What does the functional-structural plant model HydroShoot tell us about the reasons for grapevine (*Vitis vinifera* L.) photosynthesis depression?

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Abstract

Grapevine is a species that get along with water deficit. Yet, it cannot always stand thirst when accompanied by high temperatures, reducing noticeably its gas-exchange rates. Elucidating the origins of this reduction is a challenge, regarded the complex hydraulic, biochemical and energy processes lying behind gas-exchanges.

In this work we analyze data collected from an experiment conducted at he whole plant scale on Syrah vines with the aid of the functional-structural plant model HydroShoot. During our experiment, we submitted grapevines to a severe water stress and observed a steep drop in whole plant photosynthetic rates at midday, that was not due to stomatal closure, suggesting that both processes were decoupled at this moment. Using HydroShoot, we explore whether this decoupling results from a direct water limitation on biochemical processes.

HydroShoot links xylem hydraulic transport to gas and energy exchanges processes at the organ level. It simulates the effect of water deficit on xylem and stomatal conductances. The biochemical reactions of photosynthesis are affected by water deficit both indirectly through diffusional limitation and directly, through a reduced electron transport rate. Temperature affects photosynthetic rates through Arrhenius functions.

Using HydroShoot, we show that photosynthetic, midday depression could not be explained by simple hydraulic limitations. Bulk leaf water potential dropped to -1.6 MPa but this drop only affected J_{max} when temperatures exceeded 34 °C. Neither the Arrhenius response, nor the water limitation considered independently were sufficient to predict the observed drops. Only when responses to water and temperature were combined were we able to reproduce these observations, suggesting that photoinhibition may have occurred under these conditions.

Apart from an evidence of photoinhibition, our simulations indicate that xylem cavitation could not explain the observed drop in bulk leaf water potential. By contrast, a decrease in soil water potential has dramatic effects, much stronger than changes in xylem conductivity. The hydraulic architecture did not seem to play a major role in triggering stomatal closure.

We conclude that an adequate prediction of grapevines water use efficiency under water deficit conditions relies strongly on soil hydraulic properties and photoinhibition predictions.

Résumé

La vigne est une espèce qui a la réputation de tolérer le déficit hydrique. Toutefois, cette tolérance est réduite lorsque la vigne est soumise à la combinaison d'un déficit hydrique accompagné de températures élevées. Sous de telles conditions, les échanges gazeux entre la plante et l'atmosphère sont réduits par un ensemble de mécanismes complexes de transport hydrauliques, d'échanges d'énergie ou d'activité biochimique dont l'identification et la caractérisation représentent un défi.

Dans ce travail, nous analysons les résultats obtenus d'une expérimentation menée sur des vignes de Syrah à l'aide du modèle HydroShoot de type structure-fonction. Au cours de notre expérimentation, nous avons soumis les vignes à un déficit hydrique sévère et avons observé une forte dépression de l'activité photosynthétique à midi qui ne s'avérait pas totalement expliquée par la fermeture stomatique. En utilisant HydroShoot, nous avons testé si ce découplage résultait d'une limitation non-diffusionnelle de l'activité photosynthétique.

Le modèle HydroShoot intègre les processus de transport hydraulique, d'échange gazeux et d'échange d'énergie dans un seule cadre commun au niveau de l'organe. Il permet de simuler la cavitation du xylème et la réduction de la conductance stomatique sous l'effet du déficit hydrique. L'activité photosynthétique est affectée par le déficit hydrique à la fois indirectement par le biais de la fermeture stomatique et directement par le biais d'une fonction de photoinhibition. La photosynthèse varie également en fonction de la température des feuilles par le biais des fonctions d'Arrhenius.

En utilisant HydroShoot, nous montrons que la dépression photosynthétique observée ne peut pas s'expliquer par la seule limitation hydraulique. Bien que le potentiel hydrique des feuilles soit extrêmement bas, de l'ordre de -1.6 MPa, cette baisse n'aurait affecté les réactions biochimiques de la photosynthèse que sous des températures supérieures à 34 ° C. Les simulations effectuées n'ont permis de prédire la dépression photosynthétique que lorsque la fonction de photoinhibition était considérée, combinant les effets des stress hydrique et thermique.

