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# ORIGINAL PAPER

# Vineyard soils under organic and conventional management—microbial biomass and activity indices and their relation to soil chemical properties

Björn Probst · Christian Schüler · Rainer Georg Joergensen

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Abstract Eight vineyards in Pfaffenheim (P) and Turckheim (T) close to Colmar, France, forming four pairs of organic and conventional vineyards, were analyzed for microbial biomass and activity indices in relation to important soil chemical properties (carbon, nutrient elements, heavy metals) and also to differences between the bottom and top positions on the vineyard slope. The question was whether the vineyard management affects especially the soil microbiological indices. Three locations were on limestone (P-I, P-II, T-II), one on granite (T-I). The gravel content (>2 mm) ranged from 9 to 47%. The management systems had no significant main effect on the contents of organic C, total N, P, and S. The mean total contents of man-derived heavy metals decreased in the order Cu (164  $\mu$ g g<sup>-1</sup> soil) > Zn (100  $\mu$ g g<sup>-1</sup> soil) > Pb (32  $\mu$ g g<sup>-1</sup> soil). The contents of microbial biomass C varied between 320 and 1,000  $\mu g g^{-1}$  soil. The significantly highest content was found at location P-II, the significantly lowest at the moderately acidic location T-I. The contents of microbial biomass N and adenosine triphosphate showed a similar trend. At location T-I, the fungal ergosterol-tomicrobial biomass C ratio and the metabolic quotient  $qCO_2$ were significantly highest, whereas the percentage of soil organic C present as microbial biomass C was lowest. Highest percentages of soil organic C present as microbial

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C. Schüler
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37213 Witzenhausen, Germany biomass C and lowest  $qCO_2$  values were found in the organic in comparison with the conventional vineyards. None of the soil microbiological indices was significantly affected by the position on the slope, but all were significantly affected by the management system. This was mainly due to the highest index levels in the organic vineyard location P-II with the longest history in organic management.

**Keywords** Microbial biomass · ATP · Ergosterol · Metabolic quotient · Gravel · Slope · Cu · Biodynamic

# Introduction

The terroir denotes the special characteristics of a wine as sum of local influences on the product (van Leeuwen and Seguin 2006). The terroir may include the climate of a vineyard, e.g., the relationship between sunshine and temperature, nutrient content, and drainage of the soil (van Leeuwen et al. 2004), but also management practices, including vine spacing, direction of rows, fertilization and harvesting techniques, pruning practices, and other factors (Bodin and Morlat 2006a, b). These factors might affect the vine plants directly and also local wild yeasts surviving in surroundings. However, the knowledge about soil microbiological processes in vineyards is generally very much restricted, probably due to steep slopes and high contents of gravel causing severe difficulties in the analysis of any soil property. This is surprising, as the behavior of soil microorganisms may be an important component for the terroir.

High levels of soluble nutrients due to inorganic fertilizer application change the microbial colonization of roots (Schloter et al. 2003), negatively affect mycorrhizal colonisation (Gryndler et al. 2006; Kleikamp and Joergensen

2006), and may also reduce the amounts of roots (Henry et al. 2005). For this reason, organic and especially biodynamic vineyard management systems have gained increasing interest as possibility for improving soil and wine grape quality (Reeve et al. 2005). Organic vineyard management systems are characterized by waiving synthetic pesticides and the use of organic fertilizers, green manure, and shallow, often reduced tillage. Biodynamic farming is based on the philosophical concept of Rudolf Steiner and an important form of organic management. It is characterized by the unique use of field and compost preparations and the use of an astrological calendar to determine times of planting and harvesting (Podolinsky 2000). Negative effects of organic, but also conventional, vineyard management may be caused by the long-term use of high CuSO<sub>4</sub> doses as fungicide (Aoyama and Nagumo 1996, 1997; Rothstein et al. 2004). Consequently, one main objective was a thorough characterization of microbial biomass [C, N, adenosine triphosphate (ATP), ergosterol] and activity (basal respiration, adenylate energy charge) indices in relation to important chemical soil properties (carbon, nutrient elements, heavy metals) and also to differences between the bottom and top positions on the vineyard slope. The question was whether organic vineyard management positively affects especially the soil microbiological indices in comparison with their conventional neighbors.

