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ORIGINAL RESEARCH ARTICLE

Evaluation of site-specific management to optimise *Vitis vinifera* L. (cv. Tannat) production in a vineyard with high heterogeneity

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ABSTRACT

The spatial variability of vineyards can be characterised through precision viticulture that will allow setting the boundaries of homogeneous management zones. This study aimed to evaluate the impact of soil and plant management (site-specific management) to increase yields and improve berry quality. During three consecutive seasons, contrasting treatments designed ad hoc for two zones of vigour pre-established by NDVI were tested: high vigour zone (HV) and low vigour zone (LV). The treatments were aimed at reducing water and nitrogen supply and improving microclimatic conditions in the cluster zone in the HV zone. In the LV zone, treatments were aimed at increasing water and nitrogen supply. Leaf removal in the HV zone was the most efficient treatment to improve productivity and quality. Moreover, the water restriction improved grape quality, especially in a rainy year. The regulated deficit irrigation strategy applied in the LV zone at specific phenological stages was shown to increase vegetative growth, yield and to improve grape anthocyanins and phenols contents. The benefits of additional nitrogen supply in the LV zone on plant nitrogen status, yield, and berry composition were highly dependent on water availability. Ultimately, this study provided new insights into the relationship between water and nitrogen availability and how this determines vigour and influences yield and grape quality and influences the deviation from a "Productive Target" pattern. The use of site-specific techniques could be adjusted on a small production scale, thanks to mapping carried out with precision viticulture technologies.

KEYWORDS: precision viticulture, sustainable production, vigour, irrigation, fertilisation, leaf removal

INTRODUCTION

Conventional crop management generally relies on a set of operational decisions which are implemented at the plot scale. Indeed, the soil and canopy management, together with the different inputs (fertiliser, irrigation, phytosanitary products), are applied in a similar way across the plot without considering the within-plot heterogeneity (Arnó, 2008). However, high variability in grape yield and composition within a vineyard (Bramley *et al.,* 2011; Filippetti *et al.,* 2013; Ferrer *et al.,* 2020a) may be triggered by non-homogeneous physical and chemical characteristics of the soil, topography, and climate (Jasse *et al.,* 2021). Inadequate soil and plant management at the plot level is ultimately likely to generate both economic loss and environmental issues (Arnó, 2008; Filipetti *et al.,* 2013; King *et al.,* 2014).

Precision viticulture (PV) combines technologies and methodologies based on data collection and data analysis to optimise production and economic and environmental aspects (Tisseyre *et al.*, 2008; Bramley *et al.*, 2011; Santesteban, 2019). The characterisation of the spatial variation within a plot allows the winegrower to apply various management strategies. Within these strategies, homogeneous management zones can be delimited and site-specific management to each zone can be applied. Another management strategy is using VRD (Variable Rate Dosing) technologies that allow achieving a dosage of inputs according to the information generated instantaneously or previously (Tisseyre *et al.*, 2008). The plot spatial variation may concern soil/plant water status (Santesteban, 2019) and soil characteristics (Arnó *et al.,* 2012; Bramley *et al.,* 2011). Topography (Bahat *et al.,* 2021), soil electrical properties (Tardaguila *et al.,* 2011), yield and the normalised difference vegetation index (NDVI) can ultimately be used to establish management zones. The NDVI is calculated from the red (R) and infrared (IR) wavelengths as follows: NDVI=[IR-R]/[IR+R] (Rouse *et al.,* 1974). Vigour differences (assessed by NDVI) are generally associated with contrasted vegetative growth, yield and grape composition (Filippetti *et al.,* 2013; Ferrer *et al.,* 2020a; Sams *et al.,* 2022). High vigour is assumed to favour high yield and vegetative growth and the development of fungal diseases that can alter grape composition (Bramley *et al.,* 2011; Filippetti *et al.,* 2013; Ferrer *et al.,* 2020a; Gatti *et al.,* 2020). In contrast, low plant vigour often induces low yield, poor vegetative growth and excessive cluster exposure to direct radiation and sunburn issues (McClymont *et al.,* 2012; Ferrer *et al.,* 2020a).

When the spatial heterogeneity in plant growth and productivity are due to the variation of physical factors presenting stability over time (Taylor *et al.,* 2010, Arnó *et al.,* 2012; Matese and Di Gennaro, 2015), it is possible to differentiate the management practices (site-specific management) within the same plot year after year. Notably, because water and nitrogen are two of the factors that determine vigour expression, contrasted water and nitrogen supplies may be imposed, depending on soil characteristics, to homogenise plant vigour at the plot scale (Taylor *et al.,* 2010; Martinez-Casasnovas *et al.,* 2012; King *et al.,* 2014). Canopy management, involving, for example, leaf removal to improve the canopy and bunch microclimate, can also be implemented for a site-specific management plan (Pedò *et al.,* 2010; Arrillaga *et al.,* 2021). A selective harvest within each zone to reduce spatial variation in yield and grape composition (Scarlett *et al.,* 2014) and reach different qualities could be another relevant strategy. Lastly, prior to planting the vineyard, soil electroconductivity mapping will guide the winegrower to better reason the vine rows' orientation, the selection of the variety and rootstock and choice of the training system to reduce the heterogeneity later on. Ultimately, PV can improve the environmental and economic sustainability of the vineyards by minimising environmental impacts and maximising the oenological potential of the grape (Arnó, 2008).

In Uruguay, the year-to-year fluctuations of rainfall, added to the heterogeneity of soils, can generate local situations of water and nitrogen deficit with consequences on plant physiology and productivity. In this context, PV appears a relevant option to tackle the combined effects of climate (meso and micro) variability and soil heterogeneity at the plot scale (Matese and Di Gennaro, 2015). The present study was aimed at evaluating the impact of site-specific soil and plant management on grapevine vigour, yield and berry composition. Thus, different water or nitrogen supply and leaf removal in the bunch zone were tested over three consecutive seasons within pre-defined zones of high and low vigour. The objective was to develop specific management practices to increase yield and quality and reduce spatial heterogeneity.

MATERIALS AND METHODS

1. Experimental site

1.1. Vineyard

The experiment was conducted in a commercial vineyard in Canelones, Uruguay (34°36'S,56°14W), over three consecutive seasons (2019–2020–2021). The vineyard of 1.1 ha was planted in 1998 with *Vitis vinifera* L. cv. Tannat, grafted on SO4 rootstock. The vine spacing was 2.5 m × 1.2 m (3333 vines ha^{-1}). Vines were pruned using a double guyot system, and the shoots were trained to a VSP (vertical shoot positioning) system. The vineyard was not irrigated and received standard fertilisation with urea, distributed half pre-flowering and half post-harvest at a total dose of 140 kg of urea (46 % N) fertiliser per ha. This vineyard was characterised by high variability of vine vigour from east to west, and Ferrer et al. (2020a) defined three vigour zones, high (HV), medium (MV) and low vigour (LV) (Supplement 1). The determination of vigour zones was made by aircraft flight (620 m altitude and speed of 50 m/s) at veraison (January in the southern hemisphere) for three years. High-resolution multispectral images were obtained at ground level (0.2 m). The classes of NDVI (high, medium and low) were consistently located in the same parts of the vineyard each year (Ferrer et al., 2020a). Soil physical and chemical characteristics also showed a strong spatial

variability, mainly regarding the percentage of clay, clay type and total available water (TAW). The TAW estimated from soil texture and root depth was greater than 180 mm in the HV with a predominance of montmorillonite (expansive clay) and less than 140 mm in the LV with higher content of illite compared to HV. Ultimately, although the vineyard was relatively small, making the application of PV unbeneficial, the high spatial gradient of vigour and its stability over the years made this vineyard of high value to test different management practices according to the vigour.

