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# Simulating the impact of climate change (elevated $CO_2$ and temperature, and water deficit) on the growth of red and white Tempranillo grapevine in three consecutive growing seasons (2013–2015)

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#### ABSTRACT

In recent decades, agricultural production is being affected by a sharp increase in atmospheric CO<sub>2</sub> concentration. Due to the greenhouse effect gases, crops are impacted by enhanced temperatures and concomitantly by increased scarce water availability. All arid and semiarid areas, including Mediterranean viticulture, must face these three climate change-related factors: atmospheric CO<sub>2</sub> concentration and temperature increases, and scarce water for irrigation. Scarce water is a problem even in irrigated viticulture, as irrigation is becoming more and more restricted. Within this context, the aim of this work was to investigate grapevine (Vitis vinifera L. cv. red and white Tempranillo) vegetative and reproductive growth. Fruit-bearing cuttings were grown under elevated CO<sub>2</sub> (around 700 µmol mol<sup>-1</sup> or ppm, versus 400), high temperature (ambient temperature + 4 °C, versus ambient) and water deficit (cyclic drought, versus full irrigated) in temperature gradient greenhouses for three consecutive growing seasons (years 2013, 2014 and 2015). Climate change impacted markedly vegetative growth. Within the abovementioned factors, vegetative growth (total vegetative mass) was significantly reduced by drought (consistent the three years) and was associated to a low substrate water status and low leaf stomatal conductance. Elevated CO2 stimulated total vegetative mass, whereas leaf area was not affected. When plants were grown under elevated CO<sub>2</sub>, the largest increases were observed in leaf (white) and root (red) growth. In these experiments, elevated CO<sub>2</sub> did not compensate the negative effects of water stress. An increase of the mean temperature 4 °C had no consequences on vegetative growth. Yield and yield-related traits were unaffected by the climate change scenario.

#### 1. Introduction

Greenhouse effect gases, such as  $CO_2$ , chlorine-fluorine-carbon (CFC) and its derivatives,  $CH_4$  and  $N_2O$ , are causing the recent increases in temperature at a global scale. Atmospheric  $CO_2$  concentration has increased since preindustrial period from 280 µmol mol<sup>-1</sup> (ppm) to currently more than 400, and the predictions for the end of the century range from 421 to 936 (Meinshausen et al., 2011). Cumulative emissions of  $CO_2$  contribute largely to the global mean surface warming. Foreseen values range from stabilization at 1.5 °C higher than the current reference period (1986–2005) to a more than 4 °C increase, depending on the mitigation measures adopted (IPCC, 2014). During the last two decades, for instance, air temperature has increased 0.85 °C (IPCC, 2014). This greenhouse effect is impacting crops, causing more frequent Mediterranean crop drought (NOAA, 2011) that results from reduced water supply and increased transpiration due to the higher temperature. Grapevine is a typical Mediterranean crop fully adapted to summer drought, but its response to climate change-related factors (elevated CO<sub>2</sub>, high temperature and drought) can vary when stress factors act individually and/or interacting (Kizildeniz et al., 2015).

An instantaneous response to an elevated atmospheric  $CO_2$  is an increased photosynthesis and a decreased transpiration, due to higher substrate availability for C fixation and stomata partial closure respectively (Lambers et al., 1998; Long, 1991). Consequently, elevated  $CO_2$  was reported to stimulate plant growth in different species (Ainsworth and Long, 2005; Drake et al., 1997; Kimball and Idso,

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Abbreviations: DW, dry weight; FW, fresh weight; IPCC, intergovernmental panel on climate change; T, ambient temperature; T + 4, 4 °C more than ambient temperature; WA, water availability; FI, full irrigation; CD, cyclic drought; TGGs, temperature gradient greenhouses

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1983). As in other  $C_3$  species where photosynthesis is limited by  $CO_2$  availability, grapevine responds to the elevated  $CO_2$  with an increased vegetative growth and yield (Bindi et al., 1996; Bowes, 1993; Rogers et al., 1994). However, long-term  $CO_2$  exposure leads to a phenomenon known as photosynthetic acclimation or down regulation that decreases grapevine photosynthetic capacity (Salazar-Parra et al., 2015), resulting in less clear effects on grapevine physiology and growth. Secondary changes in morphology, allocation and chemical composition may also influence growth (Poorter et al., 1997).