Nous avons constaté par ailleurs que la simulation de la cavitation du xylème n'a que faiblement amélioré la prédiction des baisses de transpiration. Cette réduction résulte plus probablement de la baisse rapide de la conductivité hydraulique du sol.

Finalement, nous concluons qu'une prédiction adéquate de l'efficacité de l'utilisation de l'eau des vignes dans des conditions de déficit hydrique dépend fortement des propriétés hydrauliques du sol et de la prédiction de la photoinhibition.

1. Introduction

Soil water deficit is the predominant abiotic stress reducing global plant photosynthesis (Nemani et al., 2003). Its impact on photosynthesis is likely to get greater as drought events are expected to increase under changing climate. This fact put the viticulture community before the question of how to sustain production levels in the future (White et al., 2006, Hannah et al., 2013) calling for an improved prediction of water deficit effects on grapevine (*Vitis vinifera* L.) production.

Water stress affects grapevine photosynthesis by two possible mechanisms: either through inhibition of CO_2 metabolism (e.g. Tezara, 1999) or through a reduced CO_2 diffusion from ambient air to carboxylation sites (e.g. Flexas et al., 2004, 2012, Grassi and Magnani, 2005). Both, diffusional and non diffusional limitations may co-occur, although experimental evidence suggests that non diffusional limitations are rather to occur under a combination of severe water deficit and elevated temperature and irradiance (Björkman & Pöwls, 1984; Escalona et al., 1999, Flexas and Medrano, 2002). This combination is not uncommon in a viticulture that is widely spread under mediterranean climate regions where the growing seasons are often dry and hot.

A reliable prediction of the impact of soil water deficit on grapevine production requires thus a reliable representation of photosynthesis response to water stress, not only through diffusional limitation but also through biochemical inhibition.

In this study we discuss the importance of including photoinhibition functions in plant gas-exchange models. We use the functional-structural plant model HydroShoot (Albasha et al., 2016) which includes all known hydraulic limitations to simulate gas-exchange dynamics of a grapevine submitted to a severe water deficit and elevated air temperatures. We explore furthermore the respective role of simulating xylem cavitation and leaves temperature on gas-exchange rates predictions.

2. Materials and Methods

2.1. Experimental setup and data collection

The experiment was conducted in the summer of 2009 at the French National Institute for Agricultural Research (INRA) centre in Montpellier (3°53” E, 43°37” N, 44 m alt), France. 20-year old grapevine (*Vitis vinifera* L., cv. Syrah) were planted in a shallow sandy loam soil, trained using the vertical shoot positioning (VSP) system, with both its maximum height and row spacing equal to 1.8 m.

The monitored plants were submitted to soil water deficit by controlling irrigation rates so that predawn water potential was maintained between -0.30 and -0.50 MPa during the experiment. Plant’s performance under water deficit was monitored by measuring diurnal courses of gas-exchange rates in addition to continuous monitoring of temperature of 10 individual leaves distributed throughout the canopy.

Figure 1 : An open portable gas-exchange chamber completely enclosing the monitored grapevine's canopy in summer 2009. The chamber is used to assess the rates of net CO_2 assimilation and H_2O transpiration of the entire plant.

Whole plant gas exchange rates, i.e. net CO_2 assimilation and H_2O transpiration, were registered continuously using an open portable gas-exchange chamber which completely surrounded the plant canopy (Fig. 1). The chamber system consisted of a cylinder of 1.5 m diameter covered by an open-top cone, with a total 3 m height and 3.36 m^3 volume. The frame of the chamber was made from flat aluminum bars covered by a thin polypropylene film (RXD32 Propafilm, Innovia Films Ltd. UK) which has 90% transmission to solar irradiance in the PAR band. Ambient air was injected through the chamber using a blower, through two holes drilled in the bottom foam. Air was uniformly distributed through the chamber using holed plastic socks ("plenums") and air flux was controlled to satisfy a tradeoff between homogenizing air temperature inside the chamber (high flux) and keeping a differential in gas concentrations between the inlets and the outlets (low flux). Air flow entering the chamber was continuously recorded using a differential pressure transducer (PX170, Omega Engineering Inc., UK).

