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Assessment of map based variable rate strategies for copper reduction in hedge vineyards

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ABSTRACT

Reduction of Plant Protection Products (PPP) has become a priority in agriculture, led by European directives and regulations due to the negative impacts and social awareness that pesticides have raised in the population. In viticulture, the use of copper to control Downy Mildew (*Plasmopara viticola*) is posing severe problems of soil, water and environment contamination, and European Commission is regulating its use through the EU regulation 2018/1981. A targeted spraying management can be done benefiting from the novel development of variable rate application (VRA) technology for orchard sprayers, resulting in savings of PPP. In this research, the quality of the application in terms of copper deposit in leaves of two VRA strategies where compared versus the conventional strategy performed by the farmer (REF). In each of the two VRA strategies, copper dosage was selected 1) maintaining copper dose per hectare (VRA) and 2) following the copper concentration listed in the product label ($VRA_{[Cu]}$). Drone based NDVI maps from three commercial vineyards were used to characterize the structure of the vegetation in every vigour zone and manual measures in field were used to determine the most appropriate spraying volume rate according to canopy characteristics.

Results show that a reduction of 33 and 44 % of copper used per hectare did not have significant impact on the quality of the application. On the contrary, while leaf deposit was comparable to that achieved by REF strategy, the product was better distributed across the entire canopy. Savings of 40 % of copper could be achieved in in the PPP application calendar along the season 2019 with no influence in disease control. Impact of PPP can be reduced by incorporating digital technologies, and the objectives of the Farm to Fork strategy and the European Green Deal can be met ensuring a more sustainable agri-food production.

1. Introduction

Plant Protection Products (PPP) have become essential to ensure a successful yield in viticulture fulfilling the demanding on quality standards. PPP are particularly important and intensively used in specialty crops, and more specifically in vineyard, accounting for the 48 % of the total active ingredient use in the EU although the crop barely occupies 7 % of the agricultural land (EUROSTAT 2002). In particular, fungicide applications account for the largest share of PPP treatments averaging 12–15 spraying applications per season (Pertot et al., 2017). One of the most common fungicides used in viticulture is copper which controls Downy Mildew caused by the oomycete *Plasmopara viticola* (Berk. & Curt.) Berl. & de Toni (Gessler et al., 2011; Cabús et al., 2017). Thus, copper in viticulture is both necessary and its use restricted due to the hazardous impact in the environment (REG 2018). Furthermore, in general terms, the use of pesticides is under a great concern for the European Commission which is elaborating a new regulation for the Sustainable Use of Pesticides (EU 2009), and fixing reduction rates of 50 % in PPPs by 2030 according to the Farm to Fork strategy implemented under the European Green Deal (COM 2019).

More targeted and sustainable spray applications are necessary for increasing PPP efficacy and efficiency. Canopy characteristics is the factor that most critically influences the calculation and selection of the optimal volume rate (and consequently the pesticide dose) to be applied in every moment along the growing season, and it is related to the quality of the spray application (Walklate et al., 2011; Codis and Douzals, 2012; Gil et al., 2014; Garcerá et al., 2017; Xun et al., 2022). Traditionally, canopy has been considered homogeneous within the vineyard, and consequently, the volume rate applied has been maintained uniform when spraying PPP (Peteinatos et al., 2013). However, it

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is well known that certain amount of variability is present in the vineyard, often induced by factors linked to soil (Martinez-Casasnovas et al., 2012). Special care should be taken under spatially variable vineyards to avoid overdosing of low vigor vines or a lack of coverage or penetration in the most vigorous zones of the plot. Imprecise or excessive use of pesticides can lead to serious problems such as traces of pesticides in food, health issues in wildlife and humans and, in the particular case of copper, intensive use of this active matter in viticulture is leading to contaminated soils (Komárek et al., 2010) causing severe environmental damage.

Accurate canopy characterization is often linked with the growing implementation of variable rate pesticide application, and it has been attempted using different sensing technologies with the objective of increasing productivity and efficiency of this process. Ground sensors such as vision, spectral and ultrasonic systems, as well as laser scanning have been broadly studied providing reliable results for canopy characterization in vineyard (Rosell et al., 2009; Llorens et al., 2011; Arnó et al., 2013; Sanz et al., 2018), and development of real time variable rate prototypes allowed reducing the liquid applied in 58 % (Gil et al., 2007; Llorens et al., 2010) and 21.9 % (Gil et al., 2013) as compared with conventional sprayers. More recently, unmanned aerial vehicles (UAV), providing flexibility for flight planning, reliability and affordability, have allowed mapping large areas with high spatial resolution. Information provided by UAV have been used for canopy characterization in vineyards using known vegetation indices in a 2D environment (Poblete-Echeverría et al., 2017; Campos et al., 2019; Campos et al., 2021) or more sophisticated methodologies based on photogrammetry in 3D (Ballesteros et al., 2015; Weiss and Baret, 2017; Mathews and Jensen 2013; Mathews 2015). Knowing the structure of vigor variability within the plot, and having successfully adjusted the optimal volume rate for every zone in the field, various spray parameters (working pressure, nozzle flow rate, etc.) can be modified according to the canopy characteristics, while maintaining a constant application rate per unit of canopy.

Although variable rate systems for vineyards are already in commercial phase, the potential savings which can be achieved by this technology still need to be studied and verified by further field tests. In turn, additional research should be conducted to evaluate whether copper concentration can be successfully reduced while maintaining the deposition and quality of the application in leaves. For this reason, the general objective of this research was to compare variable rate application strategy with conventional practices carried out by farmers in terms of leaf copper deposition and homogeneity of the application. Specific objectives are: 1) to determine if copper deposit in leaves was significantly different for the reference and variable rate approaches, 2) to test if a variable rate application combined with a rational dose expression linked to canopy characteristics allowed to reduce the use of copper without compromising the quality of the application and leaf deposition, 3) to evaluate the real savings in terms of water and copper used and the viability for rapid adoption of such application techniques.

2. Materials and methods

Within this section, the complete process for variable rate application (VRA) of pesticides based on canopy maps is described following the methodology presented in Campos et al. (2019, 2020, 2021). For the consecution of the objectives listed in this study the following steps were accomplished: 1) drone based canopy maps were produced in each of the test sites, 2) site specific canopy characterization and subsequent determination of PPP volume rate according to canopy development and 3) VRA based on prescription maps was compared versus a reference spraying management by assessing copper deposition on leaves.

2.1. Study site

Field trials were conducted during 2019 in the viticulture zone of appellation of origin Penedès, one of the recognized areas of wine and sparkling wine (Cava) production in Spain with more than 16.500 ha of vineyards and around 2,800 producers registered. Experiments were conducted in Torrelavit (X: 391,600.00 Y: 4,588,005.50 UTM31 ETRS89) where three representative commercial parcels of Cabernet Sauvignon comprising an area of 3.84, 3.14 and 1.53 ha were included in the study (Fig. 1). Vineyards were planted at distances of 2.2 m between rows and 1.2 m in the row. Vine ages were 20, 19 and 27 years respectively and were planted in a terrain with moderate slope (5–10 %). Plants were in full production, non-irrigated and trained in a double cordon spur pruning system with green pruning when shoot length was approximately 10 cm. After green pruning, training wires were lifted in order to better define the vegetation wall and to promote canes grow upright position. Weeds were managed by regular harrowing of soil in between canopy rows and under de vines.

