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Grapevine leaf ionome is shaped by soil factors and plant age

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Abstract: The concept of *terroir* relates the sensory attributes of wines attributable to the environmental conditions of the grapevines, and therefore it represents an important descriptor of the connection between wines and their origins. In ongoing efforts to improve the monitoring and geochemical fingerprinting of grapevine products, knowledge of the variability of grapevine elemental compositions, and factors that have strong influences on this, can significantly improve the traceability of wines to their origins. Here, we demonstrate a strong connection of grapevine elemental composition to the micro-location of the individual vineyard, with an important contribution from the biotic soil factors. The differences in measured leaf elements appear to be more closely connected to the grapevine age than to the viticultural practice (biological *vs.* conventional). Soil microbial communities have a substantial impact on grapevine leaf elements, with differences seen between fungi and bacteria. Bacteria appear to be more closely related to the environment in vineyards than fungi, with changes in their interplay reflected in the elemental composition of the grapevines. Nevertheless, both microbial groups explain 15% to 17% of the variation in the grapevine leaf elements, making the soil fungal and bacterial communities critical factors in the *terroir* concept.

Keywords: *Vitis vinifera* L.; copper; biotic factor; mycorrhizal fungi; nutrient

In viticulture, the concept of *terroir* connects the sensory attributes of wines to the vineyard's environmental conditions (OIV 2010). The soil characteristics are the most important factors for the *terroir*, as they have a powerful impact on the grapevine mineral composition (Likar et al. 2015).

Several important soil factors can influence the mineral composition of plants (Humphries et al. 2007). However, considering the biological approach, in particular, contemporary agriculture recognises plants as part of a complex agroecosystem where many organisms co-exist and interact. This approach emphasises the importance of interactions between the soil, the microbe communities, and the plants, as these influence the growth, physiology and yield of crop plants (Likar et al. 2015, Bao et al. 2022). Biotic factors such as grapevine cultivar and rootstock affect the uptake of elements from the soil (Wooldridge et al. 2010). Furthermore, soil microbiomes can profoundly affect the availability of the soil elements and hence their uptake by grapevines (Lewis et al. 2018).

In viticulture, fungicide application (Trouvelot et al. 2015), acidification of the soil due to fertiliser input (Muñoz-Leoz et al. 2012), and tillage practices (Likar et al. 2017) can cause severe adverse effects on the soil microbial communities, including beneficial soil microbes, such as the mycorrhizal fungi. In contrast, the low-input measures of biological vineyard management can provide better conditions for the support of higher diversity of beneficial microbes (Radić et al. 2014). However, copper use in the biological approach can lead to its accumulation within the topsoil (Brundretto et al. 2016), which

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can also profoundly affect the microbe communities. Viticultural practices can thus change the microbe-grapevine interactions and result in differences in grapevine elemental composition.

In the ongoing efforts to develop techniques for the geochemical fingerprinting of wines, knowledge of the variability of the grapevine elemental composition on small and large geographical scales and the factors that have a strong influence on it would significantly improve the traceability of wines to their origins. The main aim of the present study was to assess the impact of the soil on the elemental composition of grapevine leaves at a landscape scale and to relate these to the type of grape production, duration of production, and within vineyard habitats. We hypothesised that the vineyard management practices (i.e., conventional *vs*. biological) influence the elemental compositions of the grapevines directly (through the changes in soil characteristics) or/and indirectly (through improved microbial diversity in organically managed vineyards).

MATERIAL AND METHODS

Study vineyards. Grapevine leaves of cv. Merlot were sampled in vineyards located near Šempas, Slovenia (45°55.614N, 13°44.568E). The winegrowing district at ~90 m a.s.l. is characterised by a sub-Mediterranean climate, with a mean annual air temperature of 12 °C and annual precipitation of 1 500 mm. The parent matter of the area is classified as Cretaceous platform carbonate rock, a limestone-dolomite non-clastic siliceous sedimentary rock with Eutric brown soil, typic and calcaric on flysch (Eutric, Calcaric Cambisols) (FAO 1974, 1988). The soils and leaves were sampled from four vineyards: vineyard B3, with 3 years of biological management; vineyard B10, with 10 years of biological management; vineyard B35, with 35 years of biological management; and vineyard CVT35, with 35 years of conventional management. The grapevines in the vineyards under ecological management are integrated into the biological/organic production, as set out in "Rules on organic production and processing of agricultural products and/or foods" (Official Gazette of RS 2001, 2003, 2006) and EC 889/2008. The detailed technological characteristics of biological and conventional vineyards included in this study are shown in Table 1.