2.2. Treatments

Treatments were carried out depending on the vigour zone to improve the yield and berry quality and reduce their heterogeneity. For this purpose, only the most contrasting vigour areas of the vineyard (HV vs LV) were selected. In the pre-established zones, as described above, HV (NDVI 0.57 to 0.61) and LV (NDVI 0.48 to 0.55), treatments were arranged in a random block design with three replications and 21 vines per replicate. For HV, the treatments are aimed at reducing water and nitrogen supply and improving the microclimatic conditions in the bunch zone. The water restriction (H-W) was implemented from veraison to harvest by covering the soil with polyethylene (white on both sides, 220 micrometres thick, with ultraviolet protection). No nitrogen was applied (0 N unit in the season) in the nitrogen restriction treatment (H-N). During two seasons (2019 and 2020), the winegrower stopped fertilising with urea on one subplot (rows 5 to 15, where this treatment was randomly installed). Leaf removal (H-L) was applied at the pre-flowering stage by removing 60 % of the leaves. In contrast, for LV, the treatments are aimed at increasing water and nitrogen supply. Additional irrigation was supplied (L+W) compared to the control (LV) to reach 100 % of the climatic demand (ETo) from budburst to flowering and from harvest to leaf fall, and 70 % of ETo from flowering to harvest. For the supplemental nitrogen treatment (L+N), 210 kg urea per ha were supplied (70 kg urea per ha in the form of urea were added prior to flowering to the 140 kg mentioned above). In addition, a treatment combining both water and nitrogen supplements (L+WN) was carried out. These treatments were compared with their respective controls for each vigour zone (HV and LV). All treatments are presented in Table 1 and Figure 1.

2. Weather measurements

2.1. Weather characterisation

Meteorological data were collected from a weather station owned by INIA (National Institute of Agricultural Research; 34°40'S, 56°20'W; 10 km from the experimental site) and managed according to the World Meteorological Organization (WMO) standards, to which added a pluviometer. The following climatic variables were monitored: mean air temperature (Tm, °C), reference Penman–Monteith evapotranspiration (ETo, mm), photosynthetically active radiation (PAR, mmol m^{-2} s^{-1}), relative air humidity (RH, %) and precipitation (mm).

2.2. Microclimate characterisation

For the microclimatic data, three HOBO® sensors (HOBO® U23 ProV2 & HOBO Pendant® loggers, USA) were distributed inside the canopy in the bunch zone for each treatment from flowering to harvest (2019 to 2021). The results for the most contrasting years in terms of weather are presented in the 3.1 section (2019 vs 2020).

TABLE 1. Description of the water, nitrogen and leaf removal treatments in the high (HV) and low (LV) pre-delimited vigor zones and years evaluated.

						Seasons		
Vigor	Treatment	Rainfall allowed	Nitrogen supplied	Leaves removed	Irrigation supplied	2019	2020	2021
High Vigor	Control (HV)	✓	✓	✗	✗	✓	✓	✓
	Water restriction (H-W)	✗[1]	✓	✗	✗	✓	✓	✗
	Nitrogen restriction (H-N)	✓	✗	✗	✗	✓	✓	✗
	Leaf removal (H-L)	✓	✓	✓	✗	✓	✓	✓
Low vigor	Control (LV)	✓	✓	✗	✗	✓	✓	✓
	Irrigation (L+W)	✓	✓	✗	✓	✗	✓	✓
	Nitrogen supply (L+N)	✓	✓ ✓	✗	✗	✓	✓	✓
	Irrigation+Nitrogen supply (L+WN)	✓	✓ ✓	✗	✓	✗	✗	✓

[1]No rain from veraison to harvest.

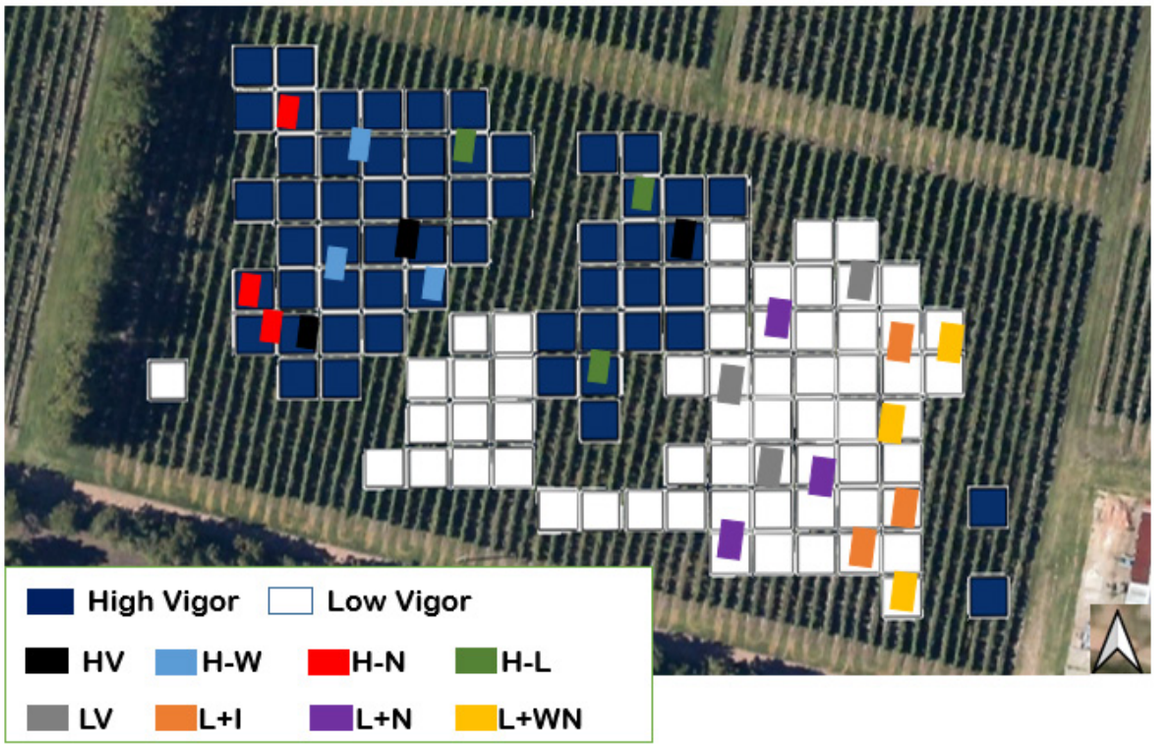


FIGURE 1. Plot location of water, nitrogen and leaf removal treatments in the high (HV) and low (LV) pre-defined vigor zones and replications (adapted from Ferrer *et al.*, 2020a) (see Table 1 for the treatment description).

The mean canopy temperature (Tmc) and relative humidity (RHc) were recorded using HOBO® U23 ProV2 loggers, while the photosynthetically active radiation in the canopy (PARc) was measured from HOBO Pendant® loggers. All variables were recorded at an hourly time step.

3. Nitrogen and water status

3.1 Soil N status

Soil samples were taken at three depths (0–20; 20–40; 40–60 cm) for each replicate in the HV and LV in winter (August) before budburst (2019 to 2021). Nitrate (NO_3^-), ammonium (NH4+), and organic matter (O.M.) were evaluated each year. The N stock was estimated from the soil analyses combined with a potential mineralisation term (Salvo *et al.,* 2014). Nitrogen leaching was not taken into account.

3.2. Leaf N status

Leaf nitrogen content was evaluated on 20 whole healthy and exposed leaves (limbs) at veraison. The samples were dried (60 ºC, 48 hours) and analysed in the soil and plant laboratory of the Facultad de Agronomía (Uruguay). Leaf nitrogen was assayed by Kjeldahl's method.

3.3. Leaf water status

From flowering to harvest, vine water status at pre-dawn was determined using a pressure chamber (SoilMoisture equipment, Santa Barbara, CA, USA). Nine healthy expanded leaves were taken from each treatment (three leaves per replicates). Leaves were covered with a plastic bag before cutting the petiole and the measurement was carried out immediately after detaching the leaf from the plant.

4. Plant and berry measurements

4.1. Vegetative growth

Vegetative growth was evaluated at veraison. Ten representative shoots (average shoots in terms of length and diameter) were collected for each treatment, and the numbers of primary and secondary leaves were counted. In addition, the number of shoots per plant was counted. The leaf area of each leaf was estimated using the Easy Leaf Area® mobile application (Easlon and Bloom, 2014). The plants evaluated had the same number of buds (12 buds). The % lignification was measured in the same ten representative shoots (described above) at harvest. The total length of the shoot and the length of the lignified borer were measured. Pruning weight (PW, g/plant) was measured during winter on the same 63 plants.