2.2. Field spray applications

Pest management was focused on one of the predominant fungal diseases in the area such as downy mildew (*Plasmopara viticola*). All spray applications were based on commercial copper oxychloride 50 % W/W formulates selected by the technicians in charge of the vineyard complying with the indications specified in the product label. Field spray applications were organized coinciding with the pest protection calendar set by the manager of the vineyard, who carried out a total of four applications during the season 2019. Deposition experiments were carried out in two vegetation growth stages namely *beginning of flowering* (BBCH 59) and *beginning of ripening* (BBCH 81) according to the BBCH monograph (Meier 1977). For each of the two growth stages where deposition experiments were carried out (i.e. BBCH 59 and BBCH 81), three different treatments were conducted: 1) reference (REF), 2) variable rate maintaining same copper dose per hectare than REF (VRA) and 3) variable rate following the copper concentration listed in the product label ($VRA_{[Cu]}$). These treatments were carried out in plots A, B, and C respectively (Fig. 1).

2.3. Variable rate map generation

Variable rate prescription maps were generated for plots B and C by assigning a specific volume rate to each vigour class. Vigour classes were based on canopy variability maps produced using high resolution remote sensing with unmanned aerial vehicle (UAV) following the methodology developed and described in Campos et al. (2019, 2020, 2021). Two flight campaigns were performed on the 6th of May, and 22nd of July 2019 coinciding with two of the most important growth stages of vegetation in vineyards. A UAV hexacopter (model: CondorBeta, Dronetools SL, Sevilla, Spain) was used as aerial platform due to its flexibility in terms of flight altitude and cruise speed. The UAV was equipped with a multispectral camera (model: RedEdge, Micasense, Seattle, USA) consisting on a camera matrix of five optics with specific spectral filters centered in the blue (B, 475 nm), green (G, 560 nm), red (R, 668 nm), red edge (RE, 717 nm) and near-infrared (NIR, 840 nm). Filter bandwidth ranged from 10 to 40 nm depending on the spectral band selected, optics focal length was 5.4 mm and sensor 1280x960 pixels (width × height). Flights were conducted 95 m above ground level (AGL) at a cruise flight speed of 6 $m{\cdot}s^{-1}$. Overlapping zones were adjusted at 80 % in the sense of flight and 60 % in the transverse sense.

The image frames acquired were processed with the photogrammetric software Agisoft Metashape (Agisoft LLC, Russia) and five orthophotomaps with a ground sample distance of 6.48 $cm{\cdot}pixel^{-1}$ were generated, one for each spectral filter in the camera. Each orthophotomap was radiometrically calibrated using gray-scale calibration standards of known reflectance (22, 32, 44, and 51 %), which were placed inwards the vineyard, close to the area where the UAV took off and landed, to ensure that they were visible in several frames. From the 12-bit digital value extracted from the calibration plates, a power function

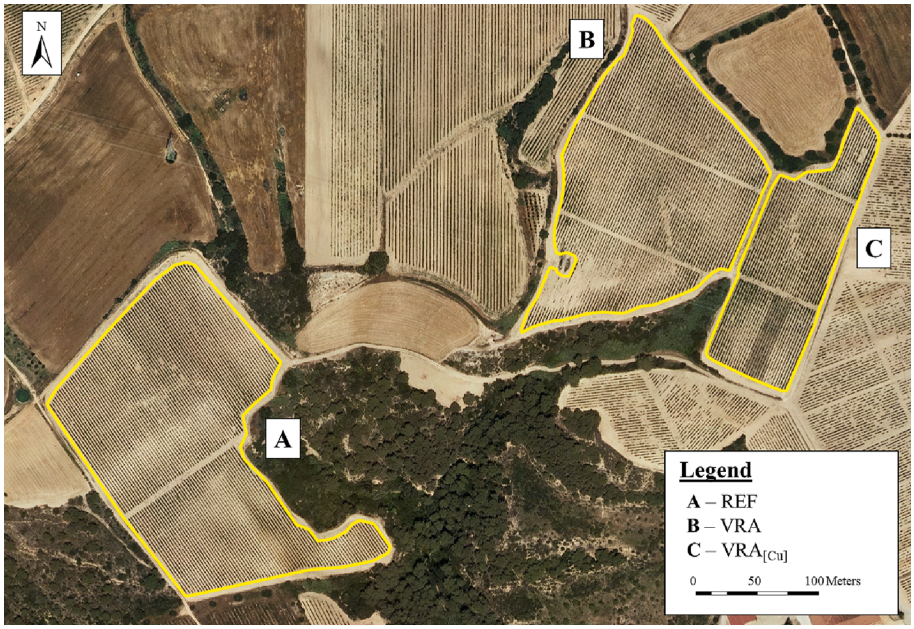

Fig. 1. Location of experimental plots. Spraying management based on reference practices (REF) were performed in plot A. Plot B and C were treated using VRA techniques, following the copper concentration listed in the product label in the case of plot C ($VRA_{[Cu]}$).

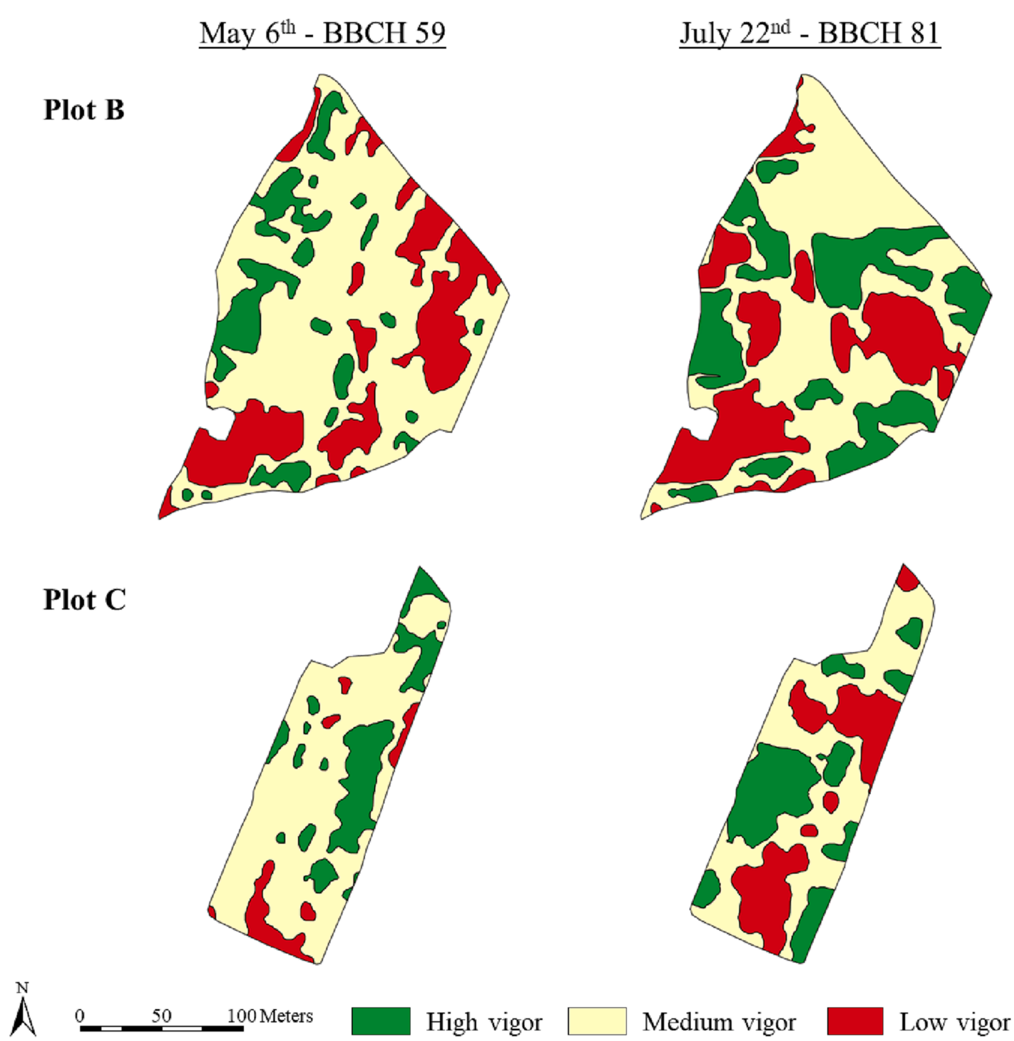

Fig. 2. Vigour maps obtained in the VRA parcels in the different BBCH studied.