Sample collection and handling. The sampling design followed a grid pattern with 35 sampling points for each vineyard (total, 140 samples), which were collected using cores of 1-dm^3 that extracted the soil to a depth of 15 cm. The sampling points were positioned under the grapevine plants in the vineyard rows (three rows for each sampling grid) and the paths between the vineyard rows (two paths for each sampling grid). The sampling points were separated by 2 m along the X-axis and 5 m along the Y-axis, and the whole grid covered an area of 240 m^2.

Leaf samples were collected in September (at veraison) from all vineyards. Ten fully expanded leaves from the whole canopy (front and back; sunny and shadowy; top and bottom of the canopy) were sampled and stored in PVC bags at 4 °C until further analysis. In total, 25 grapevines were sampled in each vineyard, as 10 leaves/vine. The elemental compositions of the grapevines were determined according to the European criteria for the determination of grapevine elements, which are based on whole-leaf analysis, in contrast to the American criteria where only the leaf petiole is analysed (Čoga et al. 2008).

Soil and leaves analysis. Before further analysis, the soil samples were air dried, ground to a fine powder in an agate mortar, and sieved to 2 mm. Total organic matter was measured by wet combustion, according to Kandeler (1995). The plant-available phosphorous was determined photometrically, according to Olsen and Sommers (1982). The soil pH was measured in soil:water extracts (1:2, *v*/*v*), using deionised water. The soil texture and water content were determined as described by Alef and Nannipieri (1995). The soil elemental composition was measured using energy dispersive X-ray fluorescence spectrometry as described in Vogel-Mikuš et al. (2010), and the elemental composition of leaves was measured using total reflection X-ray spectrometry (Seifert, Ahrensburg, Germany) as described in Nečemer et al. (2008).

Fungal and bacterial community analysis. Due to a large number of samples, the fast-fingerprinting approach of automated ribosomal intergenic spacer analysis (ARISA) was used. The DNA of the microbial communities was extracted using soil DNA extraction kits (PowerSoil; MO BIO Laboratories, Carlsbad, USA), following the manufacturer's instructions. Analysis of fungal and bacterial communities using ARISA was performed as described in Likar et al. (2017).

Statistical analysis. All of the mathematical and statistical computations were carried out using the R 4.1.2 software (Vienna, Austria). Differences in element leaf concentrations between individual vineyards were analysed using one-way ANOVA and Holm-Bonferroni post hoc tests at $P < 0.05$. Pearson correla-

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Table 1. Technological characteristics of the biological and conventional management in studied vineyards

	Ecological vineyards		Conventional vineyard	
	measure	frequency – growing season	measure	frequency – growing season
Downy mildew (*Plasmopara viticola* (Berk. & M.A.Curtis) Berl. & De Toni in Sacc.)	spraying with copper hydroxide ($Cu(OH)_2$) and copper oxychloride ($3Cu(OH)_2$ $CuCl_2$) rate of 28 kg/ha of copper over a period of 7 years (i.e. on average 4 kg/ha/year)[a]	up to 5–7 times	spraying with copper-based and other different allowed chemical agents[b]	copper agents commenly up to 2 times; other agents commenly up to 6–10 times
Powdery mildew (*Uncinula necator* (Schwein.) Burrill)	spraying with sulphur in solid forms (powder) at a maximum of 3–8 kg per year/hectare[c]	up to 7–15 times	spraying with sulphur-based and other different allowed chemical agents[b]	sulphur agents commenly up to 5–10 times; other agents commenly up to 5–8 times
Fertilisation	organic fertiliser (animal manure) and/or low solubility mineral fertilisers; max of 170 kg of organic N/ha/year of agricultural area[d]	according to the chemical analysis of the soil and fertilising plan; up to 2 times	high solubility mineral fertilisers and/or organic fertilisers; max of 110 kg of N/ha/year[b]	according to the chemical analysis of the soil and fertilising plan; up to 2 times
Soil management	100% of vineyards with permanent green cover between rows; mechanical or no chemical weed control under vines[c]	mulching, mowing and/or covering; up to 3 times	at least 50% of the vineyard with permanent green cover between rows; mechanical and/or chemical control of weeds under vines[b]	mulching and/or mowing up to 3 times; chemical control up to 2 times