4.2. Yield

The harvest date was fixed according to the evolution of pH (3.3 to 3.4) and berry weight to prevent dehydration of the berry (avoid weight loss); details of dates are presented in the Supplementary information (Table S2). Yield (Y, kg/vine), the number of bunches (B/vine) and the individual bunch weight (WB) at harvest were determined on 63 plants for each treatment (21 plants for each replicate). As the main bunch disease detected was Botrytis bunch rot (*Botrytis cinerea* Pers.), the percentage of bunch rot (BR, %) was evaluated at

harvest. Lastly, the individual berry weight was determined at harvest on samples of 100 berries for each replication.

4.3. Grape composition

At harvest, two samples of 100 berries were collected from the central zone of the bunch (Deloire *et al.,* 2019) for each treatment. One sample was crushed using a juicer, Phillips HR2290 (Phillips, Netherlands). The following berry composition variables were determined: total soluble solids (TSS) by refractometer (Atago, Japan); pH by pH-meter (Hanna Instruments, Italy); and acidity by titration (gH2SO4/L), following the protocols established by the OIV (2014). The available yeast nitrogen (YAN, mg/l) content in the musts was determined using formaldehyde quantification (Aerny, 1996). A second sample was used to determine the content of total anthocyanins (A, mg/l) and the total phenol index (TPI) according to the methodology proposed by Glories and Agustin (1993) as modified by González-Neves *et al.* (2004). The measurements were performed in duplicate with a spectrophotometer (Shimadzu UV-1240 Mini, Japan) using glass cells for anthocyanin analysis (absorbance at 520 nm) and quartz cells for phenols (absorbance at 280 nm) with a 1 cm optical path length.

5. Data analyses

5.1. Effect of the treatments

Statistical analyses were conducted with the statistical package InfoStat version 2011. Analyses of variance, followed by the Tukey test for means comparison, were conducted to determine the effect of the different treatments on plant and berry variables for each zone of vigour (HV, LV) over the vintages 2019 to 2021.

5.2. Derivation from an optimal "Productive Target" pattern

The spatial variations in the HV and LV zones from a "Productive Target" pattern for a sustainable and cost-effective production over vintages (2019 to 2021) of a few selected important productive and economic indicator variables (PW, Y, BR, TSS, YAN, A, and TPI) were analysed. The definition of this "Productive Target" pattern was based on reported values considered as favourable for reaching a balanced vine (Ravaz Index, 5-7 defined by Ferrer *et al.* (1997) for Tannat) and high-quality wines for Tannat (González-Neves *et al.,* 2012; Ferrer *et al.,* 2018; Ferrer *et al.,* 2020b). The variables considered were categorized into four classes or scores, where score 3 corresponded to the optimal "Productive Target", which includes the following values considered: PW = 650 g, Y = 5 kg/vine, BR = 5 %, TSS = 238 g/l, YAN = 150 mg/l; A = 2540 mg/l and TPI = 80. The level of BR used was proposed by Ky *et al.* (2012), which established an acceptable tolerance range of 5 % of botrytised berries to avoid severe oenological consequences in the wines.

For each treatment, values in the same score or higher than the Productive Target were considered beneficial for that variable. Conversely, scores below 3 (scores 1 and 2) were assumed to be not beneficial for those selected variables.

The values considered for each score and variable were:

PW (g) = ***Score 1***: < 400; ***Score 2***: 400–600; ***Score 3***: 600–800; ***Score 4***: > 800

Y (kg/pl) = ***Score 1***: < 3.5; ***Score 2***: 3.5–5.0; ***Score 3***: 5.0–6.5; ***Score 4***: > 6.5

BR (%) = ***Score 1***: >10; ***Score 2***: 10–5; ***Score 3***: 5–0; ***Score 4***: 0

TSS (g/l) = ***Score 1***: <192; ***Score 2***: 190–215; ***Score 3***: 215–238; ***Score 4***: >238.

YAN (mg/l) = ***Score 1***: <100; ***Score 2***: 100–140; ***Score 3***: 140–180; ***Score 4***: >180.

A (mg/l) = ***Score 1***: < 1800; ***Score 2***: 1800–2200; ***Score 3***: 2200–2600; ***Score 4***: > 2600.

IPT= ***Score 1***: < 25; ***Score 2***: 25–50; ***Score 3***: 50–80; ***Score 4***: >80.

The difference in score for each variable from the optimal "Productive Target" pattern was represented using a spider graph for each treatment (R, I, N, IN, L) and vigour (HV, LV) zone and an average of the three seasons (2019, 2020 and 2021) (Figure 5).

RESULTS

1. Weather and microclimatic conditions

The monthly water demand (ETo) and supply (rainfall) for the study area are presented in Supplement 3. The crop seasons markedly differed in terms of water availability. Notably, 2019 was a rainy year with 885 mm over the season, while 2020 and 2021 were the drier years, with accumulated rainfall over the cropping season of 484 mm and 539 mm, respectively. In addition, the distribution of rainfall within each season varied. In 2019 and 2021, up to 57 % of the rainfall occurred during the ripening period. In contrast, in 2020, 60 % of rainfall occurred from bud break to flowering, while the period from flowering to harvest was drier. Years also differed in terms of reference evapotranspiration (ETo). The accumulated ETo over the season reached 806 mm in 2019, 853 mm in 2021 and up to 903 mm in 2020. Ultimately, the climatic water balance (accumulated rain – accumulated ETo) over the cropping season was positive in 2019 (79 mm) and negative in 2020 (–419 mm) and 2021 (–300 mm).

The effects of the water, nitrogen and leaf removal treatments on the canopy microclimate were evaluated by averaging the daily evolution (from flowering to harvest) of temperature, relative humidity, and light in the bunch zone for the wet and dry years, respectively 2019 and 2020 (Figure 2).

The daily dynamics of mean air temperature (Tm) increased from sunrise to a maximum value at 4 pm for both years (Figure 2.1). The maximum temperature was 25 °C in 2019 (Figure 2.1A), while in 2020, it was 27 °C (Figure 2.1C). For each vigour zone, the mean canopy temperature (Tmc) differed between the treatments in 2019 and 2020 (Figure 2.1). In the HV (Figure 2.1AC), the H-W treatment had the

