NEW DISEASE-RESISTANT GRAPEVINE VARIETIES RESPONSE TO DROUGHT UNDER A SEMI-ARID CLIMATE

Authors: Luciana WILHELM DE ALMEIDA[1,2*], Anne PELLEGRINO[2], Bénédicte FONTEZ[3], Laurent TORREGROSA[1, 2], Hernán OJEDA[1]

[1]Unité Expérimentale de Pech Rouge (UE 0999), INRAE, 11430 Gruissan, France

[2]UMR LEPSE, Univ Montpellier, INRAE, CIRAD, Institut Agro Montpellier, 2, place P. Viala, 34060 Montpellier Cedex, France

[3]UMR MISTEA, Univ Montpellier, INRAE, Institut Agro, Montpellier, 2, place P. Viala, 34060 Montpellier Cedex, France

*Corresponding author: luciana.wilhelm-de-almeida@inrae.fr

Keywords: water deficit, acclimation, water use efficiency, yield, quantitative analysis

Introduction

In many regions, climate change leads to an increase in air temperature combined with a reduction of rainfall, intensifying climatic demand and water deficits (WD) (Cardell et al. 2019), which in turn may negatively impact grapevine development, yield and grape composition (Santos et al. 2020). In addition, climate change may also increase disease pressure, leading to further yield and quality losses, besides increasing costs due to increased vineyard spraying (Santos et al. 2020) and reducing viticulture acceptability by consumers (Guichard et al. 2017). Adopting new resistant varieties appears as a promising long-term solution to better manage vine protection, but unfortunately little is known regarding their behavior in front of WD. Indeed, most grapevine breeding programs do not consider drought performance as a primary attribute for selection, which is related to the difficulty to identify relevant phenotypes building a drought-performant behavior of a perennial fruit crop in the short and long terms.

Grapevine's first acclimation to WD is the reduction of transpiring surfaces, reducing leaf area and canopy development (Simonneau et al. 2017) by primarily reducing secondary growth with later effects on main shoot (Pellegrino et al. 2005). Reproductive organs are also importantly affected by WD, normally leading to reductions in number of clusters, number of berries and berry weight, with greater impact if WD occurs during pre-véraison stage (Levin et al. 2020). In addition to these seasonal processes, the interseasonal effects include reduced reserve repletion and bud-fruitfulness (Guilpart et al. 2014), impairing vineyard longevity, a factor of great importance to be considered for a perennial fruit crop.

Despite many studies addressing WD effects on grapevine development and on grape yield and quality, there is a lack of knowledge regarding the plant processes involved in the acclimation from one season to the next, and how the new fungi-resistant genotypes behave in front of abiotic constraints, such as drought. Understanding these points is essential for anticipating vineyard durability and genotype selection in a changing climate.

Research Objectives

The objectives of the present work were to quantitatively evaluate the effects of WD over four consecutive years on gas exchanges, vegetative development and yield components, in a set of six new resistant-varieties issued from the INRAE breeding program. An original methodological approach was proposed, to reduce experimental noise linked to open field experiments, by downscaling the phenotyping unit to the individual vine. This allowed a fine traceability of the pluriannual effect of water deficit at plant level. Furthermore, by reasoning harvest date at the arrest of phloem unloading into berries (a precise physiological development point) it was possible to estimate the maximum fresh yield and to compare the concentration of solutes at a similar fruit physiological stage avoiding errors linked to berry water loss.

Material and methods

Experimental site and plant material
The experiment was conducted with field-grown vines (2019-2022) at the INRAE unit of Pech Rouge, Gruissan, France. The panel of genotypes consisted of 6 INRAE resistant varieties (*V. vinifera* L. x *M. rotundifolia* hybrids) and the *V. vinifera var.* Syrah, as the control. The resistant varieties included two certified varieties: Artaban and Floréal and 4 varieties in the final process of certification: 3176N, 3159B, G14 and G5, with the two last ones carrying the sugarless trait (Bigard et al. 2022). A total of 30 plants per genotype were individually phenotyped. From those, half were irrigated from flowering until harvest, receiving 40 L per plant per week, while the other half was not irrigated. All genotypes were situated in close-by plots with the same planting density (2.5m x 0.9 m), orientation of rows (SW-NE) and managed with the same pruning method.