# Materials and methods

#### Locations and sampling

Vineyard soils were sampled on 28 and 29 September 2004 from two locations at Pfaffenheim (P-I, P-II; 47°57'N, 07° 17'E) and two locations at Turckheim (T-I, T-II; 48°05'N, 07°16'E) in the southwest of Colmar, Elsas, France (Table 1). The mean annual precipitation is around

530 mm, the mean annual temperature is about 10°C, and the mean annual sunshine period is approximately 1,700 h. The four locations formed four pairs of organically and conventionally managed vineyards growing different varieties of grapes (Vitis vinifera L. ssp. Vinifera; Table 1). The samples were taken with 500-ml steel cores at 0- to 10-cm depth after removal of the aboveground biomass. In each vineyard, eight samples were taken, four at the bottom and four at the top sampling area, approximately 40 to 50 m apart from each other. Each sampling area formed a  $1.5 \times 1.5$  square within the spacing between two rows of grape plants. The distance between the neighboring organically and conventionally managed vineyards was approximately 6 m at the bottom and at the top sampling areas. The samples were preincubated for 2 weeks at room temperature, sieved (<2 mm) during this time, and then stored in polyethylene bags at 4°C. The gravel >2 mm was dried and weighed.

The organic vineyards were managed according to the biodynamic regulations of the Demeter (Pfaffenheim) or Biodyvin (Turckheim) organizations. Both biodynamic winegrowers used the field preparations 500 (horn manure) and 501 (horn quartz powder) three times and two times per year, respectively. Since 2000, the field preparation 500 was partially replaced by the cow mist compost preparation (Podolinsky 2000). The compost used at Turckheim was a mixture of cattle, sheep goat, and horse manure obtained from organic farms in the neighborhood. The vineyards under conventional management received customary amounts of NPK fertilization, i.e., approximately 60 kg N ha<sup>-1</sup>, 10.3 kg P ha<sup>-1</sup>, and 20 kg K ha<sup>-1</sup>. The organic vineyards were tilled once a year and the conventional vineyards three to four times a year using a spade machine.

#### Soil chemical analysis

Soil pH was measured in water using a soil/water ratio of 1:2.5. Subsamples of dried soil material were homogenized

 Table 1
 Vine classification, parent material, slope, facing, start of organic management, grape variety, yield, grow up of grass and herbs, and compost addition at the four vineyard locations

	P-I	P-II	T-I	T-II
Parent material	Limestone	Limestone	Granite	Limestone
Slope (%)	4	20	25	45
Facing	East	East/southeast	South/southeast	East/southeast
Start of organic management	1981	1970	1998	1998
Variety-organic	Weißburgunder	Spätburgunder	Riesling	Riesling
Variety-conventional	Riesling	Gewürztraminer	Riesling	Gewürztraminer
Yield-organic (hl ha <sup>-1</sup> )	70	35	35	40
Yield-conventional (hl ha <sup>-1</sup> )	95	50	60	70
Grow up-organic	Spontaneous	Spontaneous	Spontaneous	Spontaneous
Grow up-conventional	Bare fallow	Spontaneous	Bare fallow	Spontaneous
Compost-organic (t ha <sup>-1</sup> )	No	10 (every fifth year)	3 (every year)	10 (every third year)

in a ball mill. Contents of total C and total N were determined using a Vario Max CN analyzer (Elementar, Hanau, Germany). Soil organic C was measured as total C minus carbonate C, which was measured gas-volumetrically after the addition of HCl. Concentrations of total P, total S, Zn, Pb, Cu, Co, Ni, and Cd were determined in soil and gravel after HNO<sub>3</sub>/pressure digestion as described by Chander et al. (2001) and analyzed by ICP-AES (Spectro Analytical Instruments/Kleve).

# Soil microbiological analysis

For the measurement of basal respiration, 50 g (oven-dry basis) of moist soil samples was weighed into a 1-1 stoppered Pyrex jar, adjusted to 50% of their maximum water-holding capacity, and reconditioned for 1 day at 25°C in the dark. Then, CO<sub>2</sub> production was measured for another 4 days. The CO<sub>2</sub> evolved was absorbed in 20-ml 0.5-M NaOH solution and determined by titration of the excess NaOH after addition of 0.5-M BaCl<sub>2</sub> solution to pH 8.3 with 0.1-M HCl. The metabolic quotient qCO<sub>2</sub> was calculated as follows: (µg CO<sub>2</sub>–C evolved in 4 days g<sup>-1</sup> soil)/(µg microbial biomass C g<sup>-1</sup> soil at the end of the 4-day incubation period)/4 days×1,000=mg CO<sub>2</sub>–C d<sup>-1</sup> g<sup>-1</sup> microbial biomass C.

