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ORIGINAL RESEARCH ARTICLE

# Under-vine vegetation in vineyards: a case study considering soil hydrolytic enzyme activity, yield and grape quality in Austria

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## ABSTRACT

In vineyards, the under-vine area is managed to control vegetation growth and to reduce the competing effect of growing plants on vines and fruit development. Applied under-vine management methods are the application of herbicides, soil tillage or the growth of spontaneous vegetation or cover crops. These methods affect pedo-climatic conditions differentially as well as the soil biota and have, therefore, consequences on soil functions and ecosystem services. In the presented case study, the effects of five under-vine management methods on the activity of soil hydrolytic enzymes, the soil water content, vine photosynthetic activity, shoot pruning weight, grape yield and quality are investigated in a vineyard in Lower Austria over three consecutive seasons. Thereby, we hypothesise that a permanent under-vine vegetation cover, either mowed or without mowing, supports the soil microbial communities and soil functions in a way to enhance water and nutrients availability for vines which partly compensates for the competition of the growing vegetation. Our results confirm effects on the soil water balance, more specifically, a reduced soil water content in 11-20 cm soil depths induced by a permanent vegetation cover as compared to herbicide application or soil tillage. Further consequences of permanent vegetation below vines were lower shoot pruning weights and lower berry weights, while total soluble solids and titratable acidity were not affected. The vine's photosynthetic activity, as well as the soil water content, were partly affected by treatments dependent on the precipitation ahead of the measurement. In parallel, the soil microbial activity was significantly enhanced by a permanent vegetation cover below the vines as compared to herbicide application, a trend which increased with the years of the project. In conclusion, permanent under-vine vegetation strongly promoted soil microbial activity without strong effects on shoot pruning weight, grape yield and quality. In the next step, a functional proof is necessary to characterise the interaction between soil microbial activity, soil water balance and vine nutrition and water status by using sensors for continuous measurements.

**KEYWORDS:** soil microbial activity, spontaneous vegetation, soil water balance, soil tillage, vineyard sustainability

## INTRODUCTION

Vineyards are highly managed agroecosystems and inter-row as well as under-vine management affects soil properties and soil functions alike. Recently, ecosystem services and disservices of cover crops in vineyards inter-rows were reviewed (Garcia *et al*., 2018), summarising the importance of cover crops in weed control (Steenwerth *et al*., 2016), pest and disease management (Paredes *et al*., 2021), water availability (Blanco-Canqui *et al*., 2015), biodiversity in soils (Burns *et al*., 2016; Winter et al., 2018), carbon sequestration or promotion of soil organic matter content and soil enzyme activity (Gattullo *et al*., 2020; Novara *et al*., 2020) as well as field trafficability. These aspects affect soil functions, e.g. aggregate stability (Six *et al*., 1999), avoidance of soil erosion and nitrogen leaching (Gaudin *et al*., 2010) or enhancement of water infiltration (Coniberti *et al*., 2018b). Apart from these positive effects, the competition for water and nutrients is considered as a major ecosystem disservice related to cover crops or permanent vegetation in vineyard inter-rows resulting in reduced vine growth, lower yield and altered fruit composition (Coniberti *et al*., 2018a; Garcia *et al*., 2018; Pou *et al*., 2011). Nevertheless, general conclusions on the benefits and disadvantages of vine growth and grape quality of cover crops or spontaneous vegetation in vineyard inter-rows as well as below vines are difficult to draw as specifically pedo-climatic conditions as well as the duration, the intensity of the vegetation cover and the plant species composition could be influencing factors (Steiner *et al*., 2021; Vanden Heuvel and Centinari, 2021). Apart from cover crops, soil tillage and herbicide application are among the repertoire of inter-row and under-vine management methods (Guerra and Steenwerth, 2012; Abad *et al*., 2020). Soil tillage incorporates plant residues into the soil leading to fast degradation of organic matter and its mineralisation through soil microorganisms which represents a source for plant available nutrients (Abad *et al*., 2021b; Curtright and Tiemann, 2021; Verdenal *et al*., 2021). Major drawbacks of soil tillage could be a depletion of soil organic matter and an increase in soil susceptibility to soil erosion in non-arid climates (Abad *et al*., 2021b; Ruiz-Colmenero *et al*., 2011). The application of herbicides to control weeds in vineyard inter-rows or under vines has led to an intense discussion on the toxicity of the herbicides, the risk for the development of herbicide resistance and the negative impacts on the environment and vines as, e.g., reduced grapevine root mycorrhisation and altered soil microbial communities (Chou *et al*., 2018; Zaller *et al*., 2018). As a consequence, cover crops or spontaneous vegetation in inter-rows in vineyards have been promoted by wine growers, stakeholders and consumers likewise, and research has been intensified (Abad *et al*., 2021b; Coniberti *et al*., 2018b; Hickey *et al*., 2016). Many soil ecosystem services and functions are provided by soil microbial communities, which are promoted by cover crops (Curtright and Tiemann, 2021; Fierer, 2017). Recently, sequencing technologies became applicable in eco-physiological studies, enhancing the knowledge of the soil microbiome (Nkongolo and Narendrula-Kotha, 2020). These metagenome studies provide a snapshot of the genetic structure and diversity of soil microbial communities but lack the potential to follow dynamic changes in their growth and functions (Burns *et al*., 2016). Functional indicators are needed to assess the effects of either environmental stressors or management changes on the activity and functions of microbial communities in time and direction. Among the parameters to monitor soil-born processes are soil microbial biomass, soil respiration and soil hydrolytic enzymes (Nkongolo and Narendrula-Kotha, 2020). Soil microorganisms use hydrolytic enzymes to mediate and catalyse soil functions, e.g., the decomposition of organic residues to provide plant available nutrients or to transform them to enhance the soil organic matter and humus content in the upper soil (Curtright and Tiemann, 2021; German *et al*., 2011). A fast and cost-effective manner to assess the hydrolytic soil enzyme activity is the usage of fluorometric-based assays which can be upscaled to high-throughput systems on 96 well plate formats (Deng *et al*., 2013).

Here, we study different under-vine management treatments (application of herbicides (H), soil tillage with rotary tillage (RT), soil tillage with an under-vine weeder (UVW), spontaneous vegetation mowing (CCM, complete cover mowing), spontaneous vegetation NO mowing (CC, complete cover)) in one vineyard in Lower Austria and monitor the effects on important soil functions and grapevine parameters. The specific objectives of the case study are: 1) to analyse the soil microbial activity by using hydrolytic soil microbial enzymes, 2) to quantify the effects of under-vine management on vine photosynthesis, grape yield and grape quality and 3) to investigate the water balance in shallow soil depth. Thereby we hypothesise that a permanent under-vine vegetation cover enhances the soil microbial activity, which partly counteracts the competition of vines for available nutrients.

## MATERIALS AND METHODS

### 1. Experimental vineyard

The experimental vineyard is located in Lower Austria, near Langenlois ("Ried Rosenhügel" 48°27'59.8"N 15°39'55.1"E, 219 m a.s.l.) and is managed by the Applied School of Viticulture and Pomology Krems (Wein- und Obstbauschule Krems). The soil type is a chernozem on loess with a loamy silt soil texture (12 % sand, 70 % silt, 18 % clay) and an alkaline pH of 7.5 and a content of soil organic matter (SOM) of 7-8 %. The climatic conditions from 2018, 2019 and 2020 on the basis of the yearly mean temperature and precipitation were: 11.4 °C, 450 mm; 12.2 °C, 305 mm and 10.8 °C, 480 mm (Figure S1). In 2008, the vineyard was planted as the experimental site of the University of Natural Resources and Life Sciences, Vienna, as a split-plot system with two clones of Pinot noir (18 Gm and 1-84 Gm) grafted on the rootstock Kober 5BB (*V. riparia* × *V. berlandieri*) and planted with a vine distance of 105 cm and inter-row width of 2.5 m. Clone 18 Gm developed stunted shoot growth

showing a growth depression of unknown nature in 2018, the first year of the experiment independent of the treatments. Therefore, all measurements on vines and grapes in 2018 and the following years were performed with clone 1-84 Gm solely, and soil samples for soil enzyme analyses and soil water content were obtained from blocks A and B, vines 1-30 and 31-60 per row, due to its higher level of homogeneity (Table S1). A vertical shoot position (VSP) trellis system is established and vines are pruned with bilateral canes with a yield expectation of 9000 kg per hectare. Plant protection reschemes follow standardised integrated production rules. Inter-row management was performed in 2019 and 2020 as complete and permanent vegetation coverage of spontaneous vegetation, with a high percentage of grass species (80 % ± 17) and herbaceous species (20 % ± 16) as determined in a previous study from 2015-2017 (Griesser *et al.*, 2022). In 2018 and the previous years, alternating inter-row management was applied with tillage of every second inter-row swapped every 2-3 years. This inter-row management by soil tillage was performed three times during the growing season (2018 soil tillage was performed on the 3rd of May, 6th of June and 3rd of July). In 2018 and 2019, the vegetation below vines in CCM and CC treatments was determined in 2 squares with 2 × 0.5 m per row (23rd May 2018, 21st May 2019). We observed a high percentage of grass species (35-75 % of the vegetation coverage), mainly *Bromus* ssp.; herbaceous species (3-15 %) and typical weed species (2-18 %) with species *Elymus repens*, *Cirsium arvense*, *Convolvulus arvensis* and *Taraxacum officinale*. Inter-rows had a similar species composition, although this was not specifically determined. In all three years of the experiment, inter-rows with vegetation were mowed end of May, end of June, end of July and end of August.