[a]Commission Implementing Regulation (EU) 2018/1981, of 13 December 2018; renewing the approval of the active substances copper compounds, as candidates for substitution, in accordance with Regulation (EC) No. 1107/2009 of the European Parliament and of the Council concerning the placing of plant protection products on the market, and amending the Annex to Commission Implementing Regulation (EU) No. 540/2011; https://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:32018R1981&rid=3; [b]Technological rules for integrated grape production; https://www.gov.si/teme/integrirana-pridelava/; [c]Technological rules for ecological grape production; https://www.gov.si/assets/ministrstva/MKGP/DOKUMENTI/KMETIJSTVO/NACINI-KMETIJSKE-PRIDELAVE/Tehnoloska-navodila-EK/8fe380eb9e/TEHNOLOSKA_NAVODILA_ZA_EKOLOSKO_PRIDELAVO_GROZDJA.pdf; [d]Regulation (EC) No. 834/2007 of 28 June 2007 on organic production and labelling of organic products

tions between soil characteristics and leaf elements were calculated in R using Pearson's R coefficients.

Principal component analysis (library factoextra v1.0.7) and heatmaps (library gplots v3.0.1.1) were used to visualise the differences in the elemental composition of the grapevine leaves and the soil characteristics for the different vineyards. The data were standardised and scaled using the scaling function in R prior to analysis. The results of the PCA and heatmap/clustering analyses were further analysed using perMANOVA on Bray-Curtis dissimilarity matrices (library vegan v2.5-7).

Matrices pertaining to space (based on the principal components of the neighbour matrices), environmental factors, and microbial soil communities were used to examine the variation partitioning (library vegan v2.5-7) of the leaf elements in the grapevines. We used the vectors from the principal components of the neighbour matrices that best accounted for the autocorrelation and then conducted forward selection (999 permutations at $P < 0.05$) to select the spatial factors that significantly influenced the community dissimilarities. Prior to analysis, the environmental factors and microbial soil communities were also

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subjected to forward stepwise redundancy analysis to reduce the number of variables used in the variation partitioning.

RESULTS AND DISCUSSION

Impact of viticulture and age of the grapevines. The concentrations of K, Ca, Mn and Zn were significantly different among the grapevines (Table 2). The leaf concentrations of Ca decreased with the increasing age of the grapevines, irrespective of the vineyard management. The leaf concentrations of K were significantly lower for conventional vineyard management compared to the other grapevines (Table 2). The leaf concentrations of Mn were significantly higher for the 35-year-old grapevines, with the lowest seen for the B10 grapevines. The leaf concentrations of Zn also tended to increase with the age of the grapevines when under biological management and peaked at 51.45 ± 4.80 mg/kg DW (dry weight) (Table 2).

The highest K concentrations were seen for the 10-year-old grapevines in the biological vineyards, which is in agreement with our previous study that compared vineyards along the Adriatic coast (Likar et al. 2015). These data suggest that K uptake can be improved under biological viticulture, which can be partly explained by greater K accumulation in soils with a permanent green cover, as it can increase soil moisture and consequently K uptake by the grapevines (Hirschfelt et al. 1992).

In addition, the concentrations of Zn showed indications of increases with the age of the grapevines under biological management. Rusjan et al. (2006) reported that Zn accumulates in vineyard soils, especially with long-term application of Cu-based fungicides, as mainly seen in the use of Bordeaux mixture, which also contains various hazardous metals. The Cu concentrations were indeed highest in the vineyard B10, which was at the peak of production age. In addition, Zn is frequently used in animal feed as a dietary supplementation, and through the fertilisation with organic (animal) manure and compost that is allowed in biological vineyards, this will also increase the Zn concentrations in the soil, and consequently also potentially in the grapevines (Yamamoto et al. 2018). The third most frequent source for the accumulation of Zn in grapevine leaves are the use of various foliar fertilisers. Grapevines treated with Mg and/or Fe fertilisers have been shown to have increased Zn concentration in their leaves (Brataševec et al. 2013). Moreover, possible interactions with symbiotic fungi should not be excluded, as Radić et al. (2014) reported higher colonisation with arbuscular mycorrhizal fungi in biological vineyards. Arbuscular mycorrhizal fungi can supply up to 24% of the total Zn, e.g. tomato (Watts-Williams et al. 2015). This suggests smaller effects of biological viticulture on symbiotic fungi compared to conventional viticulture and, consequently, a greater impact of these fungi on the element nutrition of the grapevines.