Microbial biomass C and microbial biomass N of soil samples used to determine basal respiration were estimated by fumigation–extraction using 0.5-M K<sub>2</sub>SO<sub>4</sub> (Brookes et al. 1985; Vance et al. 1987). The organic C content of extracts was measured as CO<sub>2</sub> by infrared absorption after combustion at 850°C using a Dimatoc 100 automatic analyzer (Dimatec, Essen, Germany). Microbial biomass C was calculated as  $E_C/k_{\rm EC}$ , where  $E_C$ =(organic C extracted from fumigated soils)–(organic C extracted from nonfumigated soils) and  $k_{\rm EC}$ =0.45 (Wu et al. 1990). Total N in the extracts was measured after combustion at 850°C using a Dima-N chemoluminescence detector (Dimatec). Microbial biomass N was calculated as  $E_N/k_{\rm EN}$ , where  $E_N$ =(total N extracted from fumigated soils)–(total N extracted from nonfumigated soils) and  $k_{\rm EN}$ =0.54 (Brookes et al. 1985).

The fungal cell-membrane component ergosterol of soil samples used to determine basal respiration was extracted with ethanol from 2-g moist soil (Djajakirana et al. 1996). Then, ergosterol was determined by reversed-phase high-performance liquid chromatography with 100% methanol as the mobile phase and detected at a wavelength of 282 nm. Adenylates [ATP, adenosine diphosphate (ADP), and adenosine monophosphate (AMP)] were extracted with an alkaline dimethyl sulfoxide buffer according to Bai et al. (1988) as described by Joergensen and Raubuch (2005) from a moist sample of 4-g soil used for determining the basal respiration. The adenylate energy charge (AEC) was calculated on a molar basis as  $(ATP+0.5 \times ADP)/(AMP+ADP+ATP)$ .

# Statistical analysis

The results presented in the tables are arithmetic means and expressed on an oven-dry basis (about 24 h at 105°C). The effects of locations, vineyard management, and position on the slope (top or bottom) on soil chemical properties as well as soil microbial biomass and activity indices were analyzed by a three-way analysis of variance (ANOVA). Significance of differences between the four vineyards was additionally tested by one-way ANOVA using the Games/ Howell test, which was used for multiple comparisons, as this test is robust against the violation of normality and equality of variance (StatView Reference Manual, SAS Inst.). These two features were not always fulfilled as shown by the  $\chi^2$  test for normal distribution and the F test for equality of variance. Neither log- nor arcsin-transformation did improve the distribution of the data. The relationships between the different soil properties were analyzed by principal component analysis using the orthotran/varimax rotation to achieve either small or large component loading and an eigenvalue of 0.1 as the lower limit. All statistical analyses were performed using StatView 5.0 (SAS Inst.).

#### Results

At the three locations on limestone, the soil pH in  $CaCl_2$  was slightly alkaline, and the carbonate content varied around 18% (Table 2). At the carbonate-free granitic location, the soil pH was moderately acidic. The bulk density varied between 1.0 and 1.2 g cm<sup>-3</sup> and was not affected by the strong differences in gravel content

**Table 2** Mean pH, mean contents of carbonate and gravel >2 mm, bulk density with gravel, and conversion factor of the contents  $g^{-1}$  soil to ml<sup>-1</sup> substrate (soil plus gravel >2 mm) in the soils of the four vineyard locations at Pfaffenheim (P-I, P-II) and Turckheim (T-I, T-II)

	Soil pH (CaCl <sub>2</sub> )	Carbonate (%)	Gravel (%)	Bulk density (g cm <sup>-3</sup> )	Conversion factor $(g^{-1}$ soil to $ml^{-1}$ substrate)
P-I	7.2 a	16 a	9 c	1.0 b	0.95 a
P-II	7.1 b	19 a	34 b	1.0 b	0.68 c
T-I	6.2 c	0 c	47 a	1.2 a	0.64 c
T-II	7.1 b	20 a	29 b	1.2 a	0.83 b
CV (±%)	1.0	26	16	8.4	10

Different letters indicate a significant difference between the locations (one-way ANOVA, P<0.05, Games/Howell, n=16)

CV Mean coefficient of variation between replicate samples per vineyard and sampling area (n=4)

(>2 mm), which ranged from 9 to 47%. However, the bulk density was significantly lower at Pfaffenheim (P-I+P-II) than at Turckheim (T-I+T-II). The conversion factor from  $g^{-1}$  soil to  $ml^{-1}$  substrate varied from 0.64 to 0.95 and reflected the differences in gravel content and bulk density.