Total plant transpiration (E) was estimated as proposed by Long et al. (1996):

$$E=u_e(h_e-h_o) \qquad (1)$$

where E is given in [µmol s^{-1}] u_e is the flow entering the chamber [mol s^{-1}], h_e and h_o are respectively the mole fraction of H_2O of the ambient air at the entry and outlet of the chamber [µmol mol^{-1}].

Analogously, net CO_2 exchange rate (A_n) is calculated as:

$$A_n=u_e(C_e-C_o) \qquad (2)$$

where A_n is given in [µmol s^{-1}], C_e and C_o are respectively the mole fraction of CO_2 of the ambient air at the entry and outlet of the chamber [µmol mol^{-1}].

The temperature of individual leaves was measured using thermocouples inserted into the primary veins of 10 individual leaves positioned on different heights from the top of the canopy to the inside, so that temperature gradient resulting from different irradiance conditions may be captured.

Finally, at the end of the experiments, plant structure was digitized using an electromagnetic 3D digitizer (Fastrak, Polhemus Inc., Colchester, VT, USA). Destructive estimation of total leaf surface was then performed in order to complete the description of shoot architecture. Digitization data were used to reproduce plant mock-ups to be used in HydroShoot.

2.2. HydroShoot model

HydroShoot (Albasha et al., 2016) is a functional-structural plant model that links explicitly plant shoot architecture to the processes of irradiance interception, xylem hydraulic architecture, stomatal conductance, photosynthesis and leaf energy budget, with retroactions between gas-exchange, xylem hydraulic transport and energy-exchange processes.

In this manuscript, we will only briefly develop the empirical approach used for the simulation of joint impact of water and heat stress on CO_2 metabolism inhibition. The readers may refer to the Appendices for the description of CO_2 assimilation model and to Albasha et al. (2016) for a complete description HydroShoot.

We assume in this study that CO_2 inhibition is mainly due to a reduced electron transport rate (J) in the thylakoid due to joint water and heat stresses. Under such conditions, we increase the deactivation energy ΔH_d in J response to temperature (Eq. A6), steepening thus its reduction under high temperatures:

$$\Delta H_{d\,min} = \Delta H_{d\,min1} - (\Delta H_{d\,min1} - \Delta H_{d\,min2}) * \min(1, \max(0, (T_{leaf} - T_{leaf1}))/(T_{leaf2} - T_{leaf1})) \quad (3)$$

$$\Delta H_d = \Delta H_{d\,max} - \max(0, (\Delta H_{d\,max} - \Delta H_{d\,min}) * \min(1, (\Psi - \Psi_{max})/(\Psi_{min} - \Psi_{max}))) \quad (4)$$

where ΔH_d is given in KJ mol^{-1}, $\Delta H_{d\,min}$ is the minimum value of ΔH_d after considering the impact of leaf temperature T_{leaf} [°C], $\Delta H_{d\,min1}$ and $\Delta H_{d\,min2}$ are empirical thresholds of ΔH_d corresponding to arbitrarily defined leaf temperatures T_{leaf1} and T_{leaf2}. The final value ΔH_d is calculated after accounting for leaf water potential effect Ψ when it falls between two thresholds Ψ_{max} and Ψ_{min}, respectively, corresponding to $\Delta H_{d\,max}$ and $\Delta H_{d\,min}$, respectively.