Leaf concentrations of Ca, and partly of Mn, showed some dependence on the age of the grapevines, irrespective of the cultivation methods. Grapevines have little requirement for Ca, and therefore Ca is classified as a secondary nutrient, with Ca deficiency rarely seen in vineyards (Rorison and Robinson 1984). Ca uptake by grapevines is passive, and it depends on soil Ca concentration and pH and the water influx that is regulated by grapevine transpiration. Clarkson (1984) reported that Ca concentrations in grapevine shoots highly correlate with higher soil Ca concentrations and higher soil pH, which are

Table 2. Concentrations for the elements analysed for the grapevine leaves from the selected vineyards for the 3-year-old to 35-year-old grapevines

Vineyard	Element leaf concentration						
	phosphorus	potassium	calcium	manganese	iron	copper	zinc
	(mg/kg DW)						
B3	1 208 ± 51	7 814 ± 496^{b}	30 533 ± 1 246^{a}	116.0 ± 7.0b^{c}	111 ± 4.9	450 ± 60.9^{b}	33.85 ± 2.5^{b}
B10	1 064 ± 136	10 085 ± 498^{a}	25 094 ± 1 142^{b}	91.7 ± 4.1^{c}	150 ± 15.8	1 667 ± 155^{a}	49.36 ± 5.08^{a}
B35	1 367 ± 107	9 068 ± 503^{ab}	21 917 ± 1 115^{bc}	129.0 ± 5.1^{ab}	159 ± 16.9	176 ± 18.7^{b}	51.45 ± 4.8^{a}
CVT35	1 164 ± 100	5 964 ± 617^{c}	19 491 ± 1 520^{c}	153.0 ± 14.8^{a}	112 ± 17.2	241 ± 31.0^{b}	32.77 ± 4.17^{b}

Data are means ± standard error (n = 15). Different letters within columns (element) indicate statistically significant differences (P < 0.05; Holm-Bonferroni post-tests). B – biological viticulture; CVT – conventional viticulture; numbers represents the age of the vineyards (in years); DW – dry weight

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characteristic of flysch soils, and therefore are to be also expected in these vineyards. However, the leaf concentrations of Ca in the present study decreased with grapevine age, irrespective of vineyard management. This observation can be partly explained by the flysch soil type in the study vineyards, which is abundant with Ca (i.e., $CaCO_3$). This will especially be the case for the young grapevines, as they were planted soon after the vineyard restoration and deep soil tillage. During grapevine ageing, limited vineyard soil management usually leads to the leaching of Ca from the soil particles and, consequently, to decreased soil pH (Mulidzi et al. 2019). In addition, young grapevines, in comparison to older grapevines, usually show higher vigour and shoot growth, which will result in greater leaf areas (i.e., larger canopy), which will, in turn, lead to higher transpiration, and thus greater Ca translocation through the water flux into the leaves (Drake et al. 1979).

Manganese is a transition metal that is frequently present in soils under intensive viticulture practices, with the main Mn sources being abundant fertilisation and spraying (Rusjan et al. 2006). The leaf concentrations of Mn for the grapevines in the present study showed an increasing trend with the age of the grapevines, irrespective of the management system. Grapevines can develop Mn deficiency when they are grown on calcareous soils (such as a flysch type), as the Mn gets immobilised in the soil at higher pH (Humphries et al. 2007). In agreement with this, we observed significant negative correlations between the leaf Mn and the soil Ca (Pearson $R = -0.46$, $P < 0.001$) and pH (Pearson $R = -0.44$, $P < 0.001$). However, there were particularly high leaf concentrations of Mn in the present study, as these were generally above the optimal concentrations (30–100 mg/kg). This is in agreement with our previous observations (Likar et al. 2015) and with a report by Čoga et al. (2008), who also observed supraoptimal concentrations of Mn in grapevine leaves collected at veraison, which suggests improved uptake during and soon after grapevine flowering.