The contents of soil organic C varied between 24 and 42 mg  $g^{-1}$  soil with the highest content at location P-II, exceeding the other three locations significantly by 60% (Table 3). The locations P-I, T-I, and T-II contained similar levels of organic C, total N, total P, and total S. An exception was the total S content of the granitic location T-I, which was significantly the lowest. The limestone gravel contained on average 3.5 mg S  $g^{-1}$  and the granite gravel only 32  $\mu$ g g<sup>-1</sup> (results not shown). The contents of total N, total P, and total S followed organic C with a mean C/N ratio of 14, a mean C/P ratio of 20, and a mean C/S ratio of 35. At the top, the contents C, N, and P clearly exceeded those at the bottom. The management systems had no significant main effect on the contents of these four elements, but the significant location x system interactions were caused by the highest contents in the organic vineyard at location P-II.

The mean total contents of heavy metals decreased in the order Cu (164  $\mu g g^{-1}$  soil) > Zn (100  $\mu g g^{-1}$  soil) > Pb

**Table 3** Mean contents of organic C, total N, total P, and total S for comparing the soils of the four vineyard locations at Pfaffenheim (P-I, P-II) and Turckheim (T-I, T-II), top and bottom of the vineyard, and vineyard management system

	Organic C	Total N	Total P	Total S	
	(mg g <sup>-1</sup> soil)				
P-I	27 b	1.8 b	1.2 c	1.2 c	
P-II	42 a	3.3 a	2.4 a	1.8 a	
T-I	24 b	1.9 b	1.3 b	0.3 d	
T-II	28 b	2.2 b	1.3 b	1.4 b	
Тор	33	2.4	1.6	1.2	
Bottom	28	2.1	1.5	1.2	
Organic	30	2.3	1.6	1.2	
Conventional	31	2.3	1.5	1.2	
Probability levels of th	e three-way A	ANOVA			
Location	< 0.01	< 0.01	< 0.01	< 0.01	
Slope	0.01	0.02	0.01	0.02	
System	0.42	0.92	0.13	0.10	
Location x slope	0.04	0.23	0.08	0.81	
Location x system	< 0.01	< 0.01	< 0.01	< 0.01	
System x slope	0.16	0.67	0.81	0.01	
CV (± %)	16	17	8.3	9.1	

Different letters indicate a significant difference between the locations (one-way ANOVA, P < 0.05, Games/Howell, n=16)

CV Mean coefficient of variation between replicate samples per vineyard and sampling area (n=4)

(32  $\mu$ g g<sup>-1</sup> soil), Ni (28  $\mu$ g g<sup>-1</sup> soil), Co (7.0  $\mu$ g g<sup>-1</sup> soil) > Cd (1.5  $\mu$ g g<sup>-1</sup> soil; Table 4). Significantly, the highest contents of Cu and Pb were found at the locations P-II and T-I, and significantly the lowest contents of Ni, Co, and Cd at the granitic location T-I. The difference between top and bottom sampling areas was not significant for the contents of Pb, Ni, Co, and Cd. The contents of Zn and Cu were significantly higher at the top, but remained unaffected by management system in contrast to the other four heavy metals.

The contents of microbial biomass C varied between 320 and 1,000 mg  $g^{-1}$  soil with the significantly highest value at location P-II and the significantly lowest at location T-I (Table 5). These results were independent from the expression of the data in  $\mu g g^{-1}$  soil or  $\mu g m l^{-1}$  substrate (Fig. 1). The contents of microbial biomass N and ATP gave a similar picture, but the differences between the locations were not always significant (Table 5). The mean microbial biomass C/N ratio was 6.2, the mean ATP/ microbial biomass C ratio was 6.0  $\mu$ mol g<sup>-1</sup>, and the mean AEC was 0.84 (results not shown). The ergosterol content did not differ significantly between the four locations (Table 5). For this reason, the values of the ergosterol-tomicrobial biomass C ratio were significantly the highest with 0.48% at the acidic and granitic location T-I. The CO<sub>2</sub> production rate was more than double at location P-II in comparison with the other three locations. At location T-I, the metabolic quotient  $qCO_2$  was significantly highest and the percentage of soil organic C present as microbial biomass C lowest (Table 6). Highest percentages of soil organic C present as microbial biomass C and lowest  $qCO_2$ values were found at the organic vineyards in comparison to the conventional vineyards. This indicates again a strong negative relationship between these two quotients (r=-0.60, n=64, P<0.0001). None of the soil microbiological indices presented in Tables 5 and 6 was significantly affected by the position on the slope, but all were significantly affected by the vineyard management system. This was mainly due to the highest index levels in the organic vineyard at location P-II as also indicated by the significant location xsystem interactions (Fig. 1).

