineyard soils DHA could be increased with the change in materials incorporated and use of the cover crops such as legumes. Shallow tillage was important for soil water preservation and DHA positively correlated with soil water content, soil organic carbon, and soil nitrogen. The reduction of tillage and planting cover crops was important for DHA. The study should be continued with the experiments of types, amounts and timing of organic matter incorporation and the analysis on yield and quality of wine.

References

- Adak T., Singha A., Kumar K., Shukla S., Singh A., Kumar Singh V. (2014). Soil organic carbon, dehydrogenase activity, nutrient availability and leaf nutrient content as affected by organic and inorganic source of nutrient in mango orchard soil. J Soil Sci Plant Nutr (2): 394–406. DOI: 10.4067/S0718-95162014005000031
- Bauer A., Black A. L. (1994). Quantification of the effect of soil organic matter content on soil productivity. Soil Sci Soc Am J 58(1): 185–193. DOI: 10.2136/sssaj1994.03615995005800010027x
- Bhatt B., Chandra R., Ram S., Pareek N. (2016). Long-term effects of fertilization and manuring on productivity and soil biological properties under rice (Oryza sativa)–wheat (Triticum aestivum) sequence in Mollisols. Arch Agron Soil Sci 62(8): 1109–1122. DOI: 10.1080/03650340.2015.1125471
- Blanco-Canqui H., Lal R. (2008). No-tillage and soil-profile carbon sequestration: An on-farm assessment. Soil Sci Soc Am J 72: 693-701
- Błońska E., Lasota J., Zwydak M. (2017a). The relationship between soil properties, enzyme activity and land use. Forest Research Papers 78(1): 39–44. DOI: 10.1515/frp-2017-0004
- Błońska E., Lasota J., Gruba P. (2017b). Enzymatic activity and stabilization of organic matter in soil with different detritus inputs. Soil Sci Plant Nut 63(3): 242–247. DOI: 10.1080/00380768.2017.1326281
- Bogunović M., Vidaček Ž., Racz Z., Husnjak S., Špoljar, A., Sraka, M. (1998). Soil maps of Croatia - FAO UNESCO. Soil Science Department, Agricultural Faculty, University of Zagreb.
- Burke I. C., Yonker C. M., Parton W. J., Cole C. V., Schimel D. S., Flach K. (1989). Texture, climate, and cultivation effects on soil organic matter content in U.S. grassland soils. Soil Sci Soc Am J 53(3): 800–805. DOI: 10.2136/sssaj1989.03615995005300030029x
- Busari M. A., Kukal S. S., Kaur A., Bhatt R., Dulazi A. A. (2015). Conservation tillage impacts on soil, crop and the environment. International Soil and Water Conservation Research 3(2): 119–129. DOI: 10.1016/j.iswcr.2015.05.002
- Cambardella C. A., Elliott E. T. (1994). Carbon and nitrogen dynamics of soil organic matter fractions from cultivated grassland soils. Soil Sci Soc Am J 58: 123–130.
- Chander K., Brookes P. C. (1991). Is the dehydrogenase assay invalid as a method to estimate microbial activity in copper-contaminated soils? Soil Biol Biochem 23(10): 909–915. doi.org/10.1016/0038-0717(91)90170-O
- Cheng W., Padre A. T., Sato C., Shiono H., Hattori S., Kajihara A., ... Kumagai K. (2016). Changes in the soil C and N contents, C decomposition and N mineralization potentials in a rice paddy after long-term application of inorganic fertilizers and organic matter. Soil Sci Plant Nut 62(2): 212–219. doi.org/10.1080/00380768.2016.11551 69
- Clemmensen K. E., Bahr A., Ovaskainen O., Dahlberg A., Ekblad A., Wallander H., Stenlid J., Finlay R. D., Wardle D. A., Lindahl B. D. (2013). Roots and associated fungi drive long term carbon sequestration in boreal forest. Science 339: 1615-1618
- Conant R. T., Paustian K., Elliott E. T. (2001). Grassland management and conversion into grassland: effects on soil carbon. Ecol Appl 11(2): 343– 355. DOI: 10.1890/1051-0761(2001)011[0343:GMACIG]2.0.CO;2
- Conant R. T., Six J., Paustian K. (2003). Land use effects on soil carbon fractions in the southeastern United States. 1. Management intensive versus extensive grazing. Bio Fertil Soils 38: 386-392. DOI: 10.1007/ s00374-003-0652-z
- Cucci G., Lacolla G., Crecchio C., Pascazio S., De Giorgio D. (2016). Impact of long-term soil management practices on the fertility and weed flora of an almond orchard. Turk J Agric For 40: 194–202. DOI: 10.3906/tar-1502-87