was built and used to transform every pixel in the image to its corresponding reflectance value for each of the orthophotomaps. The mosaics were georeferenced using natural ground control points in the study area which were georeferenced with a global navigation satellite system with real-time kinematic (RTK) correction (model: GPS1200+, Leica Geosystems AG., Heerbrugg, Switzerland).

Assessment of canopy vigour and zoning of vegetation into homogeneous growth areas was done based on the normalized differential vegetation index (NDVI) (Rouse et al., 1973), which combines the spectral response of leaves in the red and NIR region of the spectrum. The NDVI has been widely used for canopy vigour assessment in different crops and environments, providing an easy, understandable, and robust way of depicting the spatial variability in the parcel (Gatti et al., 2017; Khaliq et al., 2019; Di Gennaro et al., 2019; Pádua et al., 2019; Matese et al., 2021).

One of the major advantages of using UAV for remote sensing operations is the high spatial resolution they provide that allows filtering out the noise and unwanted elements from the image. This is especially important in crops that are planted out in rows where the plot area is a combination of canopy vegetation, bare soil, weeds, shadows, and other objects. Prior to building the canopy vigour variability maps, the vine canopy was segmented from undesired noise by generating a vine-only binary mask. This was achieved by applying a threshold to the original NDVI image and setting the pixels above the set threshold as vineyard pixels (coded as "1" in the binary mask), whereas pixels below the set threshold were considered noise and set to "0". Once the NDVI threshold was applied, an inverse distance weighting interpolation (IDW) was performed to generate a continuous NDVI map which was classified into three different classes namely high, medium and low vigour. The classification was made based on a union of quintiles (P20, P40, P60, and P80) into three new classes where NDVI values lower than P20, between P20 and P80, and higher than P80 were categorized as low, medium, and high vigour, respectively (Fig. 2).

2.4. Prescription map generation

The most appropriate volume rate to be used in each growth stage, plot and vigour zone was selected based on the canopy characteristics. For this, a decision support system (DSS, DOSAVIÑA®) (Gil et al., 2011, Gil et al., 2019) was used in the field before each of the spray application. The system determines the most appropriate volume rate to be applied according to the canopy development which includes canopy height and width, categorical measure of leaf wall density, and sprayer design. Once the georeferenced canopy vigour maps were generated, the vigour zones were identified in the field by using a GNSS device and the specific areas were characterized by collecting 15 manual measurements of canopy height and width per vigour zone. Vine selection was made based on a random sampling methodology in each of the vigour zones determined in the different canopy maps used. Manual measurements were conducted using a regular measuring tape following the EPPO standard (EPPO, 2021). Each measurement included 95 % of the canopy, excluding protruding branches (Manktelow and Praat, 1997), and Leaf Wall Area – LWA (Morgan, 1981) was calculated being a recognized method related to canopy structure.

The measurements taken within a specific vigour zone were averaged and introduced in the DSS DOSAVIÑA® where the corresponding volume rate was calculated (Table 1 and 2). For the reference treatment (plot A), the volume rate as well as working conditions were set by the farmer managing the vineyard based on their previous experience. Nevertheless, although no variable rate was performed in plot A, the vegetation was also measured to have a sense of canopy development and characteristics which served for a better interpretation of the results found.

The products used were Codimur 50 and Beltasur 500. These products are wettable powder contact fungicides based on Copper Oxychloride 50 %. The recommended dose expressed in concentration for the Codimur 50 ranged from 0.3 to 0.4 % and expressed in a dose per hectare 1.8–3 kg ha^{-1} with a volume rate between 600 and 1000 L ha^{-1}, and for the Beltasur 500 0.3 % or 1.8–2.5 kg ha^{-1} with a volume rate between 600 and 830 L ha^{-1}. Selection of the dose of copper for the spraying application was done following different approaches for REF, VRA and $VRA_{[Cu]}$ treatments. In the REF treatment (Plot A) the dose of copper (kg ha^{-1}) was selected based on the experience provided by the technician in charge of the vineyard, but never overpassing the recommended dose per hectare listed on the product label. On the other hand, while VRA (Plot B) maintained the quantity of copper (kg ha^{-1}) used in REF treatment (Table 1), a reduction of copper was achieved in $VRA_{[Cu]}$ (Plot C) by following the recommended concentration (%) listed on the product label. The total quantity of copper to be applied was calculated based on the recommended volume rate determined by DOSAVIÑA® in every vigor zone.

2.5. Sprayer configuration

All spray applications were carried out with a conventional trailed axial flow air sprayer with air deflectors (Saher Maquinaria Agrícola S. L., Barcelona, Spain) with a 1000 L tank and an axial fan of 800 mm diameter. The sprayer was modified in order to enable a variable application based on prescription maps. In order to achieve this, the sprayer was equipped with a pressure sensor GEMS 1200 series (Gems Sensors & Controls, Plainville, CT, USA) to allow monitoring working pressure during the spraying work. It also included a control unit consisting on a WAATIC electronic controller (Estel Grup S.L., Barcelona, Spain) with an embedded GNSS receiver with a frequency of 1 Hz, a touchscreen and an automatic section controller. The system reads the coordinate position of the sprayer in the field, finds its location in the variable rate prescription map loaded in the controller and its corresponding volume rate. The section control unit would modify the working pressure using the electronic valve available in the sprayer to adjust the nozzle flow rate to achieve the desired volume rate.

Working parameters for each spraying application has to be initially set in the electronic controller touchscreen prior to any application (Tables 1 and 2). Number and type of nozzles were selected in each spray application based on the best combination of desired volume rate, forward speed and working pressure. It is important to notice that the farmer decided to use 8 nozzles in the spraying application performed in

Table 1
Specific working conditions for the spray applications conducted in growth stage BBCH 59. Forward speed was common for all treatments at 6.8 km h^{-1}. Copper used consisted on copper oxychloride 50 % W/W.

Treatment	Vigour Zone	LWA (m^2_{canopy} ha^{-1})	Volume (L ha^{-1})	Pressure (bar)	Nozzle nr. and type	Cu (kg ha^{-1}) (% reduction)	Working width (m)
REF	N.A.	5336	195	7.6	8 ATR orange	0.48	4.4
VRA	Low	4073	123	5.5	4 ATR yellow	0.48 (0 %)	2.2
	Medium	5009	151	8.4			
	High	5645	170	10.7			
$VRA_{[Cu]}$	Low	4409	133	6.5	4 ATR yellow	0.32 (33 %)	2.2
	Medium	5491	166	10.2			
	High	5818	176	11.5			

Table 2
Specific working conditions for the spray applications conducted in growth stage BBCH 81. Forward speed was common for all treatments at 6.8 km h^{-1}. Copper used consisted on copper oxychloride 50 % W/W.