The different soil properties could be assigned to three different factors by the principal component analysis, explaining 53, 22, and 12% of the variance (Table 7). ATP, ergosterol, microbial biomass N, microbial biomass C,  $CO_2$  production, soil organic C, and total N formed the first factor, characterizing the function of soil organic matter as habitat for soil microorganisms. The contents of total Cd, Ni, Co, and S were combined as the second factor, characterizing the rock-derived element load. The contents of total Pb, Cu, Zn, and P were summarized as third factor, characterizing the man-derived element load. Total Zn was additionally affected by factor 2 of the principal component analysis and total N by factor 3.

**Table 4** Mean contents of total heavy metals (Zn, Pb, Cu, Ni, Co, and Cd) for comparing the soils of the four vineyard locations at Pfaffenheim(P-I, P-II) and Turckheim (T-I, T-II), top and bottom of the vineyard, and vineyard management system

	Total Zn	Total Pb	Total Cu	Total Ni	Total Co	Total Cd		
	$(\mu g \ g^{-1} \ soil)$	$\overline{(\mu g \ g^{-1} \ soil)}$						
P-I	74 c	20 b	89 b	33 a	7.4 a	1.4 b		
P-II	123 a	39 a	243 a	32 a	8.3 a	1.9 a		
T-I	92 b	43 a	227 а	12 b	4.2 b	0.8 c		
T-II	111 a	27 b	95 b	34 a	8.1 a	2.2 a		
Тор	104	33	173	28	7.2	1.6		
Bottom	96	31	154	27	6.9	1.5		
Organic	100	29	166	29	7.5	1.7		
Conventional	100	36	161	26	6.5	1.5		
Probability levels of the t	three-way ANOVA							
Location	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01		
Slope	0.01	0.20	< 0.01	0.52	0.07	0.07		
System	0.92	< 0.01	0.28	< 0.01	< 0.01	< 0.01		
Location $x$ slope	0.08	0.06	< 0.01	0.44	0.15	0.46		
Location $x$ system	0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01		
System x slope	0.12	0.80	0.54	0.62	0.91	0.47		
CV (± %)	9.0	12	8.8	6.7	7.7	15		

Different letters indicate a significant difference between the locations (one-way ANOVA, P<0.05, Games/Howell, n=16) CV Mean coefficient of variation between replicate samples per vineyard and sampling area (n=4)

#### Discussion

The soil microbiological properties of the four organic and four conventional vineyards are in the range of other permanent cultures such as grassland (Djajakirana et al. 1996; Höper and Kleefisch 2001) or fruit orchards (Swezey et al. 1998; Goh et al. 1999). The small-scale spatial variability of the different soil physical, soil chemical, and

**Table 5**Mean contents of microbial biomass C, microbial biomass N, ATP, ergosterol, and the  $CO_2$  production for comparing the soils of the fourvineyard locations at Pfaffenheim (P-I, P-II) and Turckheim (T-I, T-II), top and bottom of the vineyard, and vineyard management system