In the present study, differences in the concentrations of elements in grapevine leaves appear to be connected more closely to the age of the grapevines rather than the viticultural practices. Furthermore, comparisons of the whole ionome revealed tighter connections to the micro-locations of the individual vineyards, with important contributions from biotic soil factors.

Principal component analysis showed close clustering of the B35 and CVT35 grapevines (Figure 1A). The B10 grapevines were the most removed from the others, with the highest dispersion of the samples. In contrast, the lowest dispersion of samples was observed for the youngest vineyard B3. The perMANOVA pairwise comparisons confirmed that the B35 and CVT35 grapevines did not differ significantly in the elemental compositions of their leaves ($P = 0.12$), whereas all other vineyard pairs showed statistical differences ($P < 0.01$).

Hierarchical clustering of the vineyards based on the soil parameters showed that the B35 and CVT35 grapevines were the most similar. On the other hand, B3 grapevines showed the least similarity to the other studied grapevines (Figure 1B). PerMANOVA of the soil parameters confirmed that all studied vineyards differed one from the other ($P < 0.01$). Among the measured soil parameters, we could observe the formation of three separated clusters. The first one grouped concentrations of Fe, K and Mn, clay content and soil water content, as well as fungal diversity. Low values of parameters in this cluster were associated with vineyard B10. The second cluster contained Zn, bacterial diversity and organic matter content, whereas the last one was comprised of Ca, potencial acidity and sand content. Low values of soil parameters from the second cluster were characteristic for vineyard B3. The parameters from the third cluster were more evenly distributed, with higher values in vineyards B3 and B10 and lower values in older vineyards (B35 and CVT35). As such, it appears that the soil parameters are the strongest determinant for the composition of the elements in these grapevine leaves and that biological vineyards show the same element nutrition of the grapevines as vineyards under conventional management.

Effects of soil factors and microbial communities. Several soil characteristics correlated with element concentrations in the grapevine leaves (Table 3). In particular, the potential soil acidity, soil Ca concentrations, and fungal diversity showed various correlations with several of the leaf elements. Phosphorous leaf concentrations showed only a positive correlation with fungal diversity. Potassium leaf concentrations were positively correlated with soil Ca and potential acidity and negatively with clay content and fungal diversity. Calcium leaf concentrations showed low positive correlations with several measured parameters (soil P, Mn, water content and clay content). Manganese leaf concentrations were negatively correlated with soil Ca concentrations and potential acidity and positively with clay content. Leaf Fe

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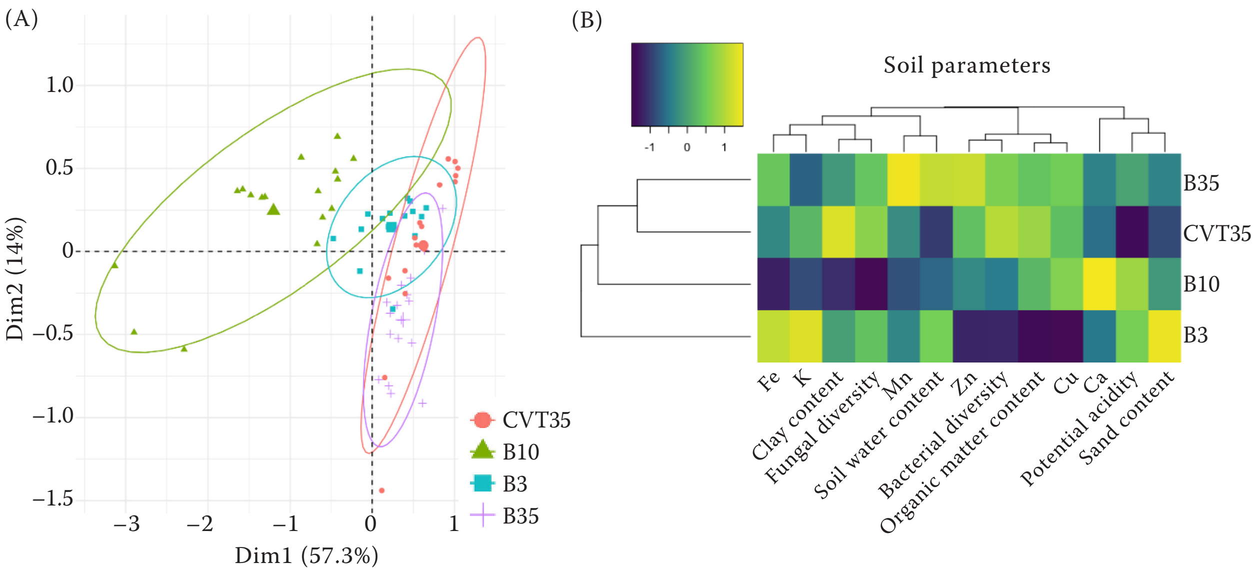