Treatment	Vigour Zone	LWA (m^2_{canopy} ha^{-1})	Volume (L ha^{-1})	Pressure (bar)	Nozzle nr. and type	Cu (kg ha^{-1}) (% reduction)	Working width (m)
REF	N.A.	7300	208	11.1	4 ATR green	0.7	4.4
VRA	Low	5555	174	6.0	4 ATR orange	0.7 (0 %)	2.2
	Medium	6500	205	8.5			
	High	7700	243	12.0			
$VRA_{[Cu]}$	Low	6009	226	10.3	4 ATR orange	0.39 (44 %)	2.2
	Medium	6782	257	13.5			
	High	7755	292	17.6			

the first growth stage, whereas in the second growth stage reduced the number of nozzles to 4. This can be explained by the fact that the farmer wanted to generate the maximum possible number of droplets, which could ensure a high coverage of the canopy in BBCH59, provided the importance of ensuring an effective control of the disease in early stages of the vegetation. A proper calibration of the machine, in concordance with canopy needs, determined that 4 nozzles were sufficient to successfully cover the entire vegetation. Alternate row spraying was employed for the reference treatment following farm management practices, meaning a working width of 4.4 m. In contrast, all rows in the field were sprayed in the variable rate application treatments (VRA and $VRA_{[Cu]}$) with a working width of 2.2 m. Fan speed was maintained the same across the three different treatments.

2.6. Leaf deposit assessment

Leaf deposit was measured using real vine leaves as natural collectors using similar methodology described by Gil et al. (2007), Llorens et al. (2010) and Salcedo et al. (2020). The experimental design consisted on three sampling blocks per treatment, containing each sampling block three replicates (vines). In VRA and $VRA_{[Cu]}$ one sampling block was placed in each of the three vigour zones in the field. For sampling purposes, each vine was segmented into different quadrats along its canopy height and width (Fig. 3). The height of the plant was divided into 2 sampling areas (A, B) for the canopy growth stage BBCH 59 and 3 areas (A, B, C) for the BBCH 81 (Balsari et al., 2008; Campos et al., 2020). In both canopy growth stages, the width of the plant was divided into 3 areas (I, II, III) to assess the deposition in the two external sides of the vegetation row as well as in the internal leaves. The combination of the subdivisions generated resulted in 6 sampling areas for BBCH 59 and 9 sampling areas for BBCH 81. In each quadrat, a total of 5 random leaves were collected after each spraying application and samples were placed on tagged zip plastic bags. This sampling design originated a total of 54 and 81 samples per treatment, for BBCH 59 and BBCH 81 respectively,

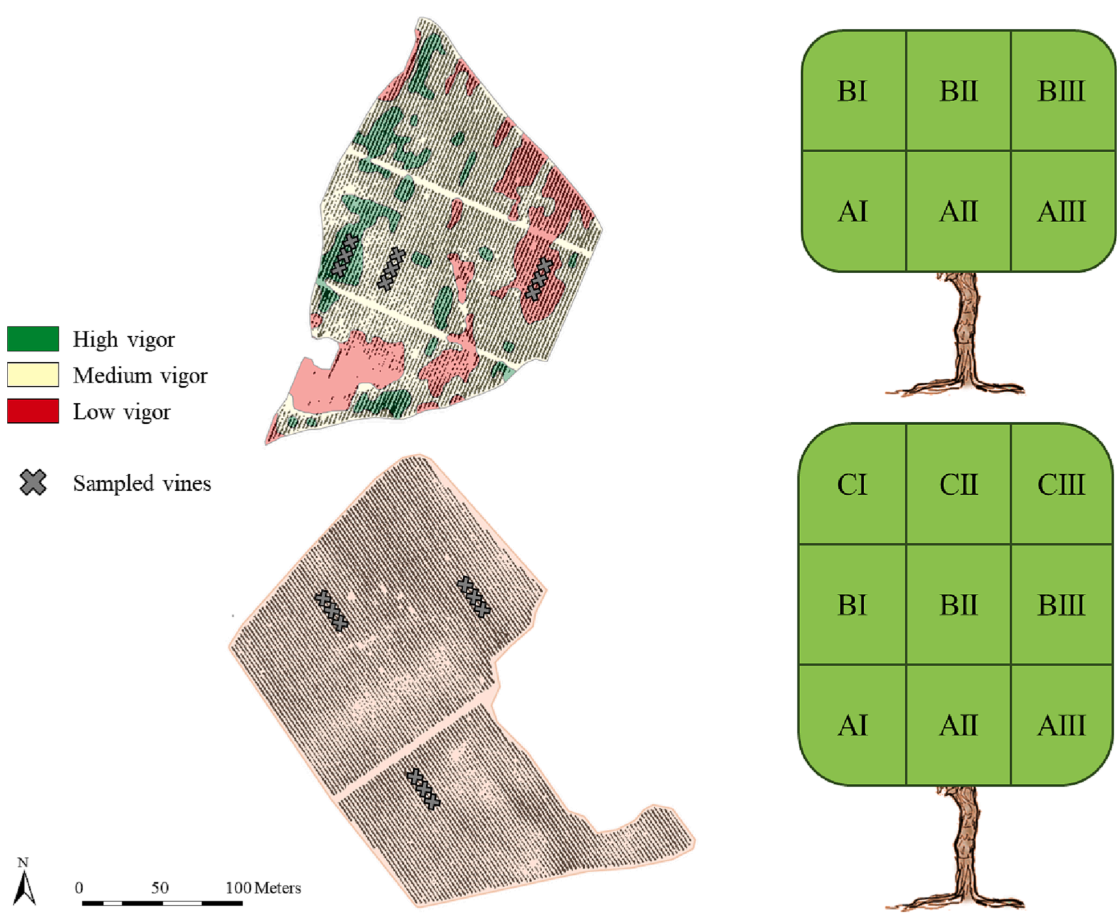

Fig. 3. (Left) Example of the location of the experimental blocs and the sampling vines in a VRA sprayed plot according to a vigour map (top) and the REF plot with constant rate (bottom). (Right) Representation of vine division in sampling areas for leaf sample collection for BBCH 59 (top) and BBCH 81 (bottom).

consisting on 5 leaves each.

Copper applied for crop protection was used as a tracer at a dedicated concentration for each spray application. Copper was selected because while being the product used to protect vineyard from fungal diseases it is easy to extract and measure in the laboratory. In addition, other authors used this product as a tracer for spray assessment (Whitney et al., 1989; Whitney and Salyani, 1991; Scapin et al., 2015; Behlau et al., 2020). Prior to the applications, 30 leaves samples were collected from every experimental block as a blank sample in order to determine the possible presence of copper before spraying. For the treatment done in the growth stage BBCH 59 only in the leaves collected in plot C ($VRC_{[Cu]}$) were found traces of copper, specifically 0.035 μg cm^{-2}. However, in the treatment carried out in the BBCH 81 were found copper on the leaves of all the plots. For the plot A (REF) were found 2.41 μg cm^{-2}, for plot B (VRC) 1.70 μg cm^{-2}, and for plot C ($VRC_{[Cu]}$) 1.68 μg cm^{-2}. These values were subtracted to the deposition obtained in each treatment.

The procedure used for copper extraction from sampled leaves consisted of adding a solution of Nitric Acid (HNO_3) 0.05 M to the plastic bags containing the leaves and mixing during 1 min. The volume of Nitric Acid solution added was 50 or 100 ml depending on copper deposition in leaves and detection range of the measurement equipment. After copper extraction by Nitric Acid, the solution was measured using an ICP-atomic emission spectrometer (ICP-AES).