Plant water status
The predawn water potential (**Ψ**p) was measured weekly for all 30 plants per genotype, from flowering until harvest, between 3:00h and 5:00h using a Scholander chamber. The accumulated **Ψ**p (Acc-**Ψ**p) was then estimated for each plant and each year as the area under the curve of **Ψ**p over time, divided by the number of days of the period comprised from flowering to harvest.

Physiological parameters
Leaf net photosynthesis (A), stomatal conductance (gs) and transpiration (E) were assessed on the preceding and following day of **Ψ**p measurement using an infrared gas analyzer system (ADC BioScientific LC pro System), equipped with a 6.25 cm² chamber. Measurements were performed between 13:00h and 16:00h, at saturated light (≥ 1000 µmol

m-2 s-1) and at CO2 concentration of 390 ppm. The WUEinst and WUEi were calculated as the ratio of A to E and to gs, respectively.

Harvest date

Harvest was defined as the moment of phloem unloading arrest, the stage at which berries reach maximum water and solute contents, corresponding to the physiological ripe stage (Vmax) (Bigard et al. 2022). During 2019 and 2020 seasons, Vmax was estimated at the cluster level, by following weekly the volume increase of 3 clusters per genotype as described by Torregrosa et al. (2008). In the 2021 season Vmax was estimated from image analysis by counting the number of pixels per cluster over time using ImageJ software (Lopes & Cadima 2021). For this purpose, 1 cluster per plant on 6 plants per genotype was photographed each week. Plants were chosen to cover a range of **Ψ**p. Bunches were harvested when their volume or number of pixels stopped increasing. For the three years, sugar content at harvest reached *ca* 23° Brix for Floréal, Artaban, 3159B, 3176N and Syrah, and 20º Brix in G5 and G14. In the 2022 season, harvest was thus targeted from those two thresholds.

Vegetative and reproductive variables

The pruning weight and number of shoots were assessed for each vine after the cropping season, during winter. The shoot weight was calculated dividing pruning weight by the number of shoots. At harvest, yield and number of clusters were directly determined. To assess individual berry weight, a triplicate of 100 berries per plant was sampled and weighed. The average cluster weight and number of berries per cluster were then calculated from the variables described above.

Statistical analysis

All graphical processing and statistical tests were performed using R studio software.

Vegetative and reproductive variables were fitted with linear regressions with Acc-**Ψ**p as continuous variable, Genotype and Year as categorical variables. The effects of Genotype, Year and their interaction on the intercepts (‘Genotype’, ‘Year’, ‘Genotype:Year’) and of Genotype on the slopes (‘Acc-**Ψ**p:Genotype’) were tested.

Physiological variables (gs, A and E) responses to **Ψ**p were linearized using log transformation. The effects of Genotype on the intercept (‘Genotype’) and on the slope (‘**Ψ**p:Genotype’) were also tested.

All regressions were performed considering each plant as one biological replicate and all non-significant factors were later dropped. The proportion of variance explained by each factor was estimated dividing its sum of squares by the total sum of squares (η^2). Multiple comparison of slopes and intercepts were performed with the emmeans package.

In addition to the regression models, a multivariate analysis (PCA) was performed to decipher the genotypic and pluriannual responses to WD (slopes, intercepts) on a subset of plant variables.

Results

Plant water status across years

The Acc-**Ψ**p in all genotypes and years decreased during the season, at a higher extent in non-irrigated vines when compared to irrigated vines. In general, the higher Acc-**Ψ**p values were seen in 2019, 2020 and 2022 seasons (-0.36, -0.33 and -0.42 MPa, respectively), while 2021 had the lowest values (-0.67 MPa) for all genotypes. Correspondingly, the factor Year explained 43% in the variation of Acc-**Ψ**p while Genotype explained 24%.

Overall Genotype and Year effects on plants responses to water deficit

The variations of **Ψ**p and their interactions with the Genotype and Year were able to explain from 50% to 86% in variations found for vegetative and reproductive variables (Figure 1). The lowest η^2 values were reported in physiological variables, reaching 17% and 24% for WUEi and WUEinst respectively.

Variations of water status (**Ψ**p or Acc-**Ψ**p), explained up to 55% of variations in berry weight, 34% in shoot weight and 36 to 48% of gas exchanges (gs, A and E). Yet, the variations of the slopes due to the Genotype (**Ψ**p:Genotype) explained a maximum of 5% in WUEi.

In contrast, the Genotype highly impacted the maximum values of the variables (intercept, 'Genotype') accounting for 50% and 45% of the variations in Nb of shoots and pruning weight, respectively. Year and the interaction Genotype:Year showed lower contributions in intercept variations, with the maximum being reported in Nb of clusters (η^2 of Year = 14%) and in Yield (η^2 of Genotype:Year = 17%) (Figure 1).