Figure 1. (A) Principal component analysis scatters plot of the element data from the leaves of the grapevines and (B) heat map of the z-scores for the soil parameters. The dendrogram represents the relationships between the vineyards (rows) and minerals (columns), with the colour in the intersections representing the magnitude of the abundance: blue, median for variable < 0; green, yellow, median for variable > 0. Dendrograms were constructed on the clusters generated using complete linkage hierarchical clustering based on Euclidean distances. B – biological; CVT – conventional; the number represents the age of the grapevines

concentration showed mainly negative correlations (soil P, organic matter and fungal diversity), although it also correlated positively with the potential acidity of the soil. A similar pattern of correlations was observed for leaf Zn, although it also negatively correlated with soil Mn.

Stepwise redundancy analysis for the leaf element composition included potential soil acidity, soil water content, soil Mn concentration, and fungal diversity. Variation partitioning showed that the environmental and fungal community datasets explained 22% (P = 0.001) and 17% (P = 0.002) of the variation in

Table 3. Pearson correlation *R* between leaf minerals and soil parameters

Soil parameter	Element leaf concentration					
	phosphorus	potassium	calcium	manganese	iron	zinc
P	–0.13	–0.12	0.27*	–0.08	–0.31*	–0.30*
K	0.24	–0.08	–0.18	0.15	0.14	0.10
Ca	–0.20	0.40***	0.13	–0.48***	0.14	0.19
Mn	0.18	–0.12	0.28*	–0.03	–0.23	–0.28*
Fe	0.15	–0.09	0.15	0.29*	0.16	0.04
Cu	–0.11	–0.08	0.22	–0.20	–0.07	–0.16
Zn	0.01	–0.16	0.23	–0.08	0.10	0.01
Organic carbon content	–0.18	–0.23	0.05	–0.01	–0.24*	–0.32**
Soil water content	0.03	0.02	0.31*	0.01	–0.05	0.03
Potential acidity	0.05	0.51***	0.27*	–0.42***	0.28*	0.31*
Clay content	–0.10	–0.37**	–0.18	0.32**	–0.01	–0.23
Sand content	0.21	0.22	–0.02	–0.12	0.10	0.26*
Fungal diversity	0.37**	–0.40***	–0.24	0.14	–0.26*	–0.44***
Bacterial diversity	–0.11	0.06	0.16	0.06	–0.18	–0.08

*P < 0.05; **P < 0.01; ***P < 0.001

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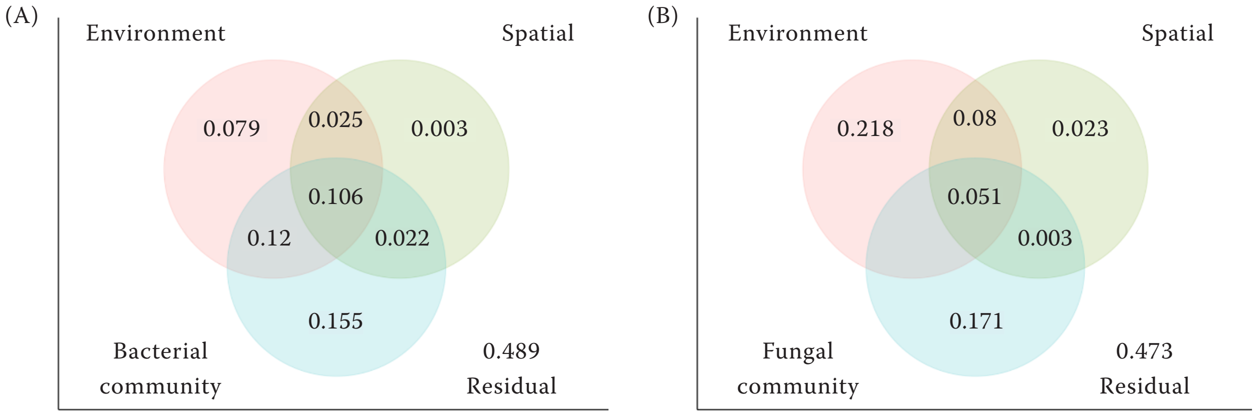