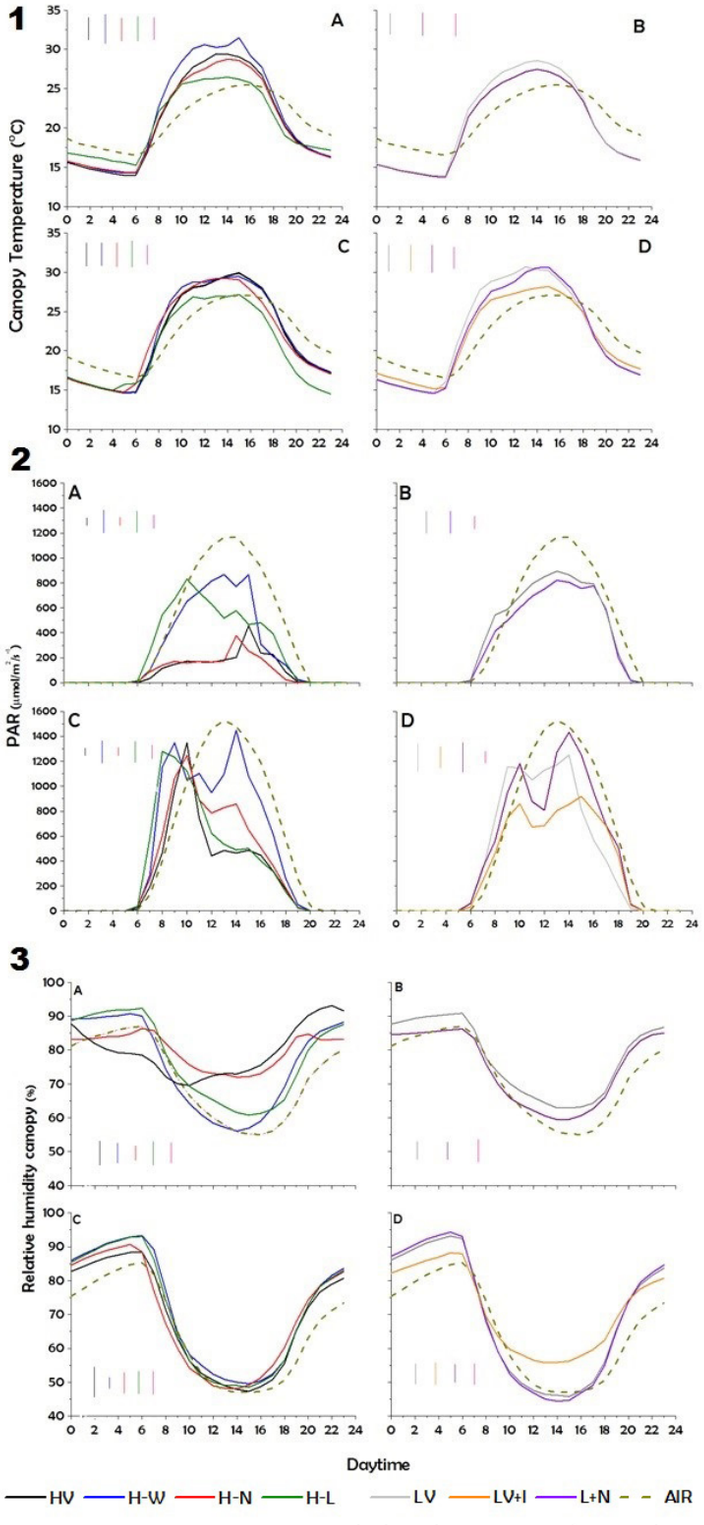


FIGURE 2. Average daily dynamics of weather and microclimatic variables in the bunch zone from bloom (November) to harvest (March) for the different treatments (water, nitrogen, leaf removal) applied in the two vigor zones, high vigor (left figures) and low vigor (right figures); and for two years, 2019 (A and B) and 2020 (C and D). The weather (air) and microclimatic (canopy) variables are the average temperature (1); the photosynthetically active radiation (PAR) (2) and relative humidity (%) (3). The vertical lines in graphs correspond to the average interval of confidence.

highest average Tmc, with temperatures reaching 30 °C from 11 am to 4 pm. The H-L treatment is the one that presented the lowest average Tmc, similar to the air temperature. The other treatments (HV and H-N) showed intermediate Tmc. In LV (Figure 2.1BD), Tmc was higher than the average air temperature during the two years. However, the L+W treatment had lower Tmc compared to LV and L+N, and it was closer to the air temperature.

The incident light (PAR) reached up to 1167 and 1525 µmol/m^2/s, respectively, in 2019 and 2020 (Figure 2.2). The light within the canopy differed between the treatments for the two years. In HV (Figure 2.2AC), the H-W and H-L treatments increased the radiation by about 300 µmol/m^2/s in the canopy compared to the HV and H-N treatments. In the LV (Figure 2.2BD), no differences were observed between the LV and L+N treatments. However, the L+W in 2020 lowered the canopy's radiation by about 150 µmol/m^2/s compared to LV and L+N treatments.

The daily dynamics of relative humidity (RH) presented a maximum value at 6 am and a minimum value at 4 pm (Figure 2.3). In 2020, the minimum RH value was 47 %, while in 2019, this minimum RH value was 55 %. High differences between the treatments were observed for the HV zone in 2019 only. The HV and H-N treatments reached the highest RHc (22 % at 4 pm), while H-W and H-L treatments presented similar RHc dynamics compared to the air RH. In the LV (Figure 2.3BD), the LV and L+N treatments showed similar RHc dynamics compared to the air RH for both seasons. In 2020, the L+W treatment resulted in higher RHc (55 %) than LV and LV+N (45 %) at midday.

2. Soil and plant nitrogen and water status

At the beginning of the trial (2018), N stocks were higher for the HV than the LV (137 and 104 kg/ha, respectively) (Figure 3A). Within each zone, treatments did not differ from each other (p-value ≤ 0.05). Nitrogen restriction in the HV lowered N stock for both the second and third seasons (reduction of 30 %). Likewise, the increased N in the LV (Figure 3B) permitted to increase in soil content (32 % higher), reaching similar values to those observed at the HV. The leaf nitrogen content (%Nl) at veraison during 2019 did not differ between the treatments within the HV (Figure 3C). In contrast, %Nl was 15 % lower for nitrogen restriction in HV compared to the other treatments in 2020. The L+N treatment had a higher %Nl (+37 %) than the LV control in 2019 (Figure 3D). In 2020, the L+W treatment was permitted to increase by 60 % the %Nl compared to the LV control, while the LV and L+N treatments did not differ from each other (Figure 3D).

The seasonal dynamics of plant water status showed differences between years (Figure 4). In 2019, the predawn water potential (Ψp) was higher (> –0.46 MPa) compared to 2020 (> –0.85 MPa). The values of Ψp from flowering to veraison for HV in 2019 were high (> –0.25 MPa), regardless of the treatments, due to high rain. However, from veraison onwards, H-W differed, from the other treatments, reaching the most negative values (–0.4 MPa at harvest). In 2020, Ψp of HV progressively decreased after flowering for all

treatments to reach –0.8 MPa at harvest for all treatments. The LV displayed higher levels of water deficit compared to HV. In 2019, the dynamics of Ψp were similar for LV and L+N treatments, and those treatments reached similar levels of water deficit, such as H-W (–0.41 MPa at harvest). In 2020, while L+W and L+N treatments reached minimum Ψp at harvest of –0.85 MPa, the irrigation treatment (LV+I) permitted to maintain the values of Ψp above –0.35 MPa through the whole seasons.

3. Vegetative growth

Differences in total leaf area (TLA) at veraison were observed between vigour zones, treatments, and years (Table 2). The total leaf area was greater for the three years on HV than on LV. The differences between HV and LV were the most pronounced for the years with higher water supply (2019 and 2021). In the HV, the H-L treatment reduced TLA by about 25 %. This decrease in leaf area was mainly due to the lower main leaf area (MLA). It should be noted that this H-L treatment also changed the canopy architecture as leaf removal was specifically performed in the bunch zone.

No difference in TLA was observed for other treatments. For LV, the L+W treatment increased TLA in 2020 and 2021 through an increase in both the primary and secondary leaf area (MLA, SLA). The L+N treatment only increased leaf area in 2019 and was similar to other treatments of LV for the other years. The L+WN treatment did not differ from the L+W treatment for vegetative growth variables.

The % lignification at harvest also showed differences between the vigour zones and treatments. For HV, the H-L treatment presented higher lignification values (> 80 %) compared to the rest of the treatments (<63 %) for the three years evaluated. For LV, the irrigation treatments, L+W and L+WN, permitted to increase in the % of lignification compared to the control (LV).

The pruning weight (PW) varied between the years and between vigour zones. The highest values were recorded in 2019, followed by 2020, and finally in 2021. HV had higher PW values (463 to 635 g/vine) compared to LV (172 to 395 g/vine).