## 2. Under-vine management treatments

The experiment with five different under-vine treatments was established in 2018 and was continued for three years (until 2020). The five under-vine management treatments were: herbicide application (H) (Roundup Ultra, Monsanto Agrar Deutschland, Düsseldorf, Germany, 5 L $ha^{-1}$, concentration 360 g $L^{-1}$, application dates see Table S2), soil tillage with rotary tillage (RT) (Alm Roller Weeder 4S, Adelheim Landtechnik Maschinenbau, Nordheim, Germany, working speed 7 km/h) soil tillage with under vine weeder (UVW) (Radius SL plus rotary tiller, Clemens Technologies, Wittlich, Germany, working speed 2.5 km/h) spontaneous vegetation mowing (CCM, complete cover mowing), spontaneous vegetation NO mowing (CC, complete cover) (Figure 1, Figure S2). In total, four replicates per treatment were distributed within 20 vine rows (subplots) of the experimental vineyard as a split-plot design was not possible due to the block arrangement of Pinot noir clones (Table S1). The under-vine management treatments followed a local schedule with recommended two times herbicide application and three times soil tillage within each vegetation period to investigate local strategies in comparison. The mowing of the complete cover mowing (CCM) treatment was performed four times per vegetation period in accordance with the mowing of the vegetation in the inter-rows end of May, end of June, end of July and end of August.

## 3. Fluorometric-based assay for soil enzyme activity and gravimetric soil water content

A modified fluorescence-based method in microplate format was used to determine the enzymatic activity of the extracellular soil hydrolytic enzymes leucine aminopeptidase (LAP), acid phosphatase (AP) and β-1,4-glucosidase (BG) (Dick *et al*., 2018; German *et al*., 2011). In short, soil samples from all five under vine treatments were collected with a soil core borer from a soil depth of 0-10 cm (2.5 cm × 10 cm length) for soil enzymatic assays and gravimetric soil water content as well as from 11-20 cm for gravimetric soil water content only. Per vine row, six samples were pooled to obtain a mixed probe for further analyses in the laboratory (N = 4 samples per sampling date and treatment). Samples were collected on: 9th May 2018 (7 days after treatment, DAT), 16th June 2018 (10 DAT), 9th July 2018 (6 DAT), 25th July 2018 (22 DAT), 2nd May 2019 (21 DAT), 12th June 2019 (7 DAT), 26th June 2019 (21 DAT), 30th July 2019 (7 DAT), 6th April 2020 (-15 DAT), 4th May 2020 (10 DAT), 3rd June 2020 (40 DAT), 30th July 2020 (1 DAT). Soil samples were kept cooled (6–8 °C) for transport and soil enzyme activity

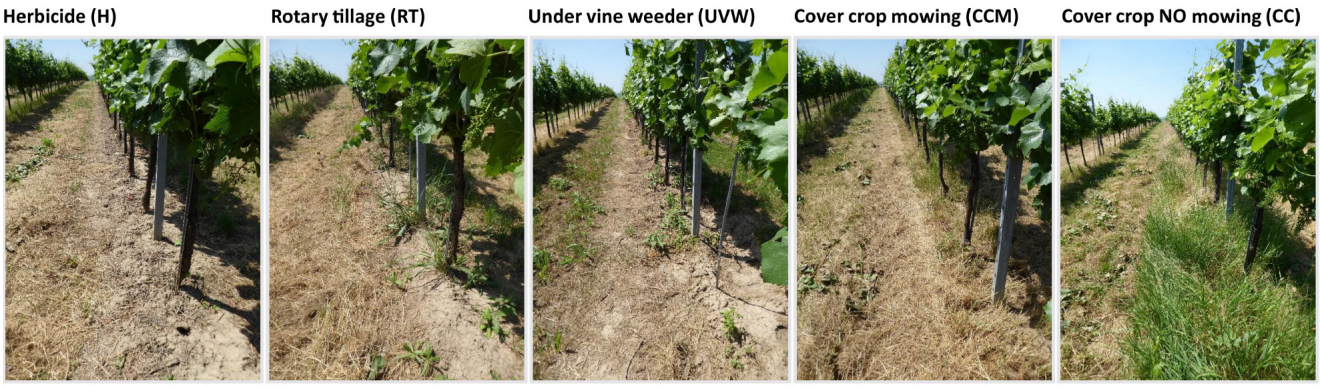



**FIGURE 1.** Under-vine management treatments.

Pictures were taken on the 26th of June 2019 after a dry period in spring. Treatments herbicide application (H), soil tillage (rotary tillage (RT), under-vine weeder (UVW)), permanent vegetation (complete cover mowing (CCM) and complete cover NO mowing (CC)) were established in spring 2018.

was determined within the next 2-3 days in the laboratory. All substrates and standards were purchased from Sigma Aldrich: MUB standard (4-Methylumbelliferone, M1381), AMC standard (7-Amino-4-methylcoumarin, A9891), substrate β-1,4-glucosidase (4-Methylumbelliferyl-β-D-Glucopyranoside, M2633), substrate acid phosphatase (4-Methylumbelliferyl phosphate, M8883), substrate leucine aminopeptidase (L-leucine-7-amino-4-methylcoumarin hydrochloride, L2145). Stock solutions and aliquots were prepared of standards MUB (100 µM), AMC (1000 µM) and all substrates (200 µM). Aliquots were stored at -20 °C and defrosted before each analysis. Standard curves for MUB were in the range of 0, 5, 10, 15, 20 and 25 µM and AMC for 25, 50, 75, 100 and 125 µM. The soil suspension was prepared with 1 g of soil and 125 mL of 50 mM TRIS buffer adjusted to the natural pH of the soil (pH = 7.5) under constant stirring inside a 150 ml glass beaker. Incubation in an ultrasonic bath at room temperature for 1 min did facilitate enzyme extraction. 200 µl of the soil suspension was used for standard curves as well as sample assay reactions with substrates. Reactions did start by adding 50 µl of standard or substrate solutions just before incubation (BG: 2 h, AP: 3 h, LAP: 20 h) in darkness to obtain a final reaction volume of 250 µl. The analyses were performed in black 96-well plates, including substrate blanks and soil slurry blanks. Reactions for standard curves were performed in duplicates, while four technical repeats were used for sample assay reactions. After incubation, the fluorescence was measured using a multiplate reader (FLUOstar Omega, BMG Labtech, Ortenberg, Germany) with an excitation wavelength of 360 nm and an emission wavelength of 460 nm with the same gain adjustment for standard and assay plates. Fluorescence signals from standards and assays were used to calculate the enzymatic activity as nmol substrate degradation per $g^{-1}$ dry soil within one hour of incubation, as previously described (German *et al*., 2011). The gravimetric soil water content was determined from the same samples collected at 0-10 cm and 11-20 cm soil depth by transferring approximately 30 g of wet soil to small paper bags and keeping them at 70 °C for five days to achieve constant weights. Dry and wet soil weights were used to calculate the gravimetric soil water content of each soil sample.

## 4. Grapevine measurements

The grapevine photosynthetic activity was measured with a portable infrared gas analyser (IRGA system, LCPro-DC (ADC BioScientific Ltd., Herts EN11 ONT, United Kingdom)) using a standardised closed air chamber. Measurements were performed during dry periods over the three years of the experiment, more specifically on the 23rd May 2018, 1st August 2018, 18th June 2019, 26th June 2019, 30th July 2020 and 12th August 2020 on six vines per subplot (vine row) on two fully developed leaves above the cluster zone between the 6th to 8th insertion (N = 12 measurements per vine row). Measurements pre-setting's were: ambient $CO_2$ levels and standardised light conditions of 1000 µmol $s^{-1}$ $mm^{-2}$ on sun-exposed leaves. The parameters assessed were: photosynthetic rate (A, µmol $CO_2$ $m^{-2}$ $s^{-1}$), respiration rate (E, mmol $H_2O$ $m^{-2}$ $s^{-1}$) and stomatal conductance (gs, mol $H_20$ $m^{-2}$ $s^{-1}$).

Grape berries (between 250 and 300 berries) were randomly collected from different grape clusters and different grape zones within each treatment and repetition prior to harvest (BBCH89; 27th August 2018, 9th September 2019, 17th September 2020) in each year from Pinot Noir clone 1-84 Gm. Collected grape berries in plastic bags were transported to the laboratory under cold condition (6-8 °C) and grape juice was obtained by squeezing berries within the plastic bag manually. Further on, the juice was filtered (Whatman filter 520 A ½, VWR International GmbH, Vienna, Austria) and centrifuged for 5 min at 3500 rpm in a 50 ml Falcon tube. The clarified juice was analysed via Fourier-Transformation-IR-Spectroscopy (FTIR, OnoeFOSS, FOSS GmbH, Hamburg, Germany) to obtain total soluble solids (TSS, °Brix), total titratable acidity (TA, g Tartaric acid $L^{-1}$) and yeast assimilable nitrogen (NOPA, mg $L^{-1}$) as alpha-amino nitrogen and ammonia. Additionally, the berry weight (N = 10) was determined from the collected grape berries.

In the vineyard, the yield was estimated by determining the weight of the clusters of five consecutive vines (kg per vine) per treatment and replicated within each study year. Additionally, the shoot pruning weight of five consecutive vines (g per vine) per treatment and repetition for each year was determined in winter. Unfortunately, both measurements were not determined on the same vines.