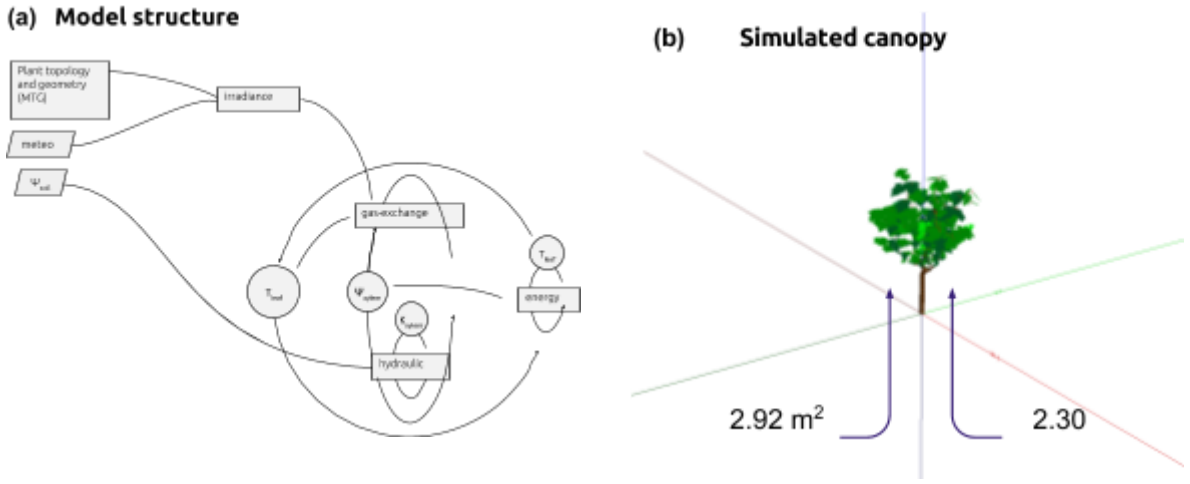

Figure 2 : (a) Structure of the HydroShoot model, (b) digital mock-up of the grapevine shoot under study.

3. Results and Discussion

Gas-exchange rates of whole plant canopy are shown in Figure 2 together with the observed air vapor pressure deficit and temperature.

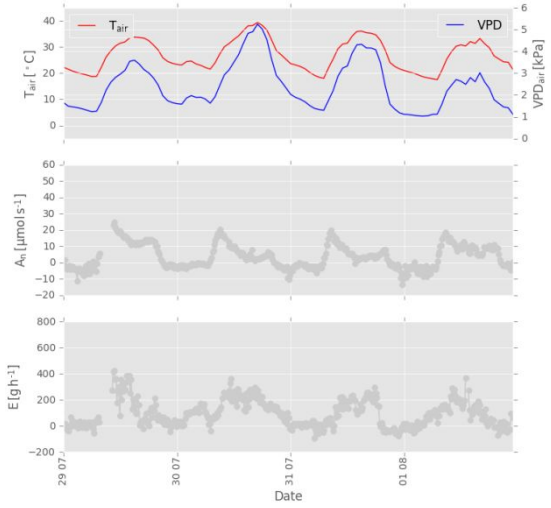

Figure 2 : Diurnal courses of observed air temperature and vapor pressure deficit (upper panel), canopy net CO_2 assimilation (middle panel) and canopy transpiration (lower panel) of a whole plant.

Air temperature inside gas-exchange chamber was high during the experiment, reaching a maximum of 39.4 °C during the second day in Fig. 2, accompanied by a strong peak in vapor pressure deficit reaching almost 5.5 kPa. These severe atmospheric conditions, combined with a strong soil water deficit, dramatically affected both A_n and E (their respective maxima reached 23 µmol s^{-1} and 410 g h^{-1}) but with distinct diurnal courses. A_n decreased drastically during the afternoon, almost

ceasing around 5 pm before recovering by the end of the day, while E showed no afternoon depression and followed the course of the canopy-to-air vapor pressure deficit.