	Microbial biomass C $(\mu g g^{-1} \text{ soil})$	Microbial biomass N (μg g <sup>-1</sup> soil)	ATP (nmol g <sup>-1</sup> soil)	Ergosterol ( $\mu g g^{-1}$ soil)	$CO_2$ production (µg C g <sup>-1</sup> soil d <sup>-1</sup> )
P-I	730 b	130 bc	4.2 a	1.3 a	23 b
P-II	1000 a	170 ab	4.8 a	2.3 a	46 a
T-I	320 c	50 d	2.4 b	1.5 a	22 b
T-II	640 b	110 c	3.5 ab	1.4 a	21 b
Тор	680	120	3.7	1.1	29
Bottom	660	110	3.8	1.3	27
Organic	740	130	4.1	1.4	31
Conventional	600	100	3.4	0.9	25
Probability levels of th	ne three-way ANOVA				
Location	< 0.01	< 0.01	< 0.01	0.02	< 0.01
Slope	0.71	0.82	0.57	0.99	0.41
System	< 0.01	0.01	0.02	0.05	0.02
Location $x$ slope	0.85	0.82	0.77	0.44	0.18
Location x system	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
System x slope	0.73	0.84	0.52	0.40	0.85
CV (± %)	25	28	27	49	32

Different letters indicate a significant difference between the locations (one-way ANOVA, P < 0.05, Games/Howell, n=16)

CV Mean coefficient of variation between replicate samples per vineyard and sampling area (n=4)



**Fig. 1** Mean contents of microbial biomass C in  $\mu g g^{-1}$  soil and in  $\mu g ml^{-1}$  substrate (soil+gravel >2 mm), comparing organic and conventional vineyard management system in the soils of the four vineyard locations at Pfaffenheim (P-I, P-II) and Turckheim (T-I, T-II)

soil microbiological properties is in the range observed for arable, grassland, and forest soils (Stork and Dilly 1998; Joergensen and Castillo 2001; Höper and Kleefisch 2001; Emmerling and Udelhoven 2002). The slight but significant

**Table 6** Mean contents of the metabolic quotient  $qCO_2$  and the microbial biomass C/organic C ratio for comparing the soils of the four vineyard locations at Pfaffenheim (P-I, P-II) and Turckheim (T-I, T-II), top and bottom of the vineyard, and vineyard management system

	Metabolic quotient $q$ CO <sub>2</sub> (mg CO <sub>2</sub> -C d <sup>-1</sup> g <sup>-1</sup> biomass C)	Microbial biomass C/ soil organic C (%)
P-I	31 c	2.4 a
P-II	46 b	1.7 b
T-I	74 a	1.4 b
T-II	35 c	1.9 ab
Тор	48	1.7
Bottom	45	1.9
Organic	43	2.0
Conventional	50	1.7
Probability leve	els of the three-way ANOVA	
Location	< 0.01	< 0.01
Slope	0.52	0.08
System	0.04	0.01
Location x slope	0.57	0.97
Location <i>x</i> system	<0.01	0.03
System x slope	0.14	0.06
CV (± %)	26	22

Different letters indicate a significant difference between the locations (one-way ANOVA, P < 0.05, Games/Howell, n=16)

CV Mean coefficient of variation between replicate samples per vineyard and sampling area (n=4)

**Table 7** Oblique solution primary pattern matrix of the principal component analysis for the different soil chemical and soil biological properties (orthotran/varimax transformation; n=64)

	Factor 1	Factor 2	Factor 3
ATP	0.97	0.09	-0.29
Ergosterol	0.93	-0.27	0.00
Microbial biomass N	0.87	0.24	-0.17
Microbial biomass C	0.84	0.28	-0.11
CO <sub>2</sub> production	0.77	-0.02	0.30
Organic C	0.73	0.11	0.30
Total N	0.62	0.16	0.42
Total Cd	-0.14	0.99	0.20
Total Ni	0.00	0.96	-0.25
Total Co	0.05	0.94	-0.02
Total S	0.29	0.77	0.03
Total Pb	-0.28	-0.20	0.91
Total Cu	0.27	-0.39	0.80
Total Zn	-0.16	0.56	0.79
Total P	0.34	0.25	0.67
Eigenvalue	7.9	3.3	1.8
Explained variance (%)	53	22	12

Italicized values Definite assignation to a certain factor

higher contents of organic matter at the top sampling area of the vineyards might be due to a reduced microbial turnover of soil organic matter caused by somewhat lower water contents (Oliveira 2001; Ramos 2006). These higher soil organic matter contents at the top indicate additionally the absence of serious erosion problems (Battany and Grismer 2000).