## 5. Statistical analyses

All statistical analyses were performed according to standardised procedures using the software program SPSS 27 (IBM SPSS Statistics, New York, USA) and R version 4.0.3 (R Core Team, 2020). More specifically, data evaluation, boxplots to visualise data and One-Way ANOVA was performed with SPSS, while regression models and their visualisation with scatter plots and effects plots were performed in R, including the packages "lme4" (Bates *et al*., 2015), "ggplot2" (Wickham, 2016), "effects" (Fox and Weisberg, 2019), "performance" (Lüdecke *et al*., 2021), "interactions" (Long, 2019) and "stats" (R Core Team, 2020). To account for heteroscedasticity, robust standard errors were calculated with functions vcov.fun = "CR", vcov.type = "CR1 of the R package "clubSandwich" (Pustejovsky, 2021). Data (soil enzyme activity, plant photosynthesis) were standardised within each sampling date (z-scores) to visualise treatment effects across collection dates and years. In linear mixed models (LMM), the random factors sampling dates within year (1|year/sampling date) and sampling dates (1|sampling date) were used to account for repeated measurements. Model comparison was performed with "performance" and factor comparisons within the linear mixed models were performed with "emmeans" (Lenth, 2021) and a Bonferroni correction for multiple testing.

## RESULTS

### 1. Soil enzyme activity is affected by under-vine treatments

Hydrolytic soil enzyme activity was determined at twelve sampling dates during the study period (Table S3). The activity of acid phosphatase (AP) did not change within each treatment and year between sampling dates (Figure S3A), while the activity of β-glucosidase (BG) was influenced by the season in all three years (Figure S3B). Especially we observed an increase in activity between the first sampling date (9th May 2018) and the fourth (25th July 2018) in both soil tillage treatments (RT, UVW) in 2018. In contrast, in the third year of the study, the activity of BG was high in spring and decreased in all treatments until the last sampling date (30th July 2020). Further on, the activity of leucine aminopeptidase (LAP) was enhanced between sampling dates in the first year in treatments RT, UVW and CCM, while LAP was not influenced by the time of sampling in 2019 and 2020.

The direction of the response of acid phosphatase (AP), β-glucosidase (BG) and leucine amino peptidase (LAP) activity to the under-vine treatments were comparable (Figure 2A–C, standardised results within each sampling date). In the first year of the experiment, especially soil tillage with the under-vine weeder (UVW) was negatively impacting the activity of all three enzymes, while differences between other treatments were marginal. In 2019 and 2020, the activity of all tested soil enzymes increased in both vegetation treatments (CCM, CC) as compared especially to the herbicide application (H) and soil tillage (UVW), while rotary tillage (RT) showed intermediate values (Figure 2).

Linear mixed regression models for each tested soil enzyme were performed with sampling dates within years as a random factor to account for repeated measurements to determine the influencing factors (Table S4). The ANOVA results of the linear mixed models confirmed for all three enzymes a highly significant influence of the treatments (AP: $F(4, 194) = 17.2801$, $p < 0.001$; BG: $F(4, 196) = 23.3937$, $p < 0.001$; LAP: $(4, 195) = 16.7342$, $p < 0.001$). Additionally, an increase in the gravimetric soil water content in 11-20 cm soil depth enhanced the activity of β-glucosidase and leucine aminopeptidase by approximately 3 nmol $g^{-1}$ $h^{-1}$ (Table S4; ANOVA results: BG: $F(1, 197) = 8.8771$, $p = 0.009$; LAP: $F(1, 199) = 7.1164$, $p = 0.010$; AP: $p = 0.9424$), while AP was not influenced by the soil water content (11-20 cm). Additionally, the gravimetric soil water content in 0-10 cm soil depth had no influence on the soil enzyme activity (Table S4). Although there is an impact of the gravimetric soil water content in 11-20 cm soil depth on BG and LAP activity, the effect sizes of the l mm confirmed a much stronger influence of the factor treatment (BG partial $\eta^2 = 0.26$; LAP partial $\eta^2 = 0.33$) as compared to the soil water content (BG partial $\eta^2 = 0.09$; LAP partial $\eta^2 = 0.04$). Pairwise comparisons of treatments within the linear mixed regression models showed for all three tested soil enzymes (Table 1) the lowest activity after herbicide application (H) and soil tillage with the under vine weeder (UVW), and the highest activity in complete cover mowing (CCM) followed by complete cover (CC) and rotary tillage (RT) when the variability due to the year of measurement and the sampling dates were excluded. A One-Way ANOVA of standardized data analysing only the fixed factor treatment confirmed the results for AP and LAP, while for BG three groups were identified (AP: $F(4, 231) = 20.867$, $p > 0.001$; BG: $F(4, 232) = 23.562$, $p > 0.001$, Tukey post hoc test a: RT, CCM, CC; b: H; c: UVW; LAP $F(4, 233) = 17.092$, $p > 0.001$).

### 2. Effects of under-vine treatments on soil water content and vine photosynthetic activity in relation to precipitation

The gravimetric soil water content in 0-10 cm and 11-20 cm soil depth was determined from the soil samples collected for the soil enzyme activity (Table S3, Figure S4). Absolute values differ between sampling dates, therefor linear mixed models with the random factor sampling dates (1|sampling dates) were set to analyse the influence of the

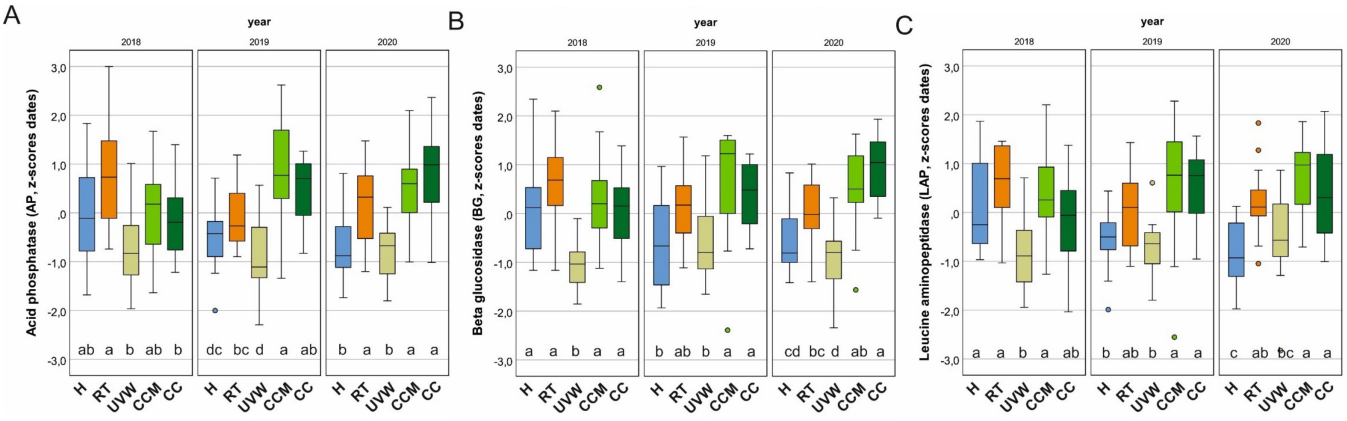



**FIGURE 2.** Soil hydrolytic enzyme activity.

Results are shown as standardised data within each sampling date (z-scores) for A) acid phosphatase (AP), B) β-glucosidase (BG) and C) leucine amino peptidase (LAP) in the years 2018-2020 for the different under-vine management treatments. Differences between treatments are indicated by different letters (One-Way ANOVA, Tukey post-hoc test, $\alpha < 0.05$, N = 16 per treatment and year).

**TABLE 1.** Treatment influence on soil enzyme activity.

| | Acid phosphatase (AP) | β-glucosidase (BG) | Leucine aminopeptidase (LAP) |
|---|---|---|---|
| Herbicide (H) | 65.1 ± 18.4 b | 109.6 ± 18.4 b | 181.1 ± 7.1 b |
| Rotary tillage (RT) | 81.4 ± 23.0 a | 127.4 ± 18.0 a | 203.2 ± 7.1 a |
| Under vine weeder (UVW) | 56.8 ± 16.0 b | 97.8 ± 18.2 b | 177.1 ± 7.0 b |
| complete cover mowing (CCM) | 87.6 ± 24.7 a | 139.8 ± 18.3 a | 216.4 ± 7.3 a |
| complete cover NO mowing (CC) | 82.3 ± 23.2 a | 137.1 ± 18.2 a | 209.3 ± 7.3 a |

Summary results from pairwise comparisons of treatments for the enzymes acid phosphatase (AP), β-glucosidase (BG) and leucine aminopeptidase (LAP) within the linear mixed models including treatment as a factor, the continuous variables gravimetric soil water content 0-10 cm and 11-20 cm soil depth and the random factor (1|year/sampling dates). Presented values are mean values ± standard errors obtained via emmeans with Bonferroni correction for multiple testing. Significant differences are shown with different letters.

factor treatment and the precipitation (as three categorial factors (0-10 mm, 11 20 mm, > 20 mm)) 7 and 14 days prior to sampling (Table S5a, Table S5b). Multiple model comparisons identified the factor treatment in combination with the precipitation 14 days prior to sampling as the best model explaining our data.