Variations of plants responses to water deficit between the Genotypes over successive Years

Principal components analysis with the slopes and intercepts, extracted from the linear fitted models are shown in Figure 2. The PCA with genotype sensitivity to water deficit (slopes) explained 56% of the variation, where the first and second principal components (PC1 and PC2) accounted for 32.7% and 23.3%. PC1 was mainly related to the slopes of photosynthesis (S_A) and berry weight (S_BW), and PC2 was related to the slopes of Yield (S_Yield) and of Ravaz index (S_RI). Individual shoot weight (S_SW), pruning weight (S_PW) and Water Use Efficiency (S_WUEi) were not correlated to those variables. Genotypes displayed different acclimations to water deficit. The genotype G5 showed a higher sensitivity of berry weight and photosynthesis to water deficit. Floréal and G14 (oppositely from Artaban) showed greater regulations in yield and Ravaz index. Floréal was also opposite to WUEi, thus indicating a lower sensitivity to water deficit of this variable. Finally, 3159B and 3176N were characterized by a greater sensitivity to water deficit of vegetative development (pruning weight and shoot weight), and lower sensitivity to water deficit of WUEi for 3176N. The cv. Syrah was poorly represented on this PCA.

The PCA with genotype and year effects on the maximum values of plant variables, i.e., under unlimited water supply (intercepts) explained 57% of the variation, where PC1 and PC2 accounted for 24.3% and 32.9%, respectively. Photosynthesis (I_A) was opposite to the pruning weight (Ii_PW) on PC1, while Yield (Ii_Yield) mostly contributed to PC2. Genotypes were spread on the two axes. Syrah and 3159B were opposite to G5 and G14 on PC1, being characterized respectively by higher vegetative biomass and photosynthetic capacity. The genotype 3176N was opposite to Artaban on PC2, indicating higher yield and lower WUEi potential. Floréal was poorly represented by this PCA (central position). Years were mainly related to PC2, separating 2020 (correlated to higher Yield potential) from 2019 and 2021 seasons (correlated to lower Yield potential). Indeed, 2019 and 2021 seasons were characterized as dryer seasons than 2020 and 2022.

Conclusion

Through a 4-year study, 7 varieties with different genotypic backgrounds were individually phenotyped regarding their physiological, vegetative and reproductive response to drought. This original methodology allowed us to implement quantitative analysis between several wide-measured indicators and Ψp. To such a degree we could quantify the genotype effect on the potential behavior under non-limiting water supply (intercept) and to a lesser extent on the sensitivity to water deficit (slope) and later rank and correlate them. In the full paper we will provide further details on the different variables and address the hierarchy of plant vegetative, reproductive and gas exchanges responses to water deficit among the genotype and over the years.

Acknowledgements

The program was financially supported (per order of contribution) by Occitanie government, the CIVL (Comité Interprofessionnel des Vins du Languedoc), INRAE and Institut Agro Montpellier. The authors thank Emmanuelle García-Adradosfor her essential technical support and management of the experimental fields and the students Emilie Barnett, Martina Baliani and Bruno Izquierdo for their contribution in data acquisition.

Literature Cited

Bigard, A., Romieu, C., Ojeda, H. and Torregrosa, L.J.-M., (2022) The sugarless grape trait characterised by single berry phenotyping. OENO One **56**, 89–102.

Cardell, M.F., Amengual, A. and Romero, R., (2019) Future effects of climate change on the suitability of wine grape production across Europe. Reg Environ Change **19**, 2299–2310.

Guichard, L., Dedieu, F., Jeuffroy, M.-H., Meynard, J.-M., Reau, R. and Savini, I., (2017) Le plan Ecophyto de réduction d'usage des pesticides en France : décryptage d'un échec et raisons d'espérer. Cah. Agric. **26**, 14002.

Guilpart, N., Metay, A. and Gary, C., (2014) Grapevine bud fertility and number of berries per bunch are determined by water and nitrogen stress around flowering in the previous year. European Journal of Agronomy **54**, 9–20.

Levin, A.D., Matthews, M.A. and Williams, L.E., (2020) Effect of Preveraison Water Deficits on the Yield Components of 15 Winegrape Cultivars. Am J Enol Vitic. **71**, 208–221.