Leaf area of each sample was determined in order to express deposition results in amount of tracer per unit leaf surface. For this purpose, for the two dates where field trials were conducted, a total of 100 leaves of different sizes were randomly collected from vines in the three parcels under study. Every leaf was weighted and its surface area (one side only) was measured using a LI 3100C (LI-COR, Lincoln, USA) electronic planimeter. A linear relationship (Eq. (1)) was built from the individual leaves in order to generate a weight ratio which was used to estimate the leaf surface area from the natural collectors used in the study (Cross et al., 2001; Gil et al., 2007; Gil et al., 2011).

$$LS(cm^2) = 47.51xLW(g)P < 0.05R^2 = 0.96 \quad (1)$$

where *LS* is the leaf surface and *LW* is leaf weight.

The amount of copper deposited per unit area of leaf was calculated following the methodology proposed by Gil et al. (2007) and Llorens et al. (2010) as presented in Eq. (2):

$$d = \frac{T_{cl} \times w}{S_a} \quad (2)$$

where d is the tracer deposited per unit sample surface (μg cm^{-2}), T_{cl} is the tracer concentration of the sample (mg L^{-1}), w is the amount of Nitric Acid solution used to extract the tracer from the sample (mL) and S_a is the area exposed of the sample (cm^2).

2.7. Data analysis

The effect introduced by the type of application, the different vigour levels within each plot as well as the homogeneity of PPP distribution in height and width along the canopy row were studied using an analysis of variance (ANOVA). Hence, to assess the deposition of copper in leaves and to determine the effect of the vigour zones, a one-way ANOVA was performed. Conversely, a two-way ANOVA was used in order to determine the effect of the type of treatment, the canopy position and a combination of both. Following ANOVA, a Tukey test for mean comparison was performed and SPSS 25.0 computing software (IBM corp., Armonk, NY, USA) was used for the analysis of data. The level of significance considered was $p < 0.05$ and previous to analysis, data was tested for normality and homoscedasticity compliance.

3. Results and discussion

3.1. Prescription map generation

The three vigour classes defined in each field from the multispectral images acquired during the drone flight campaigns, in growth stages BBCH 59 and BBCH 81, showed statistical differences in several of the parameters describing canopy characteristics that were manually measured in the field (Table 3). The well-established parameter for canopy characterization and dose expression LWA was consistently different between classes (no matter the growth stage and plot selected) justifying the separation into three classes proposed for these maps. Hence, high confidence could be given to the canopy maps produced by drone based imagery for representing the spatial variability at plot level. This is in concordance with previous research focused on using NDVI for yield prediction or canopy structural characterization in vineyard (Acebedo-Opazo et al., 2008; Martinez-Casasnovas et al., 2012; Bonilla et al., 2015; Darra et al., 2021; Matese and Di Gennaro, 2021; Campos et al., 2021).

Moreover, it was proven that comparison between treatments for a particular BBCH could be done with reliance since, in general terms, the LWA values did not show significant differences when comparing pairwise vigour classes (e.g. Low vigour for VRA and $VRA_{[Cu]}$).

It was noted that the structure of spatial variability changed throughout the vineyard season which disagreed with the research conducted by Kazmierski et al., 2011 that highlighted an intra-annual stability within vineyard season of NDVI patterns. However, this study used an airborne imagery with a resolution of 3 m pixel and only the last vineyard stages (85 days before harvest).

Prescription maps for plots B and C were created based on the values provided by the DSS DOSAVIÑA® for each canopy vigour class. A general increase in volume rate is observed between the two growth stages which is closely related with canopy vigour. On the contrary, in plot A, constant volume rate was used throughout the season averaging 200 L ha^{-1} regardless of the vegetation found in the field (Fig. 4).

3.2. Copper deposition on leaves

The results of copper deposition on leaves obtained in the field experiments have been organized in three separate sections. The first block describes the influence introduced by the type of spraying strategy (reference or variable) assessed by the differences in leaf deposition of copper. The second evaluates the influence of canopy vigor on the

Table 3
Canopy characteristics in each growth stage. Different letters represent statistical differences between vigour classes in the same treatment. * represents statistical differences between treatments in the same vigour class and BBCH. Statistical significance was assessed for α = 95 %.

Treatment	Vigour Zone	Canopy height (m)	Canopy width (m)	LWA (m^2 ha^{-1})
		BBCH59		
REF	N.A.	0.59	0.41	5336
VRA	Low	0.45 a	0.33 a*	4072 a
	Medium	0.55 b	0.39 b*	5009 b
	High	0.62 c	0.42 b*	5645 c
$VRA_{[Cu]}$	Low	0.48 a	0.37 a*	4409 a
	Medium	0.60 b	0.45 b*	5490 b
	High	0.64 b	0.48 b*	5818 b
		BBCH81		
REF	N.A.	0.80	0.39	7300
VRA	Low	0.61 a*	0.32 a*	5554 a*
	Medium	0.71 b	0.34 a*	6500 b
	High	0.85 c	0.36 a*	7700 c
$VRA_{[Cu]}$	Low	0.66 a*	0.37 a*	6009 a*
	Medium	0.75 b	0.42 b*	6781 b
	High	0.85 c	0.43 b*	7754 c