The soil water content in 0–10 cm was weakly influenced by the treatment ($F(4, 216) = 2.5001$, $p = 0.047$; partial $\eta^2 = 0.05$) and the precipitation (14 days) ($F(1, 219) = 6.3505$, $p = 0.048$; partial $\eta^2 = 0.37$). The soil water content increased by about 3 percent points between precipitation categories (0–10 mm, 11–20 mm, >20 mm) in all treatments, while rotary tillage (RT) lowered the soil water content by about 1 percent point as compared to herbicide application (Table S5a, Figure 3A). In contrast, the soil water content in 11–20 cm soil depth was much stronger influenced by the under-vine treatments ($F(4, 196) = 6.9318$, $p < 0.001$; partial $\eta^2 = 0.13$), while no influence of the precipitation was determined ($F(1, 199) = 1.5474$, $p = 0.2487$; partial $\eta^2 = 0.16$) (Figure 3B, Table S5a, b). Treatments rotary tillage (RT), complete cover mowing (CCM) and complete cover (CC) reduced the soil water content (11-20 cm) between 1.5-1.7 %-points (Figure 3B, Table S5a) as compared to herbicide application.

Pairwise comparisons with the mixed model results (Table 2) gave no significant differences between treatments for soil water content in 0-10 cm, while One-Way ANOVA of standardised data identified two groups for treatments (GW10: $F(4, 215) = 3.123$, $p = 0.016$; a: H, ab: UVW, CCM, CC, b: RT). The soil water content in 11–20 cm soil depth was highest in H and lowest in RT, CCM and CC, as shown

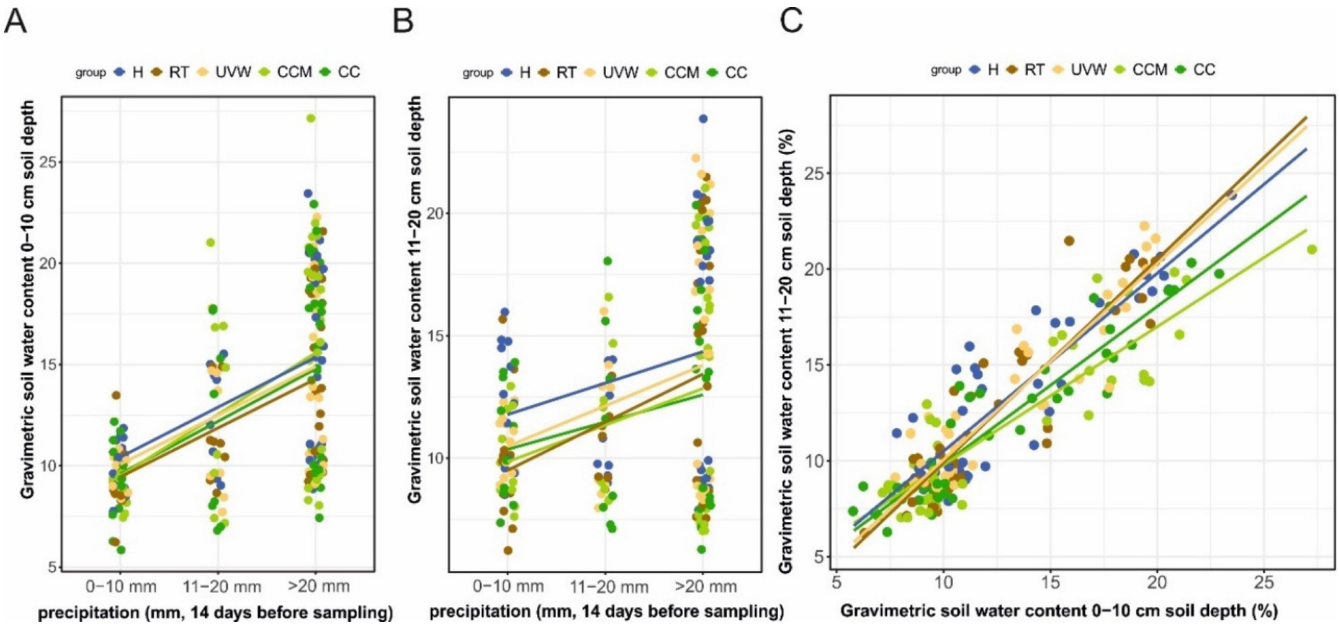



**FIGURE 3.** Soil water content.

Effect plot of linear regression models of A) the gravimetric soil water content in 0-10 cm with the precipitation 14 days prior to sample collection, B) the gravimetric soil water content in11-20 cm soil depth with the precipitation 14 days prior to sample collection and C) the relationship between the gravimetric soil water content 0-10 cm and 11-20 cm soil depth for each under-vine treatment. Data collected at twelve sampling dates in 2018-2020 are shown, and trend lines represent effects for the individual treatments in the linear mixed model.

**TABLE 2.** Treatment influence on soil water content and vine photosynthetic activity.

| | Soil water content 0–10 cm | Soil water content 11–20 cm | Assimilation rate (A) | Stomatal conductance (gs) |
|---|---|---|---|---|
| herbicide (H) | 13.7 ± 1.2 | 13.5 ± 1.3 a | 12.7 ± 0.67 | 0.174 ± 0.022 |
| rotary tillage (RT) | 12.7 ± 1.3 | 12.1 ± 1.3 b | 12.8 ± 0.68 | 0.180 ± 0.023 |
| under vine weeder (UVW) | 13.3 ± 1.2 | 12.7 ± 1.2 ab | 12.6 ± 0.68 | 0.174 ± 0.021 |
| Complete cover mowing (CCM) | 13.5 ± 1.4 | 11.9 ± 1.1 b | 12.5 ± 0.67 | 0.172 ± 0.021 |
| Complete cover NO mowing (CC) | 13.1 ± 1.3 | 11.9 ± 1.2 b | 12.5 ± 0.66 | 0.175 ± 0.022 |

Summary results from pairwise comparisons of treatments on the soil water content in 0-10 cm and 11-20 cm soil depth and the assimilation rate (A) and stomatal conductance (gs) within the linear mixed models including treatment as a factor, the precipitation (as three categorial factors (0-10 mm, 11-20 mm, > 20 mm)) 7 and 14 days prior to sampling and the random factor (1| sampling dates). Presented values are mean values ± standard errors obtained via emmeans with Bonferroni correction for multiple testing. Significant differences are shown with different letters.

by the pairwise comparison and confirmed by ANOVA of standardised data (GW20: $F(4, 195) = 7.744$, $p < 0.001$; a: H, ab: UVW, b: RT, CCM, CC) (Table 2). We further analysed the linear relationship between the soil water content from both soil depths for each treatment (Figure 3C). There is a shift in the slope of the relationship for both vegetation cover treatments (CCM, CC), indicating a reduced soil water content in 11-20 cm soil depth in these treatments when compared with the same soil water content level in 0–10 cm soil depth with herbicide application or soil tillage (RT, UVW).

The photosynthetic activity was determined at six measurement dates between 2018–2020 and absolute values of assimilation rate (A) and stomatal conductance (gs) varied strongly between measurement dates (Table S6, Figure S5). Within measurement dates, a significant influence on the assimilation rate was obtained on the 26th of June 2019 and the 12th August 2020 and for stomatal conductance on the 26th of June 2019. The direction of response to the applied treatments was different between these two days (a decrease in CCM and CC on the 26th of June 2019 and an increase in CC on the 18th of August 2020), indicating that other factors may interact. Linear mixed models with sampling dates as random, treatment as fixed factor and precipitation as a categorical variable confirmed an interaction between the under-vine treatments and the precipitation 7 days prior to measurements (Table S7; $\alpha = 0.1$). The interaction was observed for both, the assimilation rate (treatment: rain7d: $F(4, 292) = 3.1828$, $p = 0.014$) and stomatal conductance (treatment: rain7d: $F(4, 292) = 2.2790$, $p = 0.061$; log-transformed data). The summary of the coefficients of the linear mixed model showed an influence of the precipitation, while the ANOVA result of the same model gave no significant result for the assimilation rate ($F(1, 295) = 3.7251$, $p = 0.125$; partial $\eta^2 = 0.48$) and a weak significant ($\alpha = 0.1$) result for the stomatal conductance ($F(1, 295) = 5.8717$, $p = 0.07$; partial $\eta^2 = 0.59$) (Table S7, Figure 4A,B). The effect size of precipitation on both measured parameters was high.

The ANOVA results for treatments were not significant, as well as the pairwise comparisons (Table 2), supporting the illustrated effect plots (Figure 4A,B) that a general interpretation of the effects of treatments independent of the precipitation is not conclusive. Lower values for A and gs are observed in both vegetation cover treatments (CCM, CC) at higher precipitation as compared to H, UVW and RT, while no differences were observed at lower precipitation until 20 mm (Figure 4A,B; Figure S6).

### 3. Effects of under-vine treatment on grape quality and vine parameters

Total soluble solids (TSS), titratable acidity (TA) and yield per vine were not affected by the under-vine treatments (Figure 5, Table S8). Additionally, a higher grape nitrogen content (NOPA) was observed after herbicide application as compared to the other treatments, but the effects were not significant (Figure 5C). The treatments of complete vegetation mowing (CCM) and complete vegetation (CC) resulted in lower shoot pruning weight and lower berry weight as compared, especially to the herbicide application and rotary tillage in the case of berry weight.