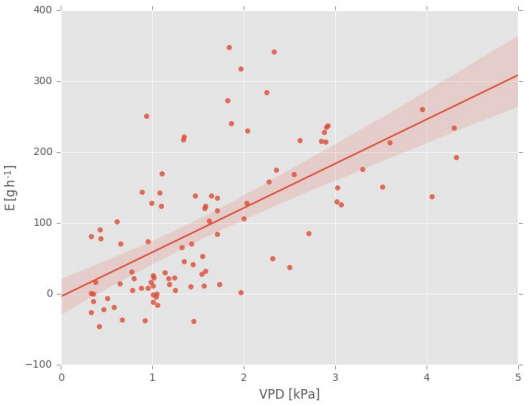

Figure 3 : Relationship between canopy transpiration (E) and air vapor pressure deficit (VPD).

The relationship between observed E and VPD (Figure 3) indicates that canopy-lumped, stomatal conductance to water (g_{sw}) was not as drastically reduced as A_n at high VPD values (i.e. during the peak of climatic demand in the afternoon). This suggests that stomatal conductance was not the only limiting factor causing the strong reduction in A_n but that the biochemical process of photosynthesis itself was limiting, probably due to some inhibition induced by co-occurring water and heat stresses. We used HydroShoot to examine this hypothesis, by performing simulations with and without a function to mimic photoinhibition. The results supported the hypothesis that diffusional pathways were not the only limiting factor behind photosynthetic depression in the afternoon (Fig. 4).

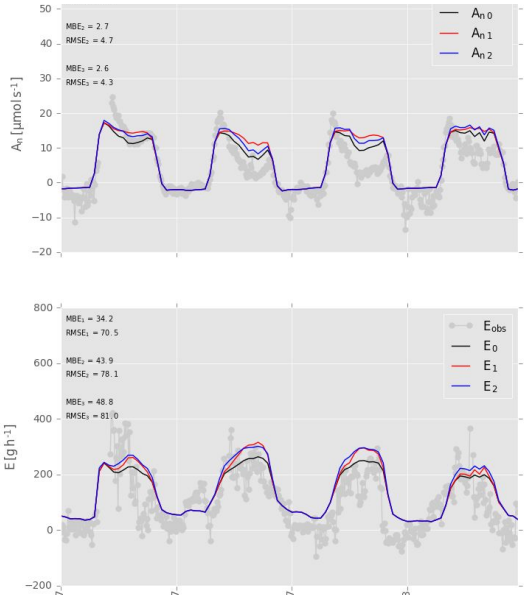

Figure 4 : Diurnal courses of observed (grey dots) and simulated (continuous lines) rates of net CO2 assimilation and transpiration for the entire canopy. The indices 0, 1 and 2 refer to simulation configurations (0: photoinhibition and hydraulic structure are considered, 1: photoinhibition is not considered while the hydraulic structure is not taken into account, 2: photoinhibition is considered while hydraulic structure is not).

Figure 4 shows that when simulations were performed with separated water and heat stress effects on A_n and gs (scenario 1 in Figure 4), HydroShoot failed to reproduce the observed diurnal patterns of A_n.

Our findings agree with reported studies in the literature, confirming that photosynthesis may be decoupled from stomatal conductance under high irradiance, heat and dry conditions, due to the inhibition of the biochemical activity of photosynthesis (Correia et al., 1990, Flexas et al., 1999, Escalona et al., 2000, Flexas and Medrano, 2002, Maroco et al., 2002, Yu et al., 2009, Wang et al., 2012). Flexas and Medrano (2002) indicated that diffusional conductance, mainly stomatal, limits the photosynthetic activity at mild to moderate drought conditions while inhibition of metabolic processes predominate in the control CO_2 assimilation as drought severity increases. This points out to the necessity of considering photosynthetic limitation by other ways than hydraulic in order to accurately predict gas-exchange dynamics under severe water-deficit conditions.

4. Conclusion

We showed in this study that midday depression in grapevine photosynthesis could not be explained only by hydraulic limitations. A reliable prediction of soil water deficit on grapevine gas-exchange rates requires thus considering the combined effect of water and temperature on CO_2 assimilation rate. Furthermore, this study case recall the predominant role of soil water conductivity on water transport in through the soil-plant-atmosphere continuum. Next to soil hydraulic reduction, that of the xylem plays only a minor role in triggering stomatal closure.