- Dorji T., Odeh I. O. A., Field D. J. (2014) Vertical distribution of soil organic carbon density in relation to land use/cover, altitude and slope aspect in the eastern Himalayas. Land 3: 1232-1250. DOI: 10.3390/land3041232
- Dikgwatlhe S. B. et al. (2014). Changes in soil organic carbon and nitrogen as affected by tillage and residue management under wheat-maize cropping system in the North China Plain. Soil Till Res 144: 110–118. DOI: 10.1016/j.still.2014.07.014
- Fernández-Calviño D., Soler-Rovira P., Polo A., Díaz-Raviña M., Arias-Estévez M., Plaza C. (2010). Enzyme activities in vineyard soils longterm treated with copper-based fungicides. Soil Biol Biochem 42: 2119–2127. DOI: 10.1016/j.soilbio.2010.08.007
- Havlin J. L., Kissel D. E., Maddux L. D., Claassen M. M., Long J. H. (1990). Crop rotation and tillage effects on soil organic carbon and nitrogen. Soil Sci Soc Am J 54(2): 448–452.
- García-Orenes F., Roldán A., Morugán-Coronado A., Linares C., Cerdà A., Caravaca F. (2016). Organic fertilization in traditional Mediterranean grapevine orchards mediates changes in soil microbial community structure and enhances soil fertility. Land Degrad Dev 27(6): 1622-1628. D: 10.1002/ldr.2496
- Gispert M., Emran M., Pardini G., Doni S., Ceccanti B. (2013). The impact of land management and abandonment on soil enzymatic activity, glomalin content and aggregate stability. Geoderma 202–203: 51–61. DOI: 10.1016/j.geoderma.2013.03.012
- Guicharnaud R. A., Arnalds O., Paton G. I. (2010). The effect of season and management practices on soil microbial activities undergoing nitrogen treatments - Interpretation from microcosm to field scale. Iceland Agr Sci 23(1): 123–134
- Lal R. (2004). Soil carbon sequestration impacts on global climate change and food security. Science 304: 1623-1627
- Lal R. (2016). Soil health and carbon management. Food and Energy Security 5(4): 212-222. doi: 10.1002/fes3.96
- Mazzoncini M., Sapkota T. B., Bàrberi P., Antichi D., Risaliti R. (2011). Long-term effect of tillage, nitrogen fertilization and cover crops on soil organic carbon and total nitrogen content. Soil Till Res 114: 165– 174. doi.org/10.1016/j.still.2011.05.001
- Montgomery D. (2007). Is agriculture eroding civilization's foundation? GSA Today 17: 4-9. DOI: 10.1130/GSAT01710A
- Okur N., Altindisli A., Cengel M., Gocmez S., Kayikcioglu H. H. (2009). Microbial biomass and enzyme activity in vineyard soils under organic and conventional farming systems. Turk J Agric For 33: 413– 423. DOI: 10.3906/tar-0806-23

- Paradelo R., Barral M. T. (2009). Effect of moisture and disaggregation on the microbial activity of soil. Soil Till Res 104: 317–319. DOI: 10.1016/j.still.2009.02.010
- Ramnarine R., Voroney R. P., Wagner-Riddle C., Dunfield K. E. (2011). Carbonate removal by acid fumigation for measuring the d13C of soil organic carbon. Can J Soil Sci 91: 247-250.
- Reeves D. W. (1997). The role of soil organic matter in maintaining soil quality in continuous cropping systems. Soil Till Res 43: 131–167
- Romic D., Romic M., Bakic H. (2009). Soil quality and monitoring of surface waters in the Vransko lake watershed, I phase - Soil quality. Zagreb.
- Shao X., Yang W., Wu M. (2015). Seasonal dynamics of soil labile organic carbon and enzyme activities in relation to vegetation types in Hangzhou Bay tidal flat wetland. PLoS ONE 10(11): 1–15. DOI: 10.1371/journal.pone.0142677
- Schrumpf M., Kaiser K., Schulze E. (2014). Soil organic carbon and total nitrogen gains in an old growth deciduous forest in Germany. PLoS ONE, 9(2), e89364. doi.org/10.1371/journal.pone.0089364.t001
- Six J., Elliott E. T., Paustian K. (1999). Aggregate and soil organic matter dynamics under conventional and no-tillage systems. Soil Sci Soc Am J 63: 1350–1358
- Steenwerth K., Belina K. M. (2008). Cover crops enhance soil organic matter, carbon dynamics, and microbial function in a vineyard agroecosystem. Appl Soil Ecol 40: 359-369
- Stockmann U. Adams, M. A., Crawford, J. W., Field, D. J., Henakaarchchi, N., Jenkins, M., ... Zimmermann, M. (2013). The knowns, known unknowns and unknowns of sequestration of soil organic carbon. Agr Ecosyst Environ 164: 80–99. DOI: 10.1016/j.agee.2012.10.001
- Swędrzyńska D., Małecka-Jankowiak I. (2017). The impact of tillaging spring barley on selected chemical, microbiological, and enzymatic soil properties. Pol J Envon Stud 26(1): 303–313. DOI: 10.15244/ pjoes/64911
- Škorić A. (1982). Manual of pedological research. Faculty of agricultural sciences, University of Zagreb, Zagreb, Croatia (in Croatian)
- Veum K. S., Goyne K. W., Holan S. H., Motavalli P. P. (2011). Assessment of soil organic carbon and total nitrogen under conservation management practices in the Central Claypan Region, Missouri, USA. Geoderma, 167–168, 188–196. doi.org/10.1016/j.geoderma.2011.09.003
- Wolinska A., Stępniewska Z., Szymańska E. (2013). Dehydrogenase activity of soil microorganisms and the total DNA level in soil of different use. J Agric Sci Technol B 3: 613–622

acs84_18