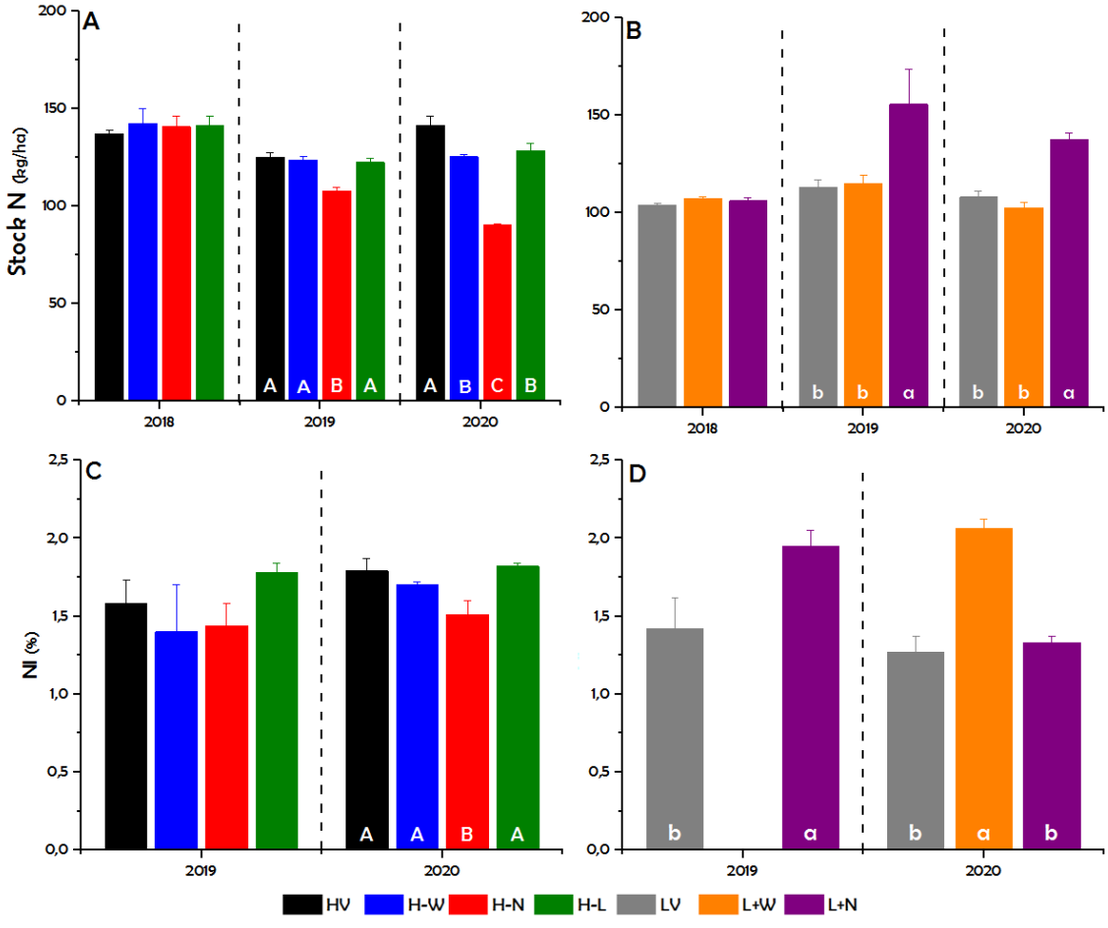


FIGURE 3. Soil and plant nitrogen dynamics. A and B: Soil Stock N at 0-40 cm (kg/ha) before budbreak according to the treatments (water, nitrogen, leaf removal) applied for the three cropping seasons (2018-2020). C and D: Percentage of nitrogen in leaves (%Nl) at veraison according to the treatments (water, nitrogen, leaf removal) for two cropping seasons. A and C: High vigor; B and D: Low vigor. In both graphs (1, 2), the bars represent the mean values (3 soil or leaf samples per treatment) and the error bars represent standard deviation. Different letters indicate significant differences according to the Tukey test (p-value < 0.05), within each vigor zone and year evaluated.

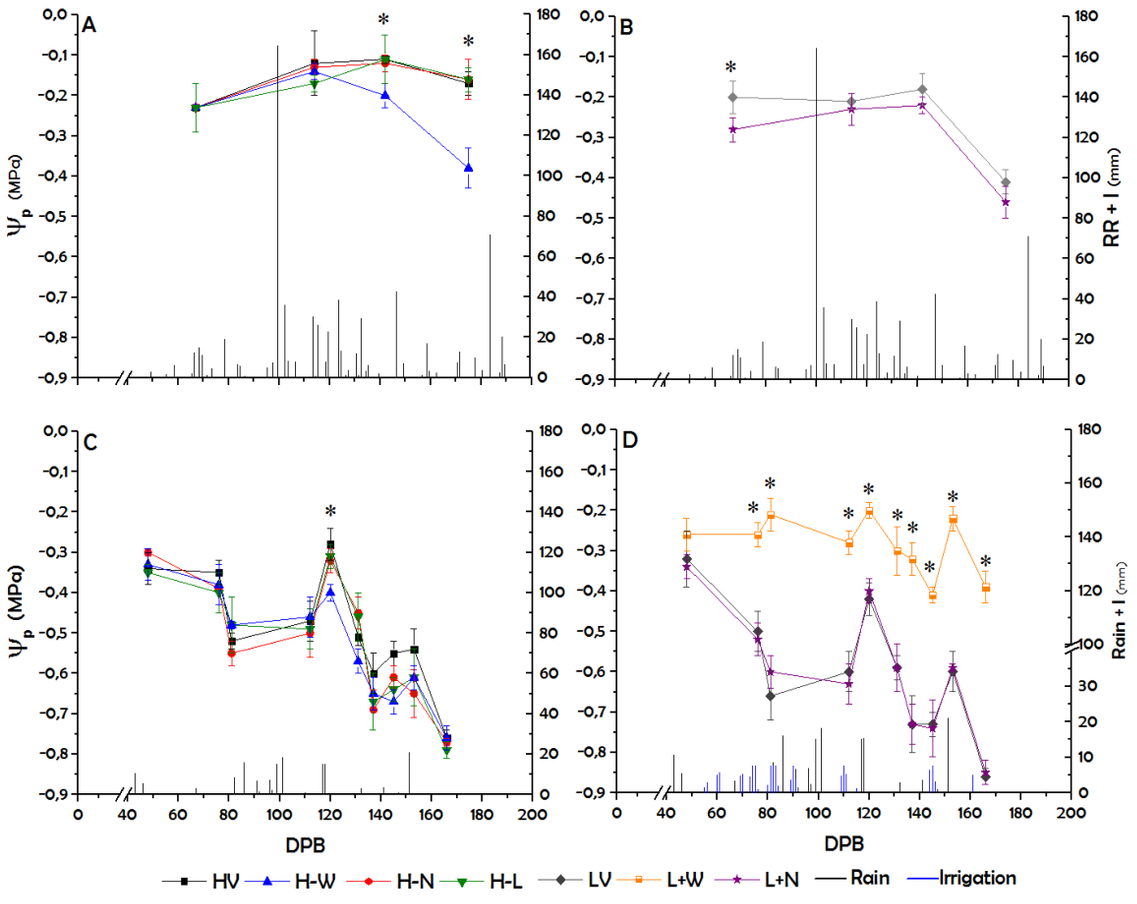


FIGURE 4. Changes of the predawn water potential (Ψp) according to the treatments (water, nitrogen, leaf removal) applied in the two vigor zones (high vigor, low vigor) for two cropping seasons (2019-2020). High vigor zone treatments in 2019 (A) and 2020 (C); Low vigor zone treatments in 2019 (B) and 2020 (D). In all graphs, the dots represent the mean values (10 leaf samples per treatment) and the error bars represent standard deviation. *Asterisks indicate significant differences (p-value < 0.05).

The treatments modified PW in 2020 and 2021 of LV only, with higher values for L+W (87 %) and L+WN treatments compared to LV and L+N.

4. Yield components

Yield clearly varied between the years and the vigour zones. The yield per vine was higher in 2019 and 2020 compared with 2021 and higher for HV compared to LV except when irrigation was supplied in LV.

In HV, H-L had the lowest yield in spite of higher berry weight for the 3 years. The H-N treatment was as high as HV and H-W in 2019 but as low as H-L in 2020 due to the reduction in berry weight. The incidence of bunch diseases reached up to 8 % for H-N and up to 17 % for HV in 2019, while it was lower than 2.5 % for all other treatments and years in this HV zone.

For LV, the L+N treatment significantly increased yield (16 %) compared to LV in 2019 only. For other years, the L+N treatment did not differ from the LV treatment for any yield variables. In contrast, the two irrigation treatments with or without additional N supply (L+W and L+WN) permitted to increase in the yield per vine (+80 %) compared with the control (LV) through the higher bunch and individual berry weights. Disease incidence was low (< 2.2 %) for all treatments of LV, regardless of the year.