Acknowledgments

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Appendix

Net CO_2 assimilation (A_n) is calculated according to Farquhar's model **(Farquhar et al., 1980)**. It is the result of the dual between carbon fixation by the Rubisco enzyme (the carboxylation activity) and carbon loss by oxygenase and day respiration. A_n may be estimated as the minimum of the Rubisco carboxylation rate (W_c) RuBP-limited carboxylation rate (W_J) and Triose phosphate utilization-limited carboxylation rate W_P :

$$V_c = min\left(W_c, W_j, W_P\right) - R_d \quad \text{(Eq. A1)}$$

$$W_c = \frac{C_c V_{c\,max}}{C_c + K_c\left(1+\frac{O}{K_o}\right)} \quad \text{(Eq. A2)}$$

$$W_j = \frac{J}{4+8\frac{\Gamma*}{C_c}} \quad \text{(Eq. A3)}$$

$$W_P = \frac{3TPU}{\left(1-\frac{\Gamma*}{C_c}\right)} \quad \text{(Eq. A4)}$$

$$J = \frac{\alpha PPFD}{\sqrt{1+\frac{\alpha^2 PPFD^2}{J_{max}^2}}} \quad \text{(Eq. A5)}$$

where V_c is carboxylation rate $[\mu mol_{CO_2}\, m^{-2}\, s^{-1}]$, R_d the Mitochondrial respiration rate in the light $[\mu mol_{CO_2}\, m^{-2}\, s^{-1}]$, $V_{c\,max}$ the maximum carboxylation rate $[\mu mol_{CO_2}\, m^{-2}\, s^{-1}]$, C_C is chloroplast partial pressure $[\mu mol\, mol^{-1}]$, K_C and K_O are respectively the Michaelis-Menten constant for the carboxylase $[\mu mol\, mol^{-1}]$ and oxygenase $[mmol\, mol^{-1}]$, O oxygen partial pressure $[mmol\, mol^{-1}]$, J is electron transport rate $[\mu mol_{electron}\, m^{-2}\, s^{-1}]$, alpha Initial quantum yield $[mmol_{CO_2}\, mmol_{photon}^{-1}]$, $PPFD$ is the photosynthetic photon flux density $[\mu mol_{photon}\, m^{-2}\, s^{-1}]$, J_{max} is the maximum electron transport rate $[\mu mol_{electron}\, m^{-2}\, s^{-1}]$ and TPU is the triose phosphates transport rate $[\mu mol_{CO_2}\, m^{-2}\, s^{-1}]$.

Leaf biochemical reactions are known to depend on temperature. This temperature dependency is described according to **Bernacchi et al. (2003)** :

$$P = exp\left(c - \frac{\Delta H_a}{R\left(T_{leaf}+273.15\right)}\right) \quad \text{(Eq. 6a)}$$

$$P = P^{25}\frac{exp\left(c-\frac{\Delta H_a}{R\,T_{leaf}}\right)}{1+exp\left(\frac{\Delta S\,T_{leaf}-\Delta H_d}{R\,T_{leaf}}\right)} \quad \text{(Eq. 6b)}$$

where P is value of either of the parameters Γ_*, K_C , K_O (Eq. 6a) or $V_{c\,max}$, J_{max}, TPU and R_d (Eq. 6b) under the actual leaf absolute temperature T_{leaf} $[K]$, P^{25} is P value of $V_{c\,max}$, J_{max}, TPU and R_d under 25 $°C$ temperature, C is a dimensionless shape parameter of the Arrhenius function, ΔH_a and ΔH_d are respectively the activation and deactivation energies $[kJ\, mol^{-1}]$, ΔS an entropy term $[kJ\, K^{-1}\, mol^{-1}]$ and R is the ideal gas constant (8.314e-6 $m^3\, MPa\, mol^{-1}\, K^{-1}$).