## DISCUSSION

Climate change forces farmers to reconsider and modify vineyard management strategies in a sustainable manner to ensure adequate yield and grape quality while enhancing vineyard resilience by supporting ecosystem services. Inter-row and under-vine management with cover crops or spontaneous vegetation can introduce a higher level of biodiversity in vineyards and support ecosystem functions (Abad *et al*., 2021a). Herein we report a three-year case study in Austria, a cool climate region, to investigate the effects of five under-vine management treatments on soil enzymatic activities and soil water content as well as vine photosynthesis and grape yield and quality.

It is well known that biodiversity and associated ecosystem services are supported by a vegetation cover in vineyards, either in inter-rows or below the area of the vine (Burns *et al*., 2016; Geldenhuys *et al*., 2021; Guzman *et al*., 2019; Paiola *et al*., 2020; Zanettin *et al*., 2021; Abad *et al*., 2020). Especially the soil microbiome is promoted, which supports manifold ecosystem functions (Curtright and Tiemann, 2021). In the presented case study, we determined an enhanced soil enzyme activity (leucine aminopeptidase

(LAP), acid phosphatase (AP) and β-glucosidase (BG)) in under-vine treatments with vegetation cover (both mowed or not mowed) and rotary tillage (RT) as compared to soil tillage by under vine weeder (UVW) or herbicide application, confirming previous studies in table grape vineyards (Gattullo *et al*., 2020). Additionally, organic and biodynamic vineyard management has a positive effect on soil microbial activity (Di Giacinto *et al.*, 2020); both systems are characterised by a high percentage of vegetation coverage in inter-rows on a temporal and spatial scale. In the current study, we could not specifically determine the causes for the observed differences in both soil tillage treatments; however, the intensive tillage of soils with the rotary tillage may have played a role. Previous studies underline the complexity as contradicting results for bacterial and fungal communities have been reported (Blanco-Canqui and Wortmann, 2020; Burns *et al*., 2016; Li *et al*., 2020; Novara *et al*., 2020). The microbial species community itself was not characterised in our study, but the soil enzyme activity could reflect dynamic changes in soil microbial activity. Additionally, we observed an influence of the period of soil sampling for the analyses for the enzymes β-glucosidase (BG) and leucine aminopeptidase (LAP), while the acid phosphatase (AP) was not influenced. The activity of β-glucosidase (BG) and leucine aminopeptidase (LAP) increased with increasing soil moisture, a result which is confirmed by a study modelling the activity of soil enzymes with different temperature and soil moisture regimes (Steinweg *et al.*, 2012). By testing different soil conditions, the study determined that β-glucosidase mainly responded to increasing temperatures, except when the soil was dry. Re-moistening of dry soils enhanced the activity, while the response in ambient or wet soils was less pronounced (Steinweg *et al.,* 2012). A positive correlation between BG and LAP to higher precipitation has also recently been determined in a shrub-encroached grassland in Mongolia (Akinyemi *et al.*, 2020). The function of β-glucosidase in the soil is in carbon cycling by cellulose degradation through hydrolysing the cellobiose residues and it can be used as an indicator for the presence of higher simple sugars for soil microbial populations and as an indicator of effects of soil management practices due to its correlation with the soil organic matter content (reviewed by Tiwari *et al*., 2019). Our study determined the under-vine treatments as major drivers of the soil enzyme activity and, to a lower extent, the soil water content. Thereby we confirm the suitability of soil hydrolytic enzymes to assess management-associated changes in soil microbial communities in vineyards.

Water availability and water balance are essential to sustain plant growth. We observed a shift in the relationship of the gravimetric soil water content until 10 cm and 20 cm soil depth under permanent under-vine vegetation. Under-vine vegetation reduced the soil water content in 10-20 cm soil depth to a greater extent than herbicide application or soil tillage at comparable soil water content in 0-10 cm soil depth. Analyses of the soil water content in 11-20 cm accounting for the variability through sampling dates, confirmed significant lower values due to rotary tillage (RT), complete vegetation mowed (CCM) and complete vegetation (CC). Lower soil water content in 35 cm soil depth has been observed in Spain with a permanent cover crop (*Brachypodium distachyon*) (Marques *et al*., 2020), while other studies observed higher water retention in a cover-cropped table grape vineyard in Southern Italy (Torres *et al*., 2017). The study in Spain could determine a seasonal influence, as the reduced soil water content was mainly observed in spring when growing

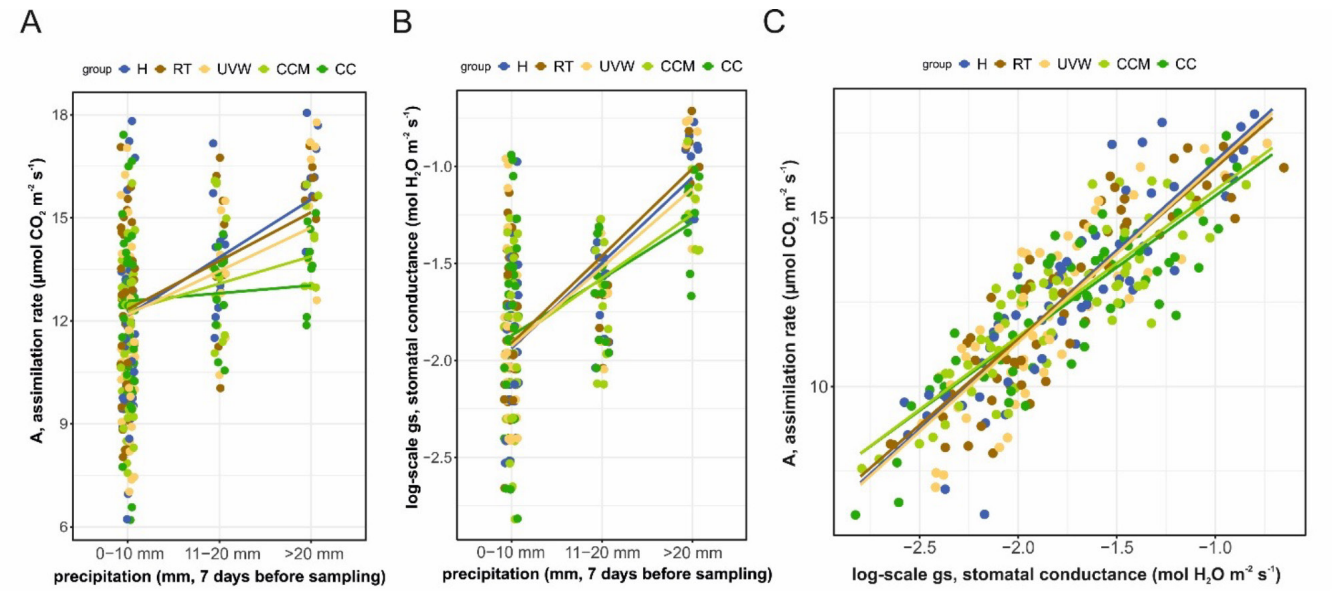



**FIGURE 4.** Photosynthetic activity.

Effect plot of linear regression models of A) the photosynthesis assimilation rate in relation to the precipitation 7 days prior to sample collection, B) the stomatal conductance in relation to the precipitation 7 days prior to sample collection and C) the linear relationship between the assimilation rate and the stomatal conductance for each under-vine treatment. Results were obtained at six sampling dates from 2018-2020, which were used as random factors in mixed models.