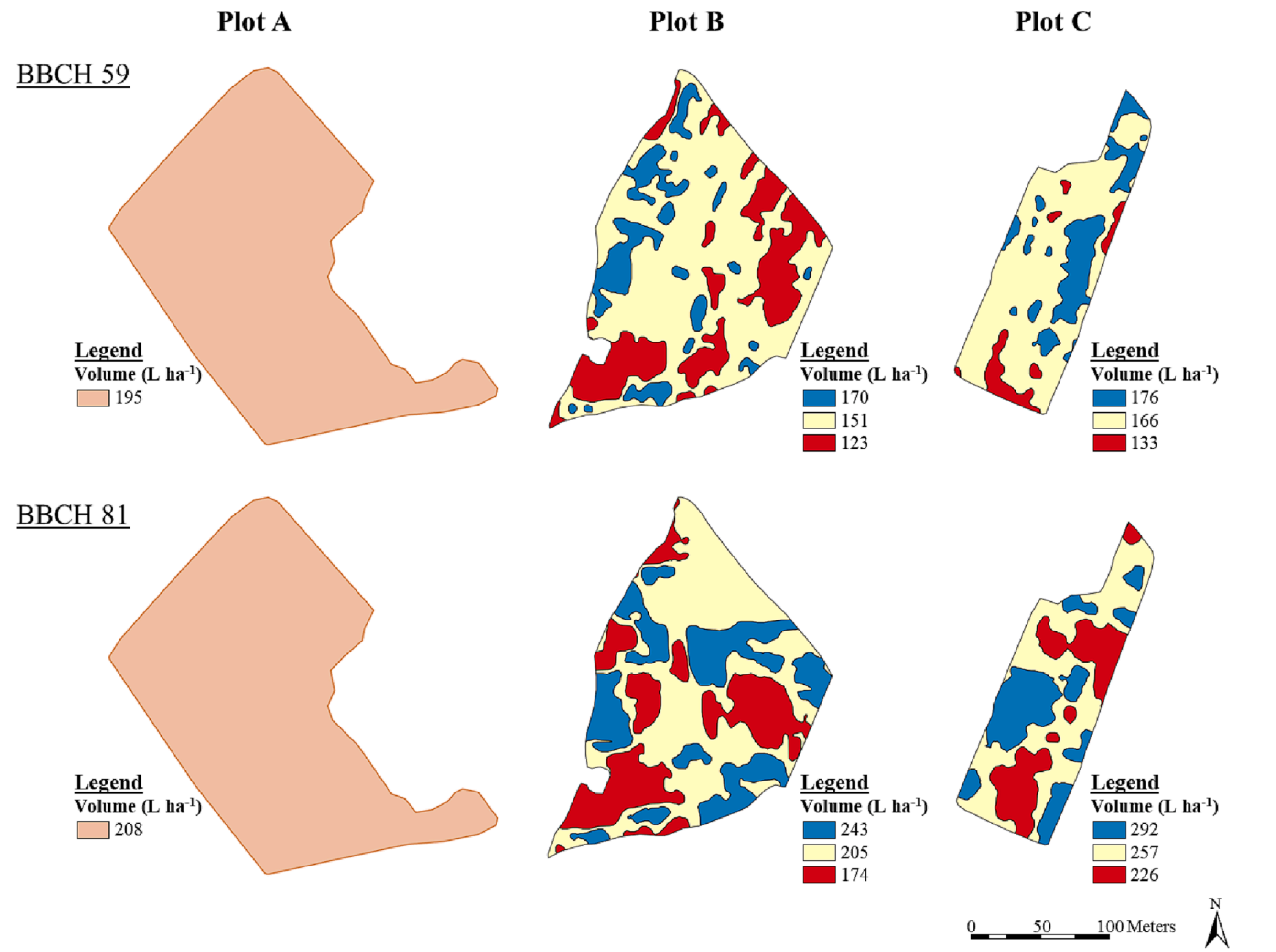

Fig. 4. Prescription maps used for each of the plots in the two growth stages where deposition assessment trials were conducted. Volume rate for plot A is set based on grower experience whereas plots B and C are based on per class canopy characterization using DOSAVIÑA® DSS.

achieved leaf deposit, and finally, the third block studies how homogeneous are the distribution of copper within the plant in each of the spraying application managements.

3.2.1. *Effect of application management strategy*

The quality of reference (REF), variable rate application (VRA) and variable rate application following the copper concentration listed in the

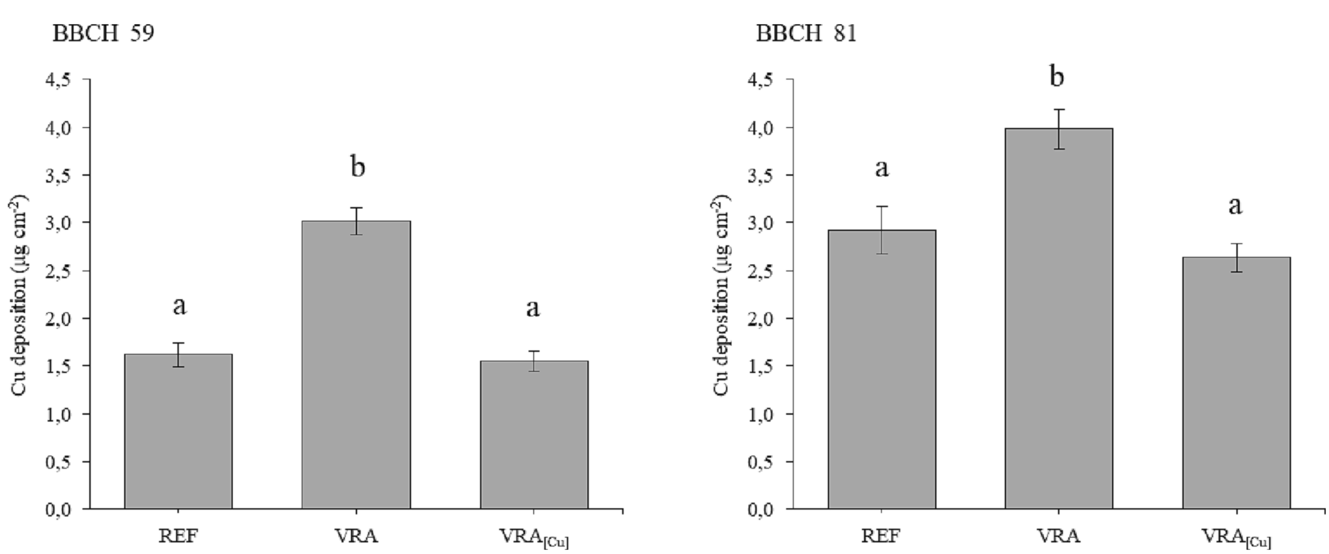

Fig. 5. Copper deposition in leaves (average ± SEM) according to the treatment, REF-Reference, VRA-Variable Rate Application and $VRA_{[Cu]}$-VRA following the copper concentration listed in the product label. Plots represented in growth stages BBCH 59 (Left) and BBCH 81 (Right). For each growth stage, different letters indicate significant differences (alpha greater than 95 %) between treatments.

product label ($VRA_{[Cu]}$) were compared and assessed based on the quantity of product recovered from sampled leaves. The two growth stages evaluated showed a greater deposition of copper in leaves when VRA management was used as compared with REF and $VRA_{[Cu]}$. This difference was consistent and statistically significant ($p < 0.05$) for BBCH 59 and BBCH 81 (Fig. 5). For the growth stage of beginning of flowering (BBCH 59), the amount of copper deposited in leaves in VRA treatment (3.02 μg cm^{-2}) was twice the amount found in the two other treatments (1.62 and 1.55 μg cm^{-2} for REF and $VRA_{[Cu]}$ respectively). Beginning of ripening (BBCH 81) was the second growth stage studied and also here significant differences among treatments were found with a higher amount of copper deposited in leaves when VRA management was used (3.98 μg cm^{-2} as compared with 2.92 and 2.64 μg cm^{-2} for REF and $VRA_{[Cu]}$ respectively).

It is important to consider the effect of proper circulation in the vineyard according to the characteristics of the spraying machine, which greatly impacted the differences in deposition between VRA and REF. Although REF was applied every second row, sprayer configuration was set aiming to obtain similar deposition in leaf as that obtained by VRA treatment (the two spraying applications worked with the same amount of copper applied per hectare). The difference in copper deposition between the two treatments, which aimed to apply the same copper per hectare, is indeed product that did not impact the leaf (46 % in the first growth stage and 28 % in the denser canopy stage) and therefore represents potential losses and contamination to the soil and air. Results also demonstrated that higher amount of copper reaches the leaf when canopy characteristics are taken into account for volume rate calculation and spraying is performed based on different vigour zones. On the other hand, reduction of 33 and 44 % of copper used per hectare in $VRA_{[Cu]}$ compared with REF did not have impact on the product deposition in leaves, achieving the same copper deposit than the application strategy used by the farmer (REF). This reduction did not have impact in the efficacy for controlling the disease during the season, and no presence of Downy Mildew was found in any of the plots used in this study. This could indicate that the deposition achieved by $VRA_{[Cu]}$ (similar to that achieved by REF) is enough for controlling Downy Mildew under the specific situation of this vineyard and conditions for the season, and there is no apparent reason to try to achieve the higher deposition observed in VRA treatment.