5. Berry composition

Differences between years and between the treatments for each vigour zone were observed for both primary and secondary metabolite concentrations in the berries (Table 4). For the primary metabolism, higher total soluble solids and total acidity concentrations were observed in 2019 and 2020 compared to 2021, while the pH and the YAN were the lowest in 2020 in all treatments. The H-W and H-L treatments permitted to increase in the sugar concentration compared to HV, although this increase was not systematic for all years. The pH was, in contrast, lower for H-W (2.78) and H-L (2.97) than HV (3.00) in 2020. The H-N treatment decreased the YAN content compared to all other treatments in HV in 2019 and 2020 (–24 %). In LV, the L+N treatment was permitted to reach higher YAN content (+20 %) than LV in 2019 and 2021 but had no effect on other primary metabolism variables. The YAN increase was even more (+60 %) when irrigation (with or without N) was applied (2020–2021). The L+W treatment also led to a higher sugar concentration and higher pH compared to the other treatments.

TABLE 2. Average values of vine total leaf area at veraison, cane production in winter and % of lignification at harvest according to treatments (water, nitrogen, leaf removal) applied in the two vigor zones (high vigor, low vigor) for the three cropping seasons.

	Treatments	TLA (m^2/vine)	MLA (m^2/vine)	SLA (m^2/vine)	Lignification (%)	PW g/vine
2019	HV	9.3 a	4.7 a	4.5 a	62 b	635
	H-W	8.3 a	4.3 ab	4.0 a	58 b	624
	H-N	8.2 a	4.8 a	3.4 ab	62 b	501
	H-L	6.9 b	3.8 b	3.1 b	87 a	572
	LV	3.1 b	1.9 b	1.2 b	49	395
	L+W	n.d.	n.d.	n.d.	n.d.	n.d.
	L+N	5.2 a	3.4 a	1.9 a	65	415
	L+WN	n.d.	n.d.	n.d.	n.d.	n.d.
2020	HV	3.8 a	3.1 a	0.7	60 b	546
	H-W	3.5 a	3.0 a	0.5	49 b	505
	H-N	3.1 a	2.6 a	0.5	51 b	477
	H-L	2.9 b	2.4 b	0.5	85 a	465
	LV	1.7 b	1.5 b	0.2 b	53 b	341 b
	L+W	5.3 a	4.6 a	0.7 a	74 a	505 a
	L+N	1.7 b	1.5 b	0.2 b	68 b	299 b
	L+WN	n.d.	n.d.	n.d.	n.d.	n.d.
2021	HV	7.3 a	5.9 a	1.3	63 b	463
	H-W	n.d.	n.d.	n.d.	n.d.	n.d.
	H-N	n.d.	n.d.	n.d.	n.d.	n.d.
	H-L	5.8 b	4.4 b	1.4	82 a	459
	LV	3.5 b	2.7 b	0.8	57 b	172 b
	L+W	7.9 a	6.4 a	1.5	66 ab	392 a
	L+N	3.9 b	3.0 b	0.9	57 b	213 b
	L+WN	7.2 a	5.9 a	1.3	76 a	351 a

TLA: Total leaf area. MLA: Main leaf area. SLA: Secondary leaf area. PW: Pruning Weight. Different letters correspond to significant differences according to the Tukey test (p-value ≤0.05). n.d.: No data available.

Regarding the secondary metabolites, 2019 and 2020 were marked by higher total anthocyanin concentrations (A) and total phenol index (TPI) in the HV zone compared to the LV zone, while the situation was the reverse in 2021.

In HV, H-L was the treatment with the highest A and TPI at harvest for all years. In contrast, the H-N treatment reached similar A and TPI as HV in 2019, but it resulted in lower concentrations than HV in 2020. For the LV, the L+W and L+WN treatments showed the highest A in 2020 and 2021 and also the highest TPI in 2020 compared to all other treatments. To a lesser extent, the L+N treatment also permitted to increase A compared to HV in 2020 and 2021 but had no effect on TPI compared to the control.

6. Relevance of the treatments when compared to an optimal "Productive Target" pattern

For both vigour zones, the treatments applied generally permitted over the three years to improve the score of a few variables compared to the controls (HV, LV), but without systematically reaching the optimal score (≥ 3) (Figure 5).

In the high vigour zone, the control HV was sub-optimal for all variables, except for the yield (Y), which reached a maximum score (score 4). Interestingly, treatments such as Leaf removal (H-L) and water restriction (H-W) presented a score of 4 for BR (no disease at all) and equalled the optimal "Productive Target" limit (score 3) for TPI, A and TSS. However, the variables PW and YAN remained sub-optimal (score 2) as for HV. Regarding yield (Y), the H-L presented a lower performance than HV (score 2), while the H-W surpassed the "Productive Target", such as HV (score 4). Similar to HV, the nitrogen reduction treatment (H-N) had higher yields than the "Productive Target", but it had lower scores than the "Productive Target" for the other selected variables. In particular, the H-N treatment reached the worst score (score 1) for YAN.

In the low vigour situation, the control LV reached optimal scores for Y and BR, but all other variables were sub-optimal.

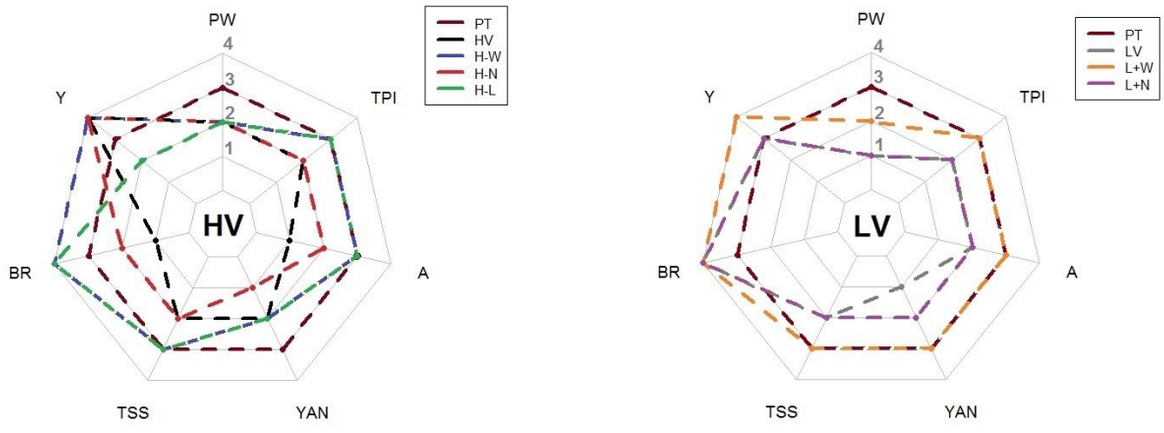


FIGURE 5. Deviation from an optimal 'Productive Target' pattern for the vineyard sustainability and profitability of the different treatments within HV and LV zones over the three years (2019 to 2021).

The optimal 'Productive Target' (score ≥ 3) is delimited by the ruby line (corresponding to score 3) for each of the 7 selected variables. PT: Productive Target; TSS: Total soluble solid; PW: Pruning Weight; Y: Yield; BR: Bunch root; YAN: Yeast assimilable nitrogen; A: total anthocyanins. TPI: total phenol index. Scores 1 and 2 were considered to be non-optimal.

The treatment with water supply (L+W) permitted to reach the "Productive Target" for all variables, except the PW variable, whose score was only 2. The contribution of N (L+N) was only permitted to favour the score of YAN compared to LV, while all other variables reached the same score as LV.