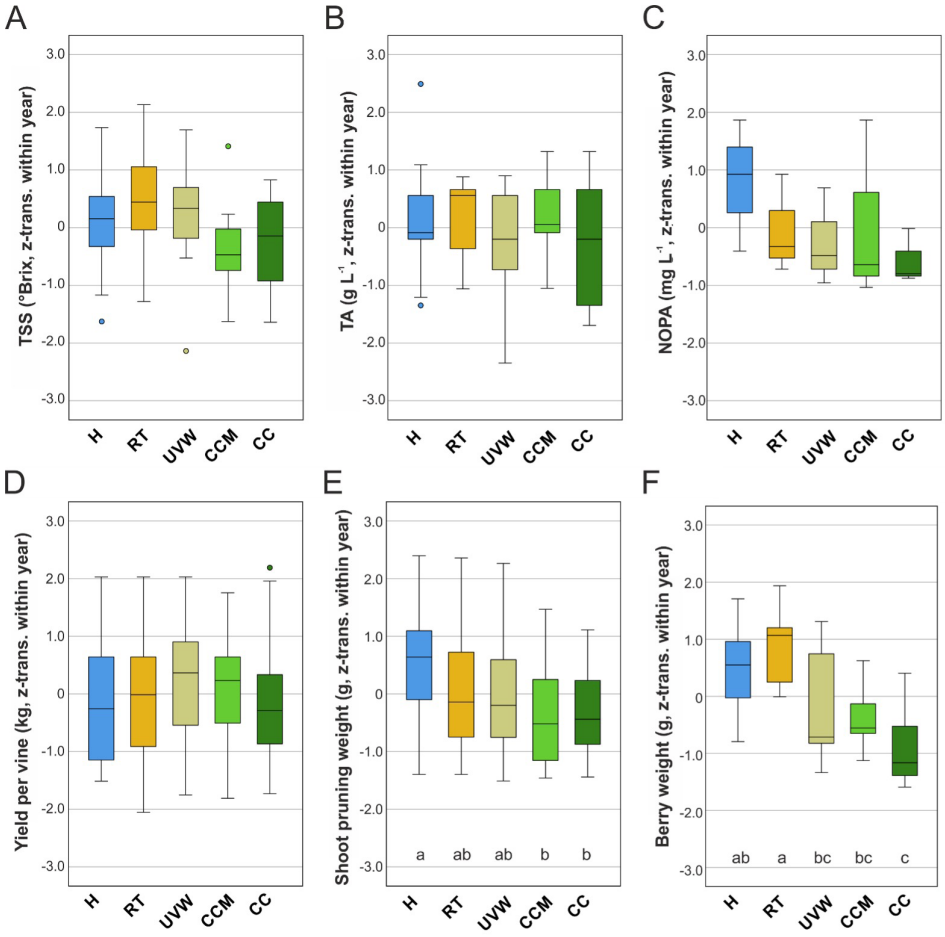



**FIGURE 5.** Grape quality, yield and pruning weight.

Summarised results of grape quality (total soluble solids, titratable acidity, NOPA, berry weight), yield and pruning weight determined in 2018–2020 with Pinot Noir clone 1-84Gm. A) Total soluble solids (TSS, °Brix, N = 45), B) Titratable acidity (TA, g L-1, N = 45), C) Yeast assimilable nitrogen (NOPA, mg L-1, N = 15), D) Yield per vine (kg, N = 45), E) Shoot pruning weight (g per vine, N = 135), F) berry weight (g per berry, N = 45). Data were standardised within each year to analyse under-vine treatment effects independent of the seasonal effect with One-Way ANOVA ($\alpha < 0.05$) and Tukey post-hoc test. Significant differences between treatments are indicated with different letters.

cover crops consume more water (Marques *et al*., 2020). This seasonality in the effects of different under-vine management on the soil volumetric water content was also observed in a study in New York state, an area with ample precipitation (Karl *et al*., 2016). The study determined lower soil water contents in areas with vegetation as compared to tillage and herbicide application in spring and early summer, while the differences in soil water content diminished later in the season, approximately at the start of berry ripening. We did observe a similar trend, a negative correlation between the soil water content (0–10 cm) and the percentage of vegetation cover in May 2019 ($R^2$ = –0.603), while in June 2019, no correlation was determined ($R^2$ = –0.142) also visible by the soil water content measurement for different treatments on 2nd May 2019 and 12th June 2019 with lower contents for CCM and CC and higher content for both treatments on the 26th June 2019 and 30th July 2019. Assuming a higher water infiltration in the treatments with vegetation cover after higher precipitation may explain the observed advantages in soil water in the summer period of 2019 in our study. Therefore, site-specific evaluation of the effects of a vegetation cover on soil water balance needs to consider environmental factors, soil temperature as well as factors related to the plant species community.

In our study, we observed a reduced photosynthetic activity measured as assimilation rate and stomatal conductance in under-vine vegetation treatments (CCM, CC) in periods of higher precipitation prior to the measurement, while in dry periods, no differences between treatments were determined. This was partly in accordance with previous reports of lower plant carbon assimilation when under-vine vegetation was applied, but these studies also highlight different trends in different years (Centinari *et al*., 2016; Hatch *et al*., 2011). Nevertheless, general conclusions on vine response to the under-vine treatments from measurements at different environmental conditions are difficult to extract, as we observed higher and lower values in CCM and CC as compared to herbicide application (Figure S5), similar to the soil water content in 2019. This inconsistent response at different

measurement dates explains the not significant influence of treatments on the photosynthetic activity as determined by linear mixed models. Additionally, our experimental setup with treatments established in individual vine rows cannot exclude interactions among adjacent treatments for physiological activity measurements, as vine root systems could be overlapping. Further on, our study did not account for vineyard variability by focusing the measurements on a more homogenous part of the vineyard. Therefore, the general conclusions drawn from our results of the treatment effects on the photosynthetic activity are difficult but could provide trends. Additionally, this snap-shot measurement of the photosynthetic activity could be entangled from the water retention of rainfall due to a different physical soil surface through vegetation and differences in soil porosity. Therefore, continuous measurements of the vine water status, the photosynthetic activity, the soil water contents in different soil layers and the evapotranspiration would be necessary to decipher the complex interaction in a spatial as well as temporal context. The positive effects of cover crops in mitigating soil erosion and precipitation run-off through the stabilisation of soil aggregates are documented manifold (Abad *et al*., 2021b; Garcia *et al*., 2019; Garcia *et al*., 2020; Novara *et al*., 2021). A study in a Mediterranean region determined that the water stock in their vineyard decreased linearly with the increase in vegetation coverage which was related to the rooting depth, the root morphology and the above-ground dry matter of the plants (Garcia *et al*., 2020). Similar outcomes were obtained in a meta-study, showing that on the one hand, cover crop mowing could be an option for a short-term water management strategy to reduce the competition of the vegetation on the vine and on the other hand, that the species composition can influence the severity of the stress on vines, especially in spring (Novara *et al*., 2021). Our case study could show that part of the vine's photosynthetic activity, as well as the soil water content, were influenced by environmental conditions, strongly supporting the necessity of studies including spatial and temporal factors as well as the plant species composition, in the case of cover crops or spontaneous vegetation in a randomised experimental block design in a vineyard to include also the factor vineyard variability.

Similarly to the photosynthetic activity, the interpretation of treatment effects on grape quality and grape yield has to consider the potential interactions of treatments due to the experimental design. Nevertheless, the grape quality parameters determined in our study (total soluble solids, titratable acidity and yeast assimilable nitrogen as NOPA) were differently affected by the under-vine treatments. We observed lower values for shoot pruning weight and berry weight in under-vine treatments with vegetation cover (CCM, CC), while the grape yield per vine itself was not affected. Other studies confirmed a reduction of vegetative growth of vines after cover crops were implemented in inter-rows (Chou and Vanden Heuvel, 2019; Guerra and Steenwerth, 2012), while other results observed this trend only in years with low precipitation (Pou *et al*., 2011) and only a few studies analysed the effects of under-vine floor management (Hickey *et al*., 2016; Vanden Heuvel and Centinari, 2021). The complexity of the agro-ecosystem studies provides conflicting results pertaining to responses of cover crops on vine growth, yield and grape quality and very often, the discrepancies can be related to different environmental conditions (Abad *et al*., 2021b). In a Mediterranean climate, cover crops reduce grape production by modifying yield components in different ways (Coletta *et al*., 2021, Muscas *et al*., 2017). In all cover crop varieties, both a yield reduction and reduced pruning weights were determined in comparison to tillage, partly confirming our observations. Other studies reported no effects on yield and limited reductions of vegetative growth in table grapes under the Mediterranean climate (Ferrara *et al*., 2021). On the other hand, in more humid vine-growing areas, one of the major aims of using cover crops is to reduce excessive vine vegetative growth and mitigate the negative consequences of heavy rainfall on soil properties and soil erosion (Vanden Heuvel and Centinari, 2021). In conclusion, it depends on the local conditions as well as on the farmers´ perspective if a competition effect of vegetation cover in inter-rows or under-vine areas is considered an advantage or disadvantage. Future studies should account for vineyard variability and complete randomised block designs should be established spanning several vine rows to exclude across-treatment effects due to the expanded grapevine root system. This is specifically true if grapevine physiology, growth and grape yield are investigated, as in our case study. Our study could not account for potential adjacent treatment effects due to the design of the experimental vineyard. Although this may have some effects on the part of our results, our study could clearly show the strong effects of different under-vine management measures on soil enzyme activity.

## CONCLUSION

The case study investigated under-vine management strategies in an Austrian vineyard with the aim of understanding the effects on the hydrolytic soil enzyme activity, the vine photosynthetic activity, and grape yield and quality. Thereby we aimed to investigate the balance between soil water relation and vine photosynthesis under different under-vine management regimes as well as if consequences on vine growth and grape quality could be observed due to vegetation competition. We hypothesised that potential negative impacts on vines of under-vine vegetation would partially be compensated by higher soil enzymatic activity. Our study could not provide the direct linkage between soil microbial activity, vine nutrition and grape quality, but we determined important results in terms of site-specific sustainable management adaptations for under-vine weed control. 1) We confirm an enhanced soil microbial activity with under-vine vegetation, independent of its regular mowing or not. 2) We observed seasonal effects on β-glucosidase activity and the soil water content, especially in 11–20 cm soil depth due to an active growing spontaneous under-vine vegetation in spring. 3) The photosynthetic activity depends

on environmental conditions, as shown by an interaction of precipitation and under-vine treatments. Last but not least, 4) only berry weight and shoot pruning weight were reduced by under-vine vegetation as compared to herbicide application in our study. The observed effects on photosynthetic activity, vine growth and grape yield may not be totally independent as interactions among adjacent treatments could not be totally excluded due to the vineyard and experimental design. In conclusion, we propose that under-vine vegetation regularly mowed could be an alternative under-vine management strategy in supplement to soil tillage and herbicide application. Additionally, the enhanced soil microbial activity could enhance vine resilience through root–microbe interaction. Nevertheless, site-specific solutions may be required to account for both the support of ecosystem services and the production of high-quality grapes.

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MG, AF, and EK conceived and designed the study and coordinated the establishment of the treatments and the measurements and data collections; MG, SK and the master students performed the measurements in the vineyard; SK and FDB established the fluorometric microplate-based method to determine soil enzyme activities and performed the analysis; MG conducted the data evaluation, the data analyses, interpreted the results and drafted the manuscript; all authors did read and approve the final manuscript.