3.2.2. Effect of canopy vigour zone in Cu deposition

The amount of copper deposited on leaves in the different canopy vigour zones were compared between VRA and $VRA_{[Cu]}$ treatments. Leaf deposition values for VRA treatment were always higher than those found for $VRA_{[Cu]}$ due to the highest amount of product applied in VRA. The importance here lays in the fact that within the same treatment, the three vigour zones received similar copper deposit in leaves, regardless of the volume rate applied (Fig. 6). This was already expected, since canopy characterization aims to adjust applied volume to achieve even deposition in leaves regardless of the leaf density. In general, it was not found statistical differences between vigour zones within the same treatment. The only exception was $VRA_{[Cu]}$ in growth stage BBCH 59 where significantly higher amount of copper deposited in leaves was observed in the area of high vigour as compared with the rest of the vigour zones. Canopy dimensions where minimal in BBCH 59 due to the short shoot length and sparse canopy density, which could have impacted the results in this initial growth stage of vines. Moreover, even when using drones and high-resolution imagery, it becomes difficult the canopy characterization of small and sparse canopies happening in the beginning of the growth stage.

The results obtained indicate that although volume rates and spraying characteristics (e.g. working pressure) were modified in real time from zone to zone based on the prescription maps, similar amount of active matter reached the leaves of the whole vineyard. Furthermore, it is well known that by adjusting volume rate to canopy characteristics and performing spray applications following best management practices, coverage of plant protection products is improved. Canopy spray distribution was assessed by Campos et al. (2020) and no significant differences were found in coverage percentage across vigour zones. Similar results were obtained in previous researches dealing with real time modification of the working parameters based on the canopy characteristics as perceived by on-board ground sensors (Solanelles et al., 2006; Gil et al., 2007; Balsari et al., 2008).

3.2.3. Uniformity of spray distribution over the target

The impact of spray application strategy on the homogeneity of copper deposition in the different sections of the vine was assessed. A two-way ANOVA was used in order to determine the interaction between the factors of application strategy and leaf position within the plant. The results for the spray applications conducted during the growth stage of beginning of flowering (BBCH 59) showed a significant effect for application strategy ($p < 0.001$) but not for position in the vine nor the interaction between the two factors ($p = 0.17$ and $p = 0.21$ in each case). As already mentioned in section 3.2.1, significant differences were found between VRA and the rest of treatments (REF and $VRA_{[Cu]}$). It is relevant to mention the visible effect of alternate row spraying in REF treatment where the copper deposition resulted especially reduced in the vine layers opposite from the sprayer trajectory. Positions A-I and

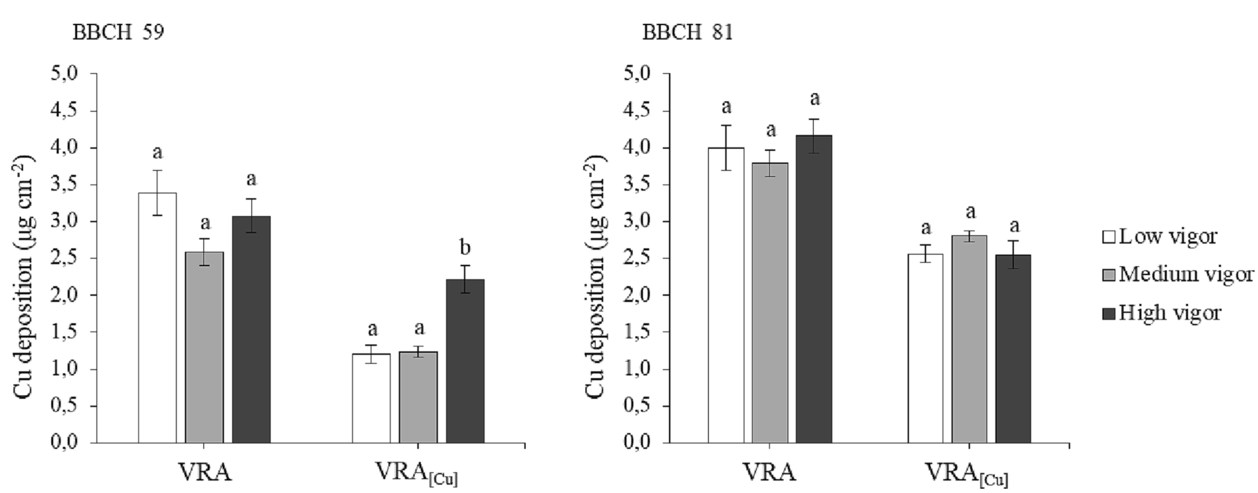

Fig. 6. Copper deposition in leaf (average ± SEM) according to the treatment, VRA-Variable Rate Application and $VRA_{[Cu]}$-VRA following the copper concentration listed in the product label, and vigour zone (low, medium, high). Plots represented in two growth stages BBCH 59 (Left) and BBCH 81 (Right). For each growth stage and treatment, different letters indicate significant differences ($p < 0.05$) between vigour zones.

B-I (Fig. 7) received the lowest amount of copper although differences as compared to other positions in the canopy were not significant statistically speaking.

On the contrary, in the fully developed growth stage with denser and taller canopy (i.e. BBCH 81), statistical significance was found for application strategy, position of leaves in the vine and the interaction between the two factors. Positions A-I, B-I and C-I in REF treatment (in BBCH 81) received a lower amount of copper because the droplets had to pass through the dense vegetation to reach the leaves placed in these positions. This effect is not produced in VRA and $VRA_{[Cu]}$ treatments where the two sides of the vegetation wall were sprayed. This work intended to compare parameters linked to deposition of copper within the canopy by three application strategies, and it was accepted the fact that the reference strategy was carried out in an inefficient way from the point of view of quality of the application, but following the normal practice used by growers in the zone. This is well known and extensively published, and the improvements of conducting a spray application covering both sides of the canopy have been widely proven in several crops (Derksen et al., 2004; Landers, 2010; Verpont et al., 2019). Nonetheless, farmers in dry areas and hot summers continue using the strategy of alternate row spraying, especially for downy mildew, balancing the probability of infection, the control efficacy in dry seasons and the time spent for every application. This implies working with nozzles with high flow rate as well as increasing the volume of air to ensure a large reach of the droplets, which increases losses of product by drift and runoff (Garcerá et al., 2017).

The effect of alternate row application is well seen when inspecting the coefficient of variation to assess the variability of product deposition in each of the application strategies. As observed in Table 4, REF treatment shows a consistent higher variation of the product deposition in leaf (44.7 and 78.1 %), especially in dense canopies where droplets have physical impediments to penetrate the vegetation wall (BBCH 81). On the contrary, both VRA strategies provide an acceptable coefficient of variation below 35 % in the first growth stage showing the effect of treating both sides of the vegetation plus adapting the volume rate to the canopy characteristics and the spatial variability of the vineyard. In the second growth stage the CV reaches 46.2 and 47.2 % for VRA and $VRA_{[Cu]}$ respectively representing approximately a reduction of 40 % as compared of that obtained by the REF treatment.

3.3. Potential copper reduction

A total of four spraying applications with copper formulates were made during the entire 2019 season in the vineyards object of this study (Table 5). VRA and REF treatments used a similar amount of copper during the season, although VRA application provided a higher deposition in the leaf (Fig. 5) and improved the homogeneity of the application in the canopy (Table 4). On the other hand, important savings of 1.02 kg Cu ha^{-1} were achieved by applying the strategy of $VRA_{[Cu]}$, representing a 40 % less copper used during the entire season as compared with the REF strategy. Most important, despite this reduction in copper use, no presence of disease was found in any of the vineyards used for this study, indicating that 1.02 kg Cu ha^{-1} could be saved with no impact on grape yield caused by Downy Mildew. Consequently, it was demonstrated that combining VRA technology, a careful calibration of the spraying machine, and a proper circulation in the vineyard according the sprayer characteristics, can provide relevant copper savings without affecting the quality of the application. Considering the tendency of the EU to progressively reduce the use of copper in viticulture (EU 2018/1981 authorizing a maximum use of 28 kg Cu ha^{-1} in metal form over a period of 7 years, resulting in an average of 4 kg Cu ha^{-1}),

Table 4
Coefficient of variation (%) average for the copper deposition in the plant according to the application strategy followed (Reference, VRA, and VRA following the copper concentration listed in the product label) for growth stages of beginning of flowering (BBCH 51) and beginning of ripening (BBCH 81).