Figure 2. Variation partitioning for the leaf minerals for the grapevines with the environmental, spatial and fungal communities (A) and the bacterial community (B). Numbers show the fractions of variation explained by each factor alone or by the interactions between two or three factors, as indicated. The unexplained variation is given in the right bottom corner

leaf element composition, respectively (Figure 2A). In contrast, the spatial dataset that contained only the physical distances between the sampling points explained only 2% of the variation and did not reach statistical significance (P = 0.136). Statistical significance of individual fractions was tested using the *anova* and *rda* functions of the Vegan package (Tables 4–5).

For the variation partitioning with bacterial communities, a greater proportion of explained variation in the grapevine leaf elements was shared between the bacteria and environmental variables (22.6%) than for the fungi (only 5%) (Figure 2B). Environmental factors, in combination with spatial and microbial communities, explained up to 35% of the variation in the grapevine leaf elements. Soil pH, moisture, Mn concentrations and fungal diversity were selected as the most important soil parameters that influence the leaf element composition of these grapevines. The fungal and bacterial community composition explained roughly equal fractions of variation from 15% to 17% in the leaf elements of the grapevines. However, for bacteria, an additional large fraction of explained variation (25%) was connected to environmental factors and the spatial component.

Soil pH, moisture, Mn concentrations and fungal diversity were selected as the most important soil parameters that influence the leaf element composition of grapevines in our study. The soil pH and moisture are the main factors that determine the availability of several elements to plants (Keller 2005) and can, therefore, heavily affect the elemental composition of the plant organs.

In contrast to P concentrations, the fungal diversity was generally negatively correlated with the concentrations of the leaf elements. Colonisation with beneficial fungi is known to modulate the concentra-

Table 4. Statistical significance of individual fractions of variation partitioning for fungal communities, environmental parameters and spatial variables

	df	Variance	*F*	Pr (> *F*)
Conditional (partial) effect of environmental factors (fraction [a])				
Model	4	10591953	7.103	0.001
Residual	49	18266848		
Conditional (partial) effect of fungal community (fraction [b])				
Model	2	7609561	10.21	0.002
Residual	49	18266848		
Conditional (partial) effect of spatial components (fraction [c])				
Model	3	2047919	1.831	0.145
Residual	49	18266848		

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Table 5. Statistical significance of individual fractions of variation partitioning for bacterial communities, environmental parameters and spatial variables

	df	Variance	*F*	Pr (> *F*)
Conditional (partial) effect of environmental factors (fraction [a])				
Model	4	4671286	3	0.016
Residual	46	17724101		
Conditional (partial) effect of bacterialcommunity (fraction [b])				
Model	5	8152307	4.232	0.003
Residual	46	17724101		
Conditional (partial) effect of spatial components (fraction [c])				
Model	3	1280621	1	0.35
Residual	46	17724101		

tions of different elements due to changed uptake or improved growth, which can lead to a dilution effect (Lewis et al. 2018). It is important to note that the use of any fungicides (also in biological viticulture) and fertilisers can affect the microbial communities (Canfora et al. 2018), resulting in changes in plant nutrition.

In addition, Mn was selected as one of the important soil factors for the elemental composition of grapevine leaves. Manganese is an important micronutrient but can also interact with other elements in the soil and thus alter their acquisition by plants (Roivainen et al. 2012).

The large fraction of explained variation connected to environmental factors in the case of bacterial communities suggests that these changed more with variations in the environment or with physical distance and supports our previous findings that the fungal and bacterial soil communities appear to respond differently to environmental conditions, and their interactions (Likar et al. 2017, 2022). These differences among these two microbial groups appear to relate to their effects on the elemental composition of these grapevines. Furthermore, it appears that the bacteria and the environment in vineyards are more closely connected, with any changes in their interplay reflected in the elemental composition of the grapevines. Nevertheless, the fungal and bacterial communities explained 15% to 17% of the variation in the leaf elements of these grapevines. Thus, these microbial communities represent an important factor in the concept of the "*terroir*".

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