## REFERENCES

Abad, J., de Mendoza, I. H., Marin, D., Orcaray, L., & Santesteban, L. G. (2021a). Cover crops in viticulture. A systematic review (1): Implications on soil characteristics and biodiversity in vineyard. *Oeno One,* 55(1), 295-312. https://doi.org/10.20870/oeno-one.2021.55.1.3599

Abad, J., de Mendoza, I. H., Marin, D., Orcaray, L., & Santesteban, L. G. (2021b). Cover crops in viticulture. A systematic review (2): Implications on vineyard agronomic performance. *Oeno One,* 55(2), 1-27. https://doi.org/10.20870/oeno-one.2021.55.2.4481

Akinyemi, D. S., Zhu, Y., Zhao, M., Zhang, P., Shen, H., & Fang J. (2020). Response of soil extracellular enzyme activity to experimental precipitation in a shrub-enroached grassland in Inner Mongolia. *Global Ecology and Conservation*, 23, e01175. https://doi.org/10.1016/j.gecco.2020.e01175

Abad, J., Marin, D., Santesteban, L.G., Cibriain, J.F., & Sagues, A. (2020). Under-vine cover crops: impact on weed development, yield and grape composition. *Oeno One* 54, 975-983. https://doi.org/10.20870/oeno-one.2020.54.4.4149

Bates, D., Mächler, M., Bolker, B., & Walker, S. (2015). Fitting Linear Mixed-Effects Models Using lme4. *Journal Statistical Software,* 067. https://doi.org/10.18637/jss.v067.i01

Blanco-Canqui, H., Shaver, T. M., Lindquist, J. L., Shapiro, C. A., Elmore, R. W., Francis, C. A., & Hergert, G. W. (2015). Cover Crops and Ecosystem Services: Insights from Studies in Temperate Soils. *Agronomy Journal,* 107(6), 2449-2474. https://doi.org/10.2134/agronj15.0086

Burns, K. N., Bokulich, N. A., Cantu, D., Greenhut, R. F., Kluepfel, D. A., O'Geen, A. T., Strauss, S. L., & Steenwerth, K. L. (2016). Vineyard soil bacterial diversity and composition revealed by 16S rRNA genes: Differentiation by vineyard management. *Soil Biology & Biochemistry,* 103, 337-348. https://doi.org/10.1016/j.soilbio.2016.09.007

Centinari, M., Vanden Heuvel, J. E., Goebel, M., Smith, M. S., & Bauerle, T. L. (2016). Root-zone management practices impact above and belowground growth in Cabernet Franc grapevines. *Australian Journal of Grape and Wine Research,* 22(1), 137-148. https://doi.org/10.1111/ajgw.12162

Chou, M. Y., Vanden Heuvel, J., Bell, T. H., Panke-Buisse, K., & Kao-Kniffin, J. (2018). Vineyard under-vine floor management alters soil microbial composition, while the fruit microbiome shows no corresponding shifts. *Scientific Reports,* 8. https://doi.org/10.1038/s41598-018-29346-1

Chou, M. Y., & Vanden Heuvel, J. E. (2019). Annual Under-Vine Cover Crops Mitigate Vine Vigor in a Mature and Vigorous Cabernet franc Vineyard. *American Journal of Enology and Viticulture,* 70(1), 98-108. DOI: 10.5344/ajev.2018.18037

Coletta, A., Toci, A. T., Pati, S., Ferrara, G., Grieco, F., Tufariello, M., & Crupi, P. (2021). Effect of Soil Management and Training System on Negroamaro Wine Aroma. *Foods,* 10(2). https://doi.org/10.3390/foods10020454

Coniberti, A., Ferrari, V., Disegna, E., Dellacassa, E., & Lakso, A. N. (2018a). Under-trellis cover crop and deficit irrigation to regulate water availability and enhance Tannat wine sensory attributes in a humid climate. *Scientia Horticulturae,* 235, 244-252. http://dx.doi.org/10.1016/j.scienta.2018.03.018

Coniberti, A., Ferrari, V., Disegna, E., Petillo, M. G., & Lakso, A. N. (2018b). Under-trellis cover crop and planting density to achieve vine balance in a humid climate. *Scientia Horticulturae,* 227, 65-74. https://doi.org/10.1016/j.scienta.2017.09.012

Curtright, A. J., & Tiemann, L. K. (2021). Intercropping increases soil extracellular enzyme activity: A meta-analysis. *Agriculture Ecosystems & Environment,* 319. https://doi.org/10.1016/j.agee.2021.107489

Deng, S. P., Popova, I. E., Dick, L., & Dick, R. (2013). Bench scale and microplate format assay of soil enzyme activities using spectroscopic and fluorometric approaches. *Applied Soil Ecology*, 64, 84-90. https://doi.org/10.1016/j.apsoil.2012.11.002

Di Giacinto, S., Friedel, M., Poll, C., Döring, J., Kunz, R., & Kauer, R. (2020). Vineyard management system affects soil microbiological properties. *OENO One*, 54 (1), 131-143. https://doi.org/10.20870/oeno-one.2020.54.1.2578

Dick, R. P., Dick, L. K., Deng, S. P., Li, X. F., Kandeler, E., Poll, C., Freeman, C., Jones, T. G., Weintraub, M. N., Esseili, K. A., & Saxena, J. (2018). Cross-laboratory comparison of fluorimetric microplate and colorimetric bench-scale soil enzyme assays. *Soil*

*Biology & Biochemistry,* 121, 240-248. https://doi.org/10.1016/j.soilbio.2017.12.020

Ferrara, G., Nigro, D., Torres, R., Gadaleta, A., Fidelibus, M. W., & Mazzeo, A. (2021). Cover crops in the inter-row of a table grape vineyard managed with irrigation sensors: Effects on yield, quality and glutamine synthetase activity in leaves. *Scientia Horticulturae,* 281. https://doi.org/10.1016/j.scienta.2021.109963

Fierer, N. (2017). Embracing the unknown: disentangling the complexities of the soil microbiome. *Nature Reviews Microbiology,* 15(10), 579-590. https://doi.org/10.1038/nrmicro.2017.87

Fox, J., & Weisberg, S. (2019). An R Companion to Applied Regression, 3rd edition. Sage, Thousand Oaks CA. https://socialsciences.mcmaster.ca/jfox/Books/Companion/index.html

Garcia, L., Celette, F., Gary, C., Ripoche, A., Valdes-Gomez, H., & Metay, A. (2018). Management of service crops for the provision of ecosystem services in vineyards: A review. *Agriculture Ecosystems & Environment,* 251, 158-170. https://doi.org/10.1016/j.agee.2017.09.030

Garcia, L., Damour, G., Gary, C., Follain, S., Le Bissonnais, Y., & Metay, A. (2019). Trait-based approach for agroecology: contribution of service crop root traits to explain soil aggregate stability in vineyards. *Plant and Soil,* 435(1-2), 1-14. https://link.springer.com/article/10.1007/s11104-018-3874-4

Garcia, L., Metay, A., Kazakou, E., Storkey, J., Gary, C., Damour, G. (2020). Optimising the choice of service crops in vineyards to achieve both runoff mitigation and water provisioning for grapevine: a trait-based approach. *Plant Soil* 452, 87-104. https://doi.org/10.1007/s11104-020-04543-y

Gattullo, C. E., Mezzapesa, G. N., Stellacci, A. M., Ferrara, G., Occhiogrosso, G., Petrelli, G., Castellini, M., & Spagnuolo, M. (2020). Cover Crop for a Sustainable Viticulture: Effects on Soil Properties and Table Grape Production. *Agronomy-Basel,* 10(9). https://doi.org/10.3390/agronomy10091334

Gaudin, R., Celette, F., & Gary, C. (2010). Contribution of runoff to incomplete off season soil water refilling in a Mediterranean vineyard. *Agricultural Water Management,* 97(10), 1534-1540. http://dx.doi.org/10.1016/j.agwat.2010.05.007

Geldenhuys, M., Gaigher, R., Pryke, J. S., & Samways, M. J. (2021). Diverse herbaceous cover crops promote vineyard arthropod diversity across different management regimes. *Agriculture Ecosystems & Environment,* 307. https://doi.org/10.1016/j.agee.2020.107222

German, D. P., Weintraub, M. N., Grandy, A. S., Lauber, C. L., Rinkes, Z. L., & Allison, S. D. (2011). Optimisation of hydrolytic and oxidative enzyme methods for ecosystem studies. *Soil Biology & Biochemistry,* 43(7), 1387-1397. https://doi.org/10.1016/j.soilbio.2011.03.017

Griesser, M., Steiner, M., Pingel, M., Uzman, D., Preda, C., Giffard, B., Tolle, P., Memedemin, D., Forneck, A., Reineke, A., Leyer, I., & Bacher, S. (2022). General trends of different inter-row vegetation management affecting vine vigor and grape quality across European vineyards. *Agriculture, Ecosystems & Environment 338*, 1-11. https://doi.org/10.1016/j.agee.2022.108073

Guerra, B., & Steenwerth, K. (2012). Influence of Floor Management Technique on Grapevine Growth, Disease Pressure, and Juice and Wine Composition: A Review. *American Journal of Enology and Viticulture, 63*(2), 149-164. https://doi.org/10.5344/ajev.2011.10001