The soils of the three locations on limestone are characterized by relatively high contents of total S, which are caused by the geogenic sulfate concentrations of the parent material (Khan and Joergensen 2006). In contrast, the high and similar contents of total P at all four locations are likely due to fertilizer application as suggested by the principal component analysis. Also, the high contents of Cu are mainly of anthropogenic origin due to the spray of CuSO<sub>4</sub> as fungicide against the grapevine downy mildew Plasmopara viticola for many decades. Probably, differences in Cu content of the four locations depended on differences in the amounts of CuSO<sub>4</sub> applied. The organic vineyard management did not lead to increased Cu levels in comparison to their conventional neighbors. However, all soils were above the current permitted limits in Germany of  $60 \ \mu g \ Cu \ g^{-1}$  soil for the application of sewage sludge to soils (AbfKlärV 1992). The Cu concentration of the present vineyard soils was similar to a highly contaminated hop yard (Zelles et al. 1994), but less than 50% of the Cu content observed in soils of the Oker floodplain contaminated with mining residues (Chander et al. 2001) and less than 25% of a highly Cu-contaminated apple orchard in Japan (Ayoama and Nagumo 1996, 1997).

In contrast to Zelles et al. (1994) and Chander et al. (2001), the present Cu concentrations did not lead to a reduction in AEC. Also, a strong shift in the microbial community structure towards fungi, as indicated by relative increases in ergosterol or fungal phospho lipid fatty acids, was not observed with increasing heavy metal contamination (Frostegård et al. 1996). The mean ergosterol-to-microbial biomass C ratio was 1.1% in heavy metal-contaminated soils (Chander et al. 2001), two times larger than at the acidic and granitic location T-I, and roughly five times larger than at the other three locations on limestone. All ergosterol-to-microbial biomass C ratios of the present vinevard soils were in the range observed in grassland soils by Djajakirana et al. (1996). High concentrations of fungicidal Cu apparently do not promote soil fungi in contrast to high levels of Zn and Pb contamination.

At the acidic location T-I, not only the highest ratio ergosterol-to-microbial biomass C was measured but also the highest metabolic quotient  $qCO_2$  and the lowest percentage of soil organic C present as microbial biomass C. High  $qCO_2$  values indicate a relatively larger proportion of younger microorganisms with low substrate-use efficiency (Anderson and Domsch 1990) and low percentages of soil organic C present as microbial biomass C, a poor availability of organic matter to soil microorganisms (Anderson and Domsch 1989). This means that more substrate is diverted towards catabolic at the expense of anabolic processes, leading to reduced microbial biomass levels in the long term (Chander and Brookes 1991) and thus to lower percentages of soil organic C present as microbial biomass C, as observed in the present set of soils. However, the present results contradict the view that fungi have generally a higher substrate-use efficiency than bacteria (Sakamoto and Oba 1994).

The metabolic quotient  $qCO_2$  and the percentage of soil organic C present as microbial biomass C were not only affected by heavy metal contamination but also by the vineyard management. The organic vineyards exhibited on average lower  $qCO_2$  values and higher percentages of soil organic C present as microbial biomass C than their conventional neighbors. Lower  $qCO_2$  values in combination with higher percentages of soil organic C present as microbial biomass C as result of organic farming and especially of organic farming has been repeatedly shown in the "bio-Dynamic, bio-Organic, and Konventionell" trial (Mäder et al. 2002) and also in the long-term soilmonitoring program of Lower Saxony, Germany (Höper and Kleefisch 2001). The exact reasons for the increases in microbial substrate-use efficiency and in availability of soil organic matter to soil microorganisms cannot be definitely assigned to a reduction in tillage intensity, the absence of synthetic herbicide and easily soluble inorganic fertilizers, or the application of organic preparations. In short-term

experiments with less than 10 years observational period, the effects of organic management systems on soil microbiological properties in comparison to integrated or conventional management system are less clear or even absent (Swezey et al. 1998; Goh et al. 1999; Glover et al. 2000a, b), although the terroir of the wine indicated already differences (Reeve et al. 2005).

This is in agreement with the present results, where the positive effect of the organic vineyard management on microbial substrate-use efficiency and availability of soil organic matter to soil microorganisms increased with the increasing time-span since the conversion from conventional management. The location P-I with the largest differences between organic and conventional management was converted in 1970 to organic farming principles and in 1981 to biodynamic management. However, positive effects of organic vineyard management might be masked by differences in land-use history. The pair-wise comparison of organic vineyards with their conventional neighbors is based on the assumption that conventional and organic vineyards had identical soil chemical and soil microbiological properties at the time of conversion. This assumption is of course not necessarily valid, emphasizing the strong demand for controlled long-term field experiments on vineyard management systems.

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