DISCUSSION

1. Changes in water and nitrogen status determine the difference in plant vigour

The productive seasons were climatically different, particularly in water supply (Supplement 3). At the time of flowering in 2019, both vigour zones were confronted with a moderate deficit level (Figure 4) due to a dry and warm spring (Supplement 3). From flowering onward, rainfall generated different water patterns according to vigour zone and treatments. The treatments recovered a good water status in the HV and remained without stress until harvest, except for the H-W treatment, while the Ψp for both LV and L+N treatments progressively declined. Inorganic mulches, such as the one used in the H-W treatment, are expected to generate a physical barrier that prevents soil water loss by evaporation and evapotranspiration through the limitation of cover crop development (Hostetler *et al.,* 2007; Ross, 2010). In our study, H-W was more stressed (Figure 4A) than control plants (HV) in 2019, despite the high rainfall. The barrier effect of the plastic mulch, preventing the entry of rainwater, coupled with the dry conditions prior to mulching, may be responsible for the low plant water status measured after flowering. In both seasons, the plastic mulch was placed after seven days without rainfall and high atmospheric demand, which caused soil moisture to drop to moderate to high-stress levels until close to harvest. For the LV and L+N, plants appeared only slightly responsive to rainfall events (Figure 4B). When controlled deficit irrigation management (L+W) was applied, plant water status was efficiently maintained at high values of Ψp (ranging from –0.2 to –0.4 MPa), according to Ojeda *et al.* (2002). The higher soil water holding capacity in HV compared to LV could explain these differences in plant water status, as reported by other authors (Bramley *et al.,* 2011; Ferrer *et al.,* 2020a).

The applied treatments modified the soil and plant nitrogen availability (Figure 3). The H-N treatment reduced foliar N starting from the second productive season (2020). The absence of effect during the first year of treatment (2019) was likely to be due to the buffering effect of N (Verdenal *et al.,* 2021). The soil N stock for the H-W treatment was also lower compared to HV in 2020. The reduction in soil moisture for this treatment in 2019 (Figure 4A) may have affected the activity of microorganisms and the rate of mineralisation of organic matter (Paul, 2007) for the second season. Similarly, in the LV zone, the effect of the treatments was dependent on the water supply of the year. Supplemental N supply (L+N) increased soil N content for all years but not plant nitrogen content. Nitrogen uptake by roots is dependent on soil water availability (Verdenal *et al.,* 2021). Thus, despite the higher N stock in the soil for the L+N treatment compared to LV in 2020, the low water availability did not permit to increase the leaf N content for this treatment. When water was supplied (L+W treatment), the higher humidity permitted to increase the leaf N content in 2020, probably due to the greater microbial activity and N solubility favouring N absorption (Ortega-Heras *et al.,* 2014).

Water conditions and nutrient supply, particularly N, determine the expression of vigour in interaction with other environmental and cultivation management strategies (Chaves *et al.,* 2007). The timing of fertilisation and also the type of fertiliser (mineral vs organic) plus the mode of application (soil and foliar application) highly impact the response of the vine to fertilisation (Gatti *et al.,* 2020). In our trial, the usual N fertilisation was the same for the whole plot. However, the two vigour zones differed in terms of root development, the latter being less abundant and more superficial in the LV compared to HV (data not shown).

Supplementary water and N supplies in the LV zone (L+W and L+WN) increased the vegetative growth (Table 2) and even exceeded the one observed for HV in 2020. It is well documented that shoots, in particular the secondary shoots (Metay *et al.,* 2014), are the most responsive organs to water and N availability (Chaves *et al.,* 2007; Vrignon-Brenas *et al.,* 2019). Excessive N applications (with water availability) can also generate excessive vigour, altering the microclimate of clusters together with the grape composition and sanitary status (Metay *et al.,* 2014; Soubeyrand *et al.,* 2014). It is clear, based on our results when considering the treatments L+W, L+WN and L+N, that water was the limiting factor and that N application without water availability had no impact on vegetative growth. The H-L treatment and the LV irrigated treatment both improved pruning weight and lignification, which may be related to a higher source:sink ratio and starch accumulation (Vrignon-Brenas *et al.,* 2019).

2. Changes in yield and grape composition

The H-L treatment reduced the yield for the three cropping seasons compared to the HV control (Table 3). Many authors have reported the effect of pre-flowering leaf removal on yield reduction for several varieties (Palliotti *et al.,* 2011; Arrillaga *et al.,* 2021; Chalfant and Dami, 2021). The restriction of carbohydrate supply due to the removal of photosynthetically active leaves during the flowering period can be critical for berry set and development (Candolfil-Vasconcelos and Koblet, 1990; Frioni *et al.,* 2018). The yield reduction was explained in the present study mainly by the reduction in berry number per bunch in accordance with other studies (Arrillaga *et al.,* 2021). Although yield was lower for the H-L treatment compared to HV, berry weight was consistently higher for the three years in the H-L treatment. The lower number of berries per cluster from the early stages of berry development and the better soil water availability in this vigour zone may have favoured the carbon balance and berry growth (Ojeda *et al.*, 2002). The higher water and nitrogen availability over the two seasons of yield elaboration were shown to have a positive impact on carbon gain and on all yield components in several studies (Ojeda *et al.,* 2002; Dos Santos *et al.,* 2003; Vasconcelos *et al.,* 2009;

TABLE 3. Average values of yield components according to the treatments (water, nitrogen, leaf removal) applied in the two vigor zones (high vigor, low vigor) for the three cropping seasons.

	Treatments	Yield (kg/vine)	Number of bunches	Bunch weight (g)	Berry weight (g)	Part of the cluster affected by bunch rot (%)
2019	HV	6.9 a	20.3	344 a	1.72 b	17 a
	H-W	6.6 a	21.3	320 a	1.70 b	0.0 c
	H-N	6.8 a	22.0	290 ab	1.71 b	7.9 ab
	H-L	5.1 b	20.2	259 b	1.85 a	0.9 b
	LV	5.3 b	25.7	214 b	1.57	2.2
	L+W	n.d.	n.d.	n.d.	n.d.	n.d.
	L+N	6.2 a	27.2	227 a	1.60	0.8
	L+WN	n.d.	n.d.	n.d.	n.d.	n.d.
2020	HV	6.7 a	26.4	251 a	1.34 b	0
	H-W	6.5 a	29.3	227 ab	1.33 b	0
	H-N	5.6 b	25.5	217 bc	1.18 c	0
	H-L	5.2 b	27.2	191 c	1.44 a	0
	LV	4.7 b	30.2	161 b	1.02 b	0
	L+W	8.3 a	30.1	283 a	1.53 a	0
	L+N	4.2 b	31.1	145 b	0.95 b	0
	L+WN	n.d.	n.d.	n.d.	n.d.	n.d.
2021	HV	5.8 a	25.1	228 a	1.42 b	2.5 a
	H-W	n.d.	n.d.	n.d.	n.d.	n.d.
	H-N	n.d.	n.d.	n.d.	n.d.	n.d.
	H-L	4.6 b	27.1	198 b	1.65 a	0.1 b
	LV	4.1 b	30.1	140 b	1.14 b	0.1 b
	L+W	7.9 a	29.8	266 a	1.50 a	0.52 a
	L+N	4.0 b	29.1	139 b	1.08 b	0.1 b
	L+WN	7.9 a	31.3	251 a	1.56 a	0.50 ab

Different letters correspond to significant differences according to the Tukey test (p value ≤0.05). n.d. No data available.

Guilpart *et al.,* 2014). In accordance with those studies, the regulated deficit irrigation strategy in the LV (L+W and L+WN), which increased plant water and N status, permitted one to improve grape yield mainly through an increase in bunch weight (Table 3).

Higher light exposure was also concomitant with higher bud fertility (Figure 2, Table 3), as reported by Sánchez and Dokoozlian (2005). The treatments H-W, H-L, LV and L+N generated an environment with greater luminosity, higher temperature, and lower relative humidity compared to HV (Figure 2). Such conditions permitted the reduction of the pressure of pathogenic fungi. The lower leaf area for these treatments (Table 2) and the better exposure of the bunches may also have improved the efficacy of chemical control (Molitor *et al.,* 2016). Grey mould incidence was shown to be reduced on grapevines with lower vegetative and reproductive growth (Valdés-Gómez *et al.*, 2008).