Application Strategy	CV (%)	
	BBCH 51	BBCH 81
REF	44.7	78.1
VRA	24.2	46.2
$VRA_{[Cu]}$	32.7	47.2

Table 5
Copper application program for the season 2019 in the commercial vineyards under study (Reference, VRA, and VRA following the copper concentration listed in the product label); Doses applied for every date and total Cu in metal form applied the entire season. Cu reduction is calculated compared to REF.

Application date	Cu in metal form (kg ha^{-1})		
	REF	VRA	$VRA_{[Cu]}$
21/05/2019	0.48	0.48	0.32
29/05/2019	0.55	0.55	0.32
13/06/2019	0.77	0.77	0.46
09/08/2019	0.70	0.70	0.39
Total Cu used in 2019	2.51	2.51	1.49
Cu reduction vs. REF	0.00	0.00	1.02

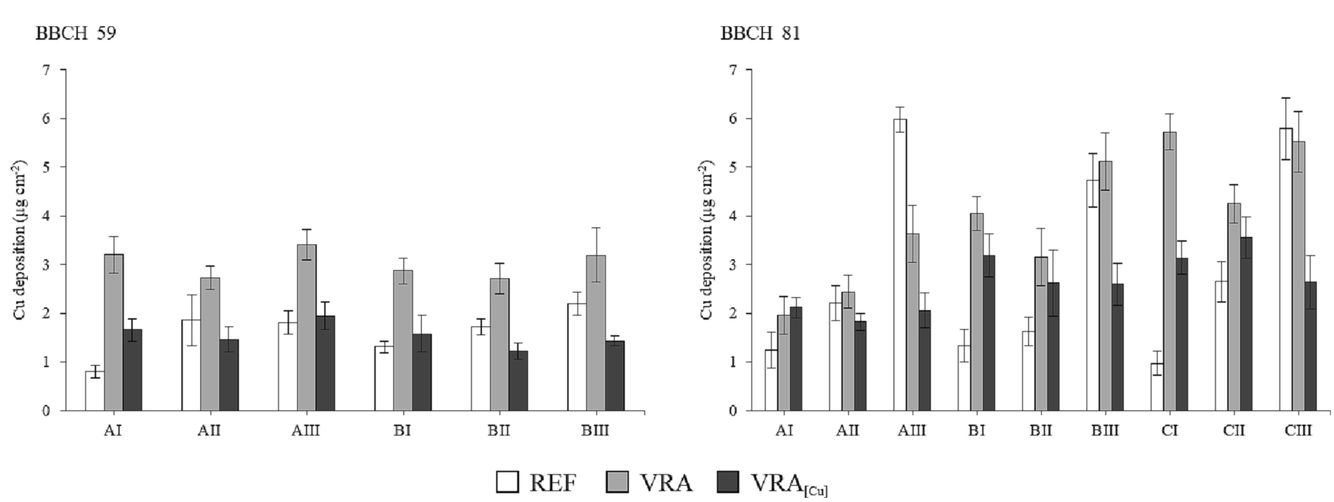

Fig. 7. Copper deposition in leaf (average ± SEM) according to the treatment, REF-Reference, VRA-Variable Rate Application and $VRA_{[Cu]}$-VRA following the copper concentration listed in the product label, for each position in the vine (A, B, C stand for lower, middle and higher positions and I, II, III for internal, middle or external side of the vegetation wall).

saving copper becomes crucial for ensuring the sustainability of the vineyard and disease control management.

Although incipient research is available regarding map based variable rate application of pesticides in vineyards, different authors have presented innovative works achieving a reduction of 47 % (Campos et al., 2019) and 25 % (Román et al., 2020) in volume rate used as compared to reference applications, being maintained the PPP concentration provided the same percentage of reduction of active matter used. It is worth noticing that in the referred works, reference volume rates used were 325 and 450 L ha^{-1} respectively (common for the region), while the highly innovative farmer selected for this research applied a maximum of 208 L ha^{-1} which restricts the margin of savings. Similarly, a reduction of 40 % of water and product has been reported by using on the go ground sensors for canopy characterization (Gil et al., 2007; Llorens et al., 2010). Similar methodology based on TRV was used to apply fertilizers in vineyards in a site specific scheme following canopy characteristics sensed by ground sensors, achieving a reduction of 80 % of the dosage (Andújar et al., 2019).

4. Conclusions

This study focuses on the reduction of copper and improvement of spraying application results by using variable rate application technology in commercial vineyards. Here we show for the first time that combination of drone and VRA technology, a rational dose expression protocol and best management practices when calibrating the sprayer, can have an important impact in the reduction of copper used in viticulture. All this, while maintaining similar results obtained by current practices carried out by the farmer, in terms of copper deposit in leaves and disease control. Furthermore, with an expected reduction of drift and runoff losses, minimizing the risk of soil and water contamination, although this needs to be confirmed in further experiments.

Drone based canopy maps where used to determine vineyard spatial variability and the canopy in every vigor zone was characterized and later used to generate variable rate prescription maps with optimal volume rate selection using the DSS DOSAVIÑA®. It was ratified that map based VRA provides a tool for better adjusting the volume rate in every area of the field based on canopy characteristics, which in the end turns out to be a vector for more homogeneous deposition in inner and outer leaves. In addition, drone based remote sensing is a reliable tool for canopy map generation although different platforms should be tested to ensure cost-effective and ready to use maps in timely manner when the farmer requires it.

A reduction of 33 % and 44 % of copper used per hectare (as compared with REF treatment) in the growth stages of beginning of flowering and veraison, respectively, does not have any negative effect on the amount of product deposited in the leaf. This fact allows reducing the active matter used while complying with concentration recommendations found in the label of the product. Findings on pesticide reduction must be always backed up with technical decisions on the best moment for pest or disease management and principle of action of the pesticide used, to ensure that enough product is available in the leaf to control the specific pest or disease.

This work also demonstrated that it is questionable the dose expression based on product per hectare, and further work is required to express the amount of pesticide to used based on canopy characteristics (L m^2 LWA). Furthermore, the benefit of VRA technology must be associated to product dose recommendation based in unit of canopy.

Results presented in this study will help promoting and adopting the use of variable rate technology and best management practices to reduce the use of PPP in viticulture in line with EU directives and the objectives of 50 % reduction of PPP and fertilizers set by the Farm to Fork strategy. Further work is needed to corroborate that the proposed working protocol is capable of controlling the diseases under favorable climate for Downy Mildew development other than the Mediterranean area.

CRediT authorship contribution statement

Francisco Garcia-Ruiz: Conceptualization, Methodology, Investigation, Writing – original draft. **Javier Campos:** Conceptualization, Methodology, Investigation, Data curation, Resources, Writing – review & editing. **Jordi Llop-Casamada:** Methodology, Investigation, Writing – review & editing. **Emilio Gil:** Conceptualization, Methodology, Resources, Writing – review & editing, Supervision, Project administration, Funding acquisition.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Data availability

Data will be made available on request.

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