Guzman, G., Cabezas, J. M., Sanchez-Cuesta, R., Lora, A., Bauer, T., Strauss, P., Winter, S., Zaller, J. G., & Gomez, J. A. (2019). A field evaluation of the impact of temporary cover crops on soil properties and vegetation communities in southern Spain vineyards. *Agriculture Ecosystems & Environment,* 272, 135-145. https://doi.org/10.1016/j.agee.2018.11.010

Hatch, T. A., Hickey, C. C., & Wolf, T. K. (2011). Cover Crop, Rootstock, and Root Restriction Regulate Vegetative Growth of Cabernet Sauvignon in a Humid Environment. *American Journal of Enology and Viticulture,* 62(3), 298-311. https://doi.org/10.5344/ajev.2011.11001

Hickey, C. C., Hatch, T. A., Stallings, J., & Wolf, T. K. (2016). Under-Trellis Cover Crop and Rootstock Affect Growth, Yield Components, and Fruit Composition of Cabernet Sauvignon. *American Journal of Enology and Viticulture,* 67(3), 281-295. https://doi.org/10.5344/ajev.2016.15079

Karl, A. D., Merwin, I. A., Brown, M. G., Hervieux, R. A., & Vanden Heuvel, J. E. (2016). Under-vine Management Impacts Soil Properties and Leachate Composition in a New York State Vineyard. *Hortscience,* 51(7), 941-949. https://doi.org/10.21273/HORTSCI.51.7.941

Lenth, R.V. (2021). emmeans: Estimated Marginal Means, aka Least-Squares Means. R package version 1.6.3. https://CRAN.R-project.org/package=emmeans

Long, J. A. (2019). interactions: Comprehensive, User-Friendly Toolkit for Probing Interactions. R package version 1.1.0, <URL: https://cran.r-project.org/package=interactions>.

Li, Y. Z., Song, D. P., Liang, S. H., Dang, P. F., Qin, X. L., Liao, Y. C., & Siddique, K. H. M. (2020). Effect of no-tillage on soil bacterial and fungal community diversity: A meta-analysis. *Soil & Tillage Research,* 204. https://doi.org/10.1016/j.still.2020.104721

Lüdecke, D., Ben-Shachar, M., Patil, I., Waggoner, P., Makowski, D. (2021). "Performance: An R Package for Assessment, Comparison and Testing of Statistical Models." *Journal of Open Source Software*, 6(60), 3139. https://doi.org/10.21105/joss.03139

Marques, M.J., Ruiz-Colmenero, M., Bienes, R., Garcia-Diaz, A., Sastre. B. (2020). Effects of a Permanent Soil Cover on Water Dynamics and Wine Characteristics in a Steep Vineyard in the Central Spain. *Air, Soil and Water Research,* 13, 1-10. https://doi.org/10.1177/2F1178622120948069

Muscas, E., Cocco, A., Mercenaro, L., Cabras, M., Lentini, A., Porqueddu, C., Nieddu, G. (2017). Effects of vineyard floor cover crops on grapevine vigor, yield, and fruit quality, and the development of the vine mealybug under a Mediterranean climate. *Agriculture, Ecosystem and Environment*, 237, 203-212. https://doi.org/10.1016/j.agee.2016.12.035

Nkongolo, K. K., & Narendrula-Kotha, R. (2020). Advances in monitoring soil microbial community dynamic and function. *Journal of Applied Genetics,* 61(2), 249-263. https://doi.org/10.1007/s13353-020-00549-5

Novara, A., Catania, V., Tolone, M., Gristina, L., Laudicina, V. A., & Quatrini, P. (2020). Cover Crop Impact on Soil Organic Carbon, Nitrogen Dynamics and Microbial Diversity in a Mediterranean Semiarid Vineyard. *Sustainability,* 12(8). https://doi.org/10.3390/su120832569

Novara, A., Cerda, A., Barone, E., & Gristina, L. (2021). Cover crop management and water conservation in vineyard and olive orchards. *Soil & Tillage Research,* 208. https://doi.org/10.1016/j.still.2020.104896

Paiola, A., Assandri, G., Brambilla, M., Zottini, M., Pedrini, P., & Nascimbene, J. (2020). Exploring the potential of vineyards for biodiversity conservation and delivery of biodiversity-mediated ecosystem services: A global-scale systematic review. *Science of the Total Environment,* 706. https://doi.org/10.1016/j.scitotenv.2019.135839

Paredes, D., Rosenheim, J. A., Chaplin-Kramer, R., Winter, S., & Karp, D. S. (2021). Landscape simplification increases vineyard pest outbreaks and insecticide use. *Ecolocial Letters,* 24(1), 73-83. https://doi.org/10.1111/ele.13622

Pou, A., Gulias, J., Moreno, M., Tomas, M., Medrano, H., & Cifre, J. (2011). Cover Cropping in Vitis Vinifera L. Cv. Manto Negro Vineyards under Mediterranean Conditions: Effects on Plant Vigour, Yield and Grape Quality. *Journal International Des Sciences De La Vigne Et Du Vin,* 45(4), 223-234. https://doi.org/10.20870/oeno-one.2011.45.4.1501

Pustejovsky, J. (2021). clubSandwich: Cluster-Robust (Sandwich) Variance Estimators with Small-Sample Corrections. R package version 0.5.3. https://CRAN.R-project.org/package=clubSandwich

R Core Team (2020). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

Ruiz-Colmenero, M., Bienes, R., & Marques, M. J. (2011). Soil and water conservation dilemmas associated with the use of green cover in steep vineyards. *Soil & Tillage Research,* 117, 211-223. http://dx.doi.org/10.1016/j.still.2011.10.004

Six, J., Elliott, E. T., & Paustian, K. (1999). Aggregate and soil organic matter dynamics under conventional and no-tillage systems. *Soil Science Society of America Journal, 63*(5), 1350-1358.

Steenwerth, K. L., Calderon-Orellana, A., Hanifin, R. C., Storm, C., & McElrone, A. J. (2016). Effects of Various Vineyard Floor Management Techniques on Weed Community Shifts and Grapevine Water Relations. *American Journal of Enology and Viticulture,* 67(2), 153-162. http://dx.doi.org/10.5344/ajev.2015.15

Steiner, M., Grace, J. B., & Bacher, S. (2021). Biodiversity effects on grape quality depend on variety and management intensity. *Journal of Applied Ecology,* 58(7), 1442-1454. https://doi.org/10.1111/1365-2664.13899

Steinweg, J.M., Dukes, J.S., & Wallenstein, M. D. (2012). Modeling the effects of temperature and moisture on soil enzyme activity : Linking laboratory assays to continous field data. *Soil Biology & Biochemistry,* 55, 85-92. https://doi.org/10.1016/j.soilbio.2012.06.015

Tiwari, R., Dwivedi, B.S., Sharma, A., & Dwivedi, A.K. (2019). Activities of β-glucosidase, phosphatase and dehydrogenase as soil quality indicators : a review. International Journal of Current Microbiology and Applied Sciences, 8(6), 834-846. https://doi.org/10.20546/ijcmas.2019.806.101

Torres, R., Ferrara, G., Soto, F., Lopez, J. A., Sanchez, F., Mazzeo, A., Perez-Pastor, A., & Domingo, R. (2017). Effects of Soil and Climate in a Table Grape Vineyard with Cover Crops. Irrigation Management Using Sensors Networks. *Ciencia E Tecnica Vitivinicola,* 32(1), 72-81. https://doi.org/10.1051/ctv/20173201072

Vanden Heuvel, J., & Centinari, M. (2021). Under-Vine Vegetation Mitigates the Impacts of Excessive Precipitation in Vineyards. *Frontiers Plant Science,* 12. https://dx.doi.org/10.3389%2Ffpls.2021.7131

Verdenal, T., Dienes-Nagy, Á., Spangenberg, J. E., Zufferey, V., Spring, J.-L., Viret, O., Marin-Carbonne, J., & van Leeuwen, C. (2021). Understanding and managing nitrogen nutrition in grapevine: a review. *Oeno One,* 55(1), 1-43. https://doi.org/10.20870/oeno-one.2021.55.1.3866

Wickham, H. (2016). ggplot2: Elegant Graphics for Data Analysis. Springer-Verlag New York. ISBN 978-3-319-24277-4, https://ggplot2.tidyverse.org

Winter, S., Bauer, T., Strauss, P., Kratschmer, S., Paredes, D., Popescu, D., Landa, B., Guzman, G., Gomez, J. A., Guernion, M., Zaller, J. G., & Batary, P. (2018). Effects of vegetation management intensity on biodiversity and ecosystem services in vineyards: A meta-analysis. *Journal of Applied Ecology,* 55(5), 2484-2495. < https://doi.org/10.1111/1365-2664.13124

Zaller, J. G., Cantelmo, C., Dos Santos, G., Muther, S., Gruber, E., Pallua, P., Mandl, K., Friedrich, B., Hofstetter, I., Schmuckenschlager, B., & Faber, F. (2018). Herbicides in vineyards reduce grapevine root mycorrhisation and alter soil microorganisms and the nutrient composition in grapevine roots, leaves, xylem sap and grape juice. *Environmental Science and Pollution Research,* 25(23), 23215-23226. https://dx.doi.org/10.1007%2Fs11356-018-2422-3

Zanettin, G., Bullo, A., Pozzebon, A., Burgio, G., & Duso, C. (2021). Influence of Vineyard Inter-Row Groundcover Vegetation Management on Arthropod Assemblages in the Vineyards of North-Eastern Italy. *Insects,* 12(4). https://doi.org/10.3390/insects12040349.