Lopes, C. and Cadima, J., (2021) Grapevine bunch weight estimation using image-based features: comparing the predictive performance of number of visible berries and bunch area. OENO One **55**, 209–226.

Pellegrino, A., Lebon, E., Simonneau, T. and Wery, J., (2005) Towards a simple indicator of water stress in grapevine (Vitis vinifera L.) based on the differential sensitivities of vegetative growth components. Australian Journal of Grape and Wine Research **11**, 306–315.

Santos, J.A., Fraga, H., Malheiro, A.C., Moutinho-Pereira, J., Dinis, L.-T., Correia, C., Moriondo, M., Leolini, L., Dibari, C., Costafreda-Aumedes, S., Kartschall, T., Menz, C., Molitor, D., Junk, J., Beyer, M. and Schultz, H.R., (2020) A Review of the

Potential Climate Change Impacts and Adaptation Options for European Viticulture. Applied Sciences **10**, 3092.

Simonneau, T., Lebon, E., Coupel-Ledru, A., Marguerit, E., Rossdeutsch, L. and Ollat, N., (2017) Adapting plant material to face water stress in vineyards: which physiological targets for an optimal control of plant water status? OENO One **51**, 167.

Torregrosa, L., Pradal, M., Souquet, J.-M., Rambert, M., Gunata, Z. and Tesniere, C., (2008) Manipulation of VvAdh to investigate its function in grape berry development. Plant Science **174**, 149–155.

Tables and Figures

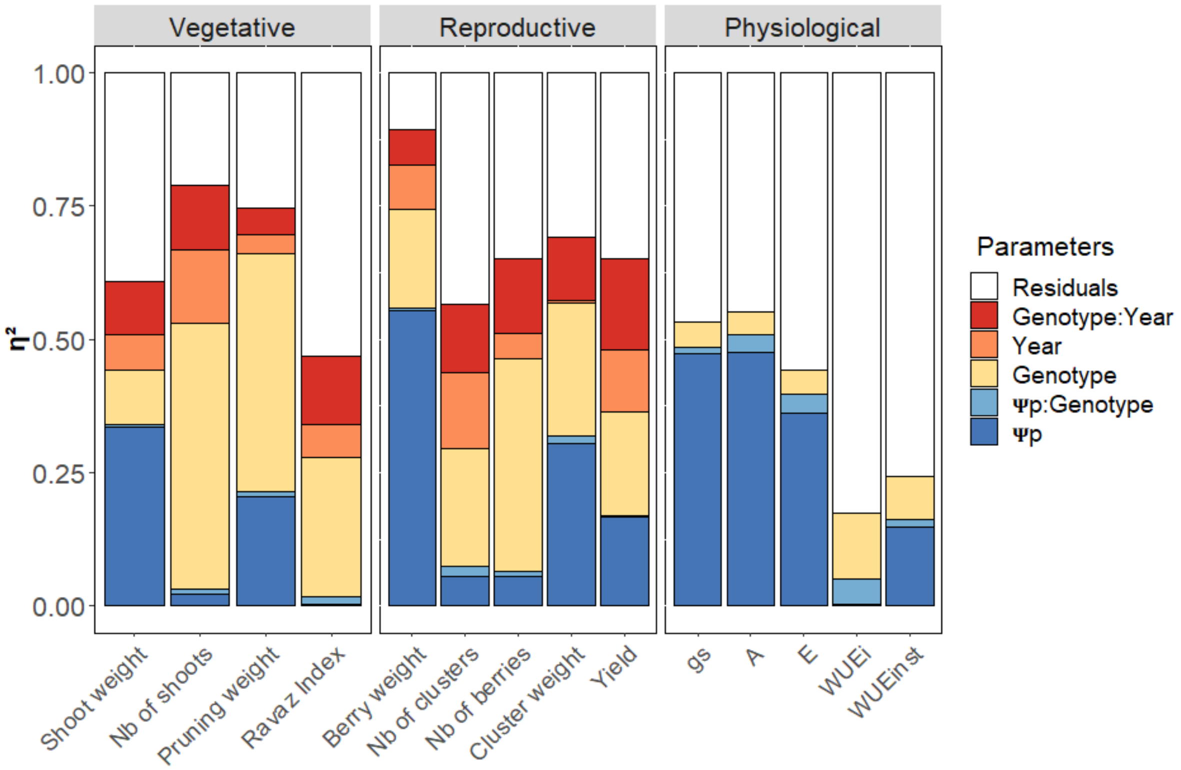

Figure 1. Proportion of variance explained (η²) by each factor and its interactions (p.value < 0.05), on slopes (blue colors) and intercepts (orange colors), in each vegetative, reproductive and physiological variable.