In this trial, the harvest date was fixed according to the evolution of pH (3.3–3.4). In addition, when these pH values were not reached for some years (2020 and 2021), priority was given to avoiding dehydration of the berries (weight loss) for each treatment. It is important to note that the treatments were harvested (Supplement 2) at maximum berry weight, thus avoiding a concentration increase in berry components resulting from berry shrivelling. Thus, at maximum bunch weight, H-L treatment improved the accumulation of sugars, anthocyanins, and phenols in HV compared to the HV control (Table 4).

The effect of pre-flowering leaf removal on grape composition is controversial in the literature. Some authors indicate an improvement in all or selected grape composition parameters (Palliotti *et al.,* 2011; Gatti *et al.,* 2012; Arrillaga *et al.,* 2021), while others report no change in grape quality (Chalfant and Dami, 2021). In our study, the lower yield for H-L treatment increased the leaf/fruit ratio and increased the light exposure in the bunch zone, which ultimately stimulated the berry growth, sugar accumulation, anthocyanin and phenol contents (Risco *et al.,* 2014, Arrillaga *et al.,* 2021). Using plastic ground cover (H-W) also improved grape composition parameters. Notably, the soluble solids and anthocyanin contents were higher, as reported by other authors (Todic *et al.,* 2008). In contrast, there was no effect

TABLE 4. Average values of berry composition according to the treatments (water, nitrogen, leaf removal) applied in the two vigor zones (high vigor, low vigor) for the three cropping seasons.

	Treatments	TSS (g/l)	Total acidity (g l-1 sulfuric)	pH	YAN (mg L^{-1})	A (mg/l)	TPI
2019	HV	213 c	4.4	3.34	147 a	2098 b	46 b
	H-W	230 a	4.2	3.32	150 a	2511 a	52 a
	H-N	218 bc	4.5	3.31	135 b	2054 b	49 b
	H-L	222 b	4.4	3.31	145 a	2646 a	52 a
	LV	200	4.2	3.37	109 b	2110	48
	L+W	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	L+N	207	4.3	3.33	128 a	2350	44
	L+WN	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
2020	HV	226 a	4.5	3.00 a	78 a	2091 b	67 b
	H-W	209 b	4.4	2.78 c	75 a	1905 c	69 b
	H-N	208 b	4.5	3.04 a	48 b	1568 d	53 c
	H-L	225 a	4.4	2.96 b	73 a	2321 a	85 a
	LV	203 b	4.5	2.93 b	67 b	1692 c	61 b
	L+W	223 a	4.4	3.10 a	125 a	2425 a	77 a
	L+N	202 b	4.8	2.80 b	73 b	1914 b	60 b
	L+WN	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
2021	HV	172 b	3.9	3.21	104	944 b	36 b
	H-W	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	H-N	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	H-L	208 a	3.9	3.20	106	1979 a	48 a
	LV	197 b	4.1ww	3.14	110 d	1694 c	52
	L+W	212 a	4.3	3.19	162 b	2110 a	57
	L+N	201 b	4.2	3.09	138 c	1938 b	52
	L+WN	200 b	4.6	3.10	184 a	2041 ab	50

TSS; Total Soluble Solid. YAN: Yeast assimilable nitrogen. A: total anthocyanins. TPI: total phenol index. Different letters correspond to significant differences according to the Tukey test (p-value ≤0.05). n.d. No data available.

of H-W on grape pH or acidity at harvest, in agreement with Hostetler *et al.* (2007) and Sandler *et al.* (2009). The effect of plastic ground cover on berry composition was particularly interesting in 2019, which was climatically less favourable (high rainfall during the ripening period). In addition to its impact on plant water status, the plastic cover permitted to increase the light reflection in the cluster zones as mentioned above (Figure 2) but also changed the wavelength (Osrečak *et al.*, 2016). Changes in the ratio of red to far-red radiation (R:FR) can occur depending on the type and colour of mulch used (Ross, 2010). The material we used presented a high diffuse reflectance (50 %, data not shown) in the wavelength belonging to the red (600 nm), improving the R:FR ratio. Therefore, this mulch could also have modified the expression or activation levels of the enzymes responsible for the biosynthesis of the molecules (Smart *et al.,* 1988).

Keeping plants under moderate water deficit (L+W) stimulated the accumulation of sugars and anthocyanins (Table 4) despite the larger leaf area found compared to LV (Table 2). Our results show that additional water supply seems to have played a more critical role in the accumulation of primary and secondary compounds (Ojeda *et al.*, 2002) than bunch exposure. Temperature and light in the canopy zone were lower for L+W compared to LV (Figure 2) due to higher vegetative growth (Table 2), but the accumulation of sugars and anthocyanins was favoured with a regulated deficit irrigation strategy (Table 4). In the seasons when water was more available (2019 and 2021), nitrogen supplementation (L+N) also improved the berry composition by increasing the total anthocyanins and YAN (Figure 3 and Table 3).

3. Valorisation of site-specific management

The spatial variability of vineyards is generally due to variations in soil characteristics (Taylor *et al.,* 2010; Arnó *et al.,* 2012; Brillante *et al.,* 2016), which affect the availability of water and nutrients. Such spatial variability was characterised by our vineyard in previous work (Ferrer *et al.,* 2020a) (Supplement 1), thus permitting us to set a boundary for two vigour zones over eight successive years of observations.

We proposed a strategy based on site-specific management, where differential cultivation techniques (irrigation, fertilisation, leaf removal) are applied according to vigour zones to optimise production and quality and increase input use efficiency (McClymont *et al.,* 2012). Water management, either with a regulated deficit irrigation strategy (L+W, L+WN) or with the use of plastic covers (H-W), allowed to achieve adequate yields with improvement in grape composition parameters (Table 4), reduce the incidence of bunch diseases and reach "Productive Target". Other studies, in agreement with our results, confirm that water management through irrigation scheduling by vigour zone and in combination with cover crops can improve water use efficiency and promote both yield and grape composition (McClymont *et al.,* 2012). Nitrogen reduction in an HV determined that the productive and compositional variables moved away from the "Productive Target" pattern (Figure 5).

The treatments H-L and L+W were the closest to the "Productive Target" for the Tannat variety and the treatments that also favoured the within subplot and between year homogenisation of all the key variables evaluated (PW, Y, TSS, BRIX, YAN, A and TPI). In addition, it should be noted that L+W treatment was able to achieve higher yields than targeted with the high berry quality levels setting in the "Productive Target".

The benefits of site-specific management can be evaluated from an environmental and economic point of view. Reasoning the inputs where and when needed brings environmental advantages compared to more 'traditional and uniform' management and reduces production costs (Arnó *et al.,* 2008). A few examples include reduced pollution due to less nutrient loss (Bongiovanni and Lowenberg-Deboer, 2004), reduced water use, and less pesticide application (Hedley, 2015), among others. Economic benefits may be more difficult to quantify. In our study, treatments showed differences compared to a "Productive Target" situation necessary to obtain good quality wines (Figure 5). H-L and L+W treatments showed the potential to produce good quality wines even with higher yields and better bunch health than HV and LV. In addition, both yield and composition parameters were more homogeneous, a situation that is desirable for winegrowers. Without intervention using management techniques, wines that would be produced from the control grapes (HV and LV) would be impossible to sell under the category of quality wines according to Uruguayan regulations because of their low alcohol level (less than 12 %). Therefore, their sale price would be automatically significantly lower than that of quality wine (USD 2.5 vs USD 7.2; 750 ml bottle). In rainy years, the hydric reduction treatment improved the sanitary condition of the clusters, improving the composition parameters and, therefore, the wine produced. Although nylon can be used for several seasons, the installation, maintenance, and removal costs are high, and the environmental impact of using non-degradable plastics must also be considered (Hostetler *et al.,* 2007).

To conclude, while a 1 ha plot can be considered homogeneous from a topographic and soil-climatic aspect, this study has demonstrated high variability of soil, production parameters and grape composition. It provided knowledge on the relationships between water and nitrogen availability, plant vigour and their impact on grape quality. Applying precision viticulture technologies, we were able to experimentally apply site-specific management strategies that improved plant productivity (yield and berry composition) and reduced heterogeneity at the plot level. This approach could be used by winegrowers on a larger scale to determine micro-terroirs and thus generate the application of site-specific techniques to obtain the potential for productivity.

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