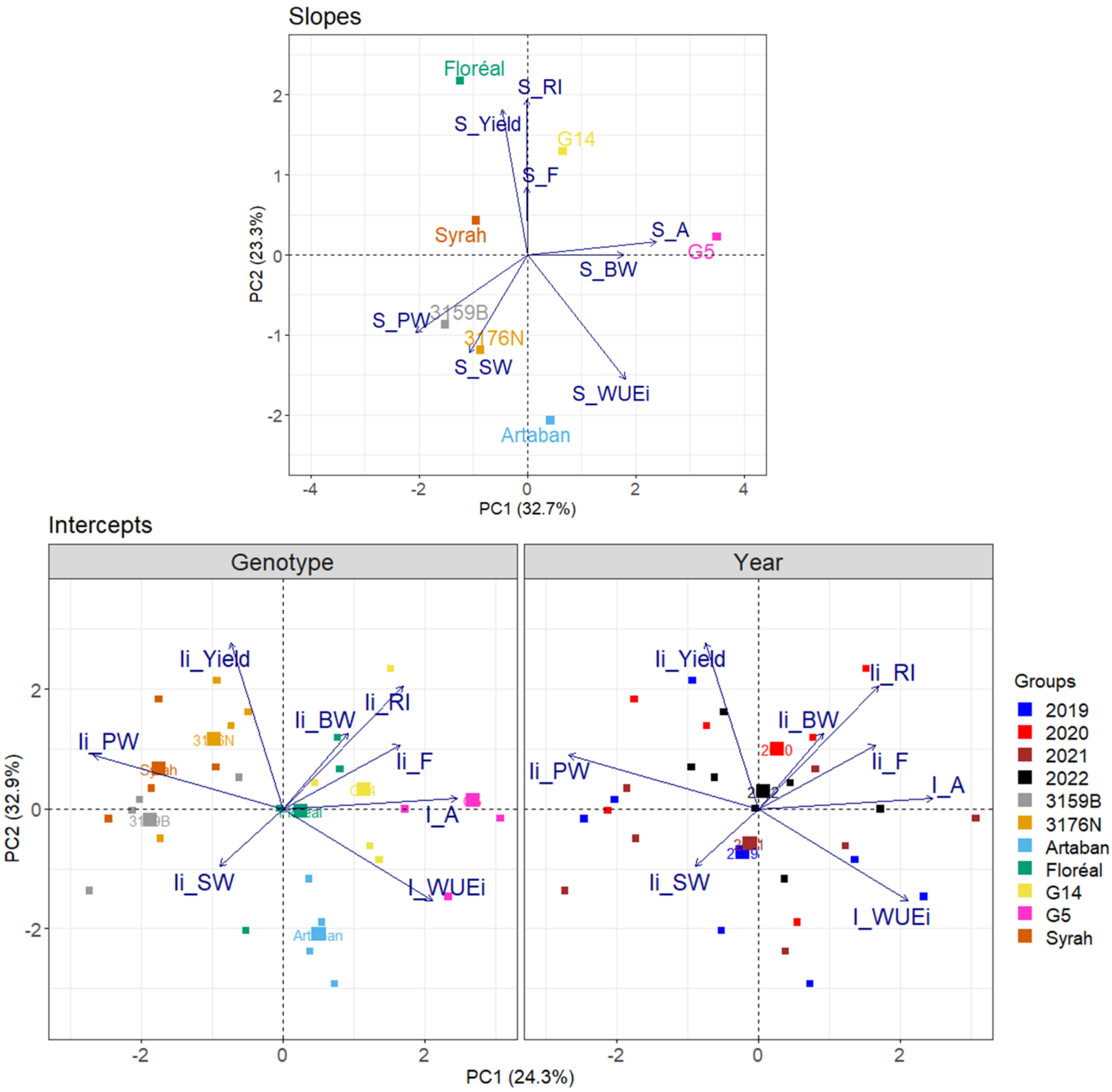

Figure 2. PCA of plant responses, slopes and intercepts, for berry weight (BW), Yield, number of clusters per shoot (F), pruning weight (PW), shoot weight (SW), Ravaz index (RI), photosynthesis (A), intrinsic water use efficiency (WUEi) per genotype and year, Gruissan - France.