Studies of elevated temperature on grapevine have been focused on berry composition, due to its association with grape ripening (Kuhn et al., 2014). Besides its effects on berries, depending on the level of elevated temperature and duration, it could also affect grapevine physiology, in particular photosynthesis, and vigor. Rates of photosynthesis can be reduced 30-50% when temperature rises over the 30 °C lightsaturated optimum (Greer and Weedon, 2013). Photosynthesis decreases with increasing temperatures were attributed only 15-30% to stomatal closure (Greer and Weedon, 2012; Zsófi et al., 2009). More extreme temperatures can also inactivate the CO<sub>2</sub> fixing Rubisco (Ribulose-1,5-biphosphate carboxylase oxygenase) enzyme, which results in a drastic reduction of photosynthesis and limits the berry sugar supply causing slow ripening (Greer and Weedon, 2013). As a consequence of photosynthesis (and sugar accumulation) decreases under high temperatures, growth and yield are expected to decrease (Medrano et al., 2003).

Grapevine is traditionally a non-irrigated crop that is cultivated on 6 out of 7 continents (Hofmann and Schultz, 2015), where it occupies quite extensive agricultural areas in arid and semiarid regions. Water is the most limiting resource in these environments, such as the Mediterranean region, where rainfall is scarce and irregularly distributed along the year. Climate change models predict even more arid conditions for the future (Ashour and Al-Najar, 2012; Collins et al., 2013). Grapevine has developed physiological and morphological mechanisms enabling it to produce under such circumstances (Koundouras et al., 2008). Irrigation is a major tool for grapevine growers to increase soil water availability, which should lead to potential higher yields with (Salon et al., 2005; Valdes et al., 2009; Williams and Matthews, 1990) or without (Reynolds et al., 2007; Smart and Coombe, 1983) quality losses. Depending on the intensity of water stress and on the grapevine developmental stage, soil water availability nevertheless has more influence on grapevine vegetative growth than on yield (Kizildeniz et al., 2015; Korkutal et al., 2011; Williams and Matthews, 1990). For viticulture purposes, a managed deficit irrigation may be used to control grapevine vegetative growth and quality (Kennedy et al., 2002).

In the literature, effects of climate change (elevated  $CO_2$ , high temperature and water scarcity) on grapevine physiology have been investigated as individual effects more than the interactions among them. When individual effects are considered, exposure time, duration and sequence are essential for growth. For instance, increased number of warm days during flowering-veraison period and low precipitation during maturation result in greater berry weights (Jones and Davis, 2000). Combination of all abovementioned climate change-related factors has more severe effects on berry weight (McCarthy, 1997). When high temperature and water deficit are combined, leaf water potential, stomatal conductance, and net photosynthesis are reduced (Edwards et al., 2011). In the long-term, if these factors persist, the leaf area can be affected. Stimulation of grapevine production by elevated  $CO_2$  is decreased or completely cancelled in presence of high temperature (Bindi et al., 2001).

Grapevine works focused on multiple stress factors are rarely performed because such studies in the field or under controlled conditions are complex, difficult and expensive to execute. The aim of this study was to analyze the influence of the three main climate change-related factors (elevated  $CO_2$ , high temperature and water scarcity), acting both individually and/or interacting, on plant vegetative and reproductive (yield) growth in fruit-bearing cuttings of two grapevine cultivars (red and white Tempranillo). Plants were grown in temperature gradient greenhouses (TGGs) during 3 consecutive growing seasons (2013, 2014 and 2015).

#### 2. Materials and methods

#### 2.1. Plant material

Dormant cuttings of *Vitis vinifera* L. cv. red (accession T43, clone RJ-43) and white Tempranillo (accession CI-101 in "La Grajera" germplasm bank, Riojás Government, Spain) were sampled from an experimental vineyard of the Institute of Sciences of Vine and Wine (ICVV) in Logroño (La Rioja, Spain). Experiments were conducted using each year new cuttings (not imposing treatments on the same plants for 3 consecutive years). Tempranillo is a local red grape variety widely cultivated in northern and central Spain, currently expanded to many other viticulture areas in the world. A new white berry somatic variant of Tempranillo ("white Tempranillo") is the result of a spontaneous, natural mutation, which was discovered in an old red Tempranillo vineyard in 1988 in Murillo de Río Leza, La Rioja, Spain (García-Escudero et al., 2011) and vegetatively propagated from one single grapevine plant.

#### 2.2. Growth conditions

We used two types of greenhouses for growing the plants. Until fruit set, plants were grown in a pre-culture greenhouse with an effective cooling system where temperature and relative humidity (RH) were set at 26/15 °C and 60/80% (day/night) respectively and with a photoperiod of 15 h using natural daylight supplemented with high-pressure metal halide lamps (OSRAM<sup>\*</sup>, Augsburg, Germany), providing a photosynthetic photon flux density (PPFD) of 500 µmol m<sup>-2</sup> s<sup>-1</sup> at inflorescence level. Variability around target temperature and RH values was typically 1.5–2% and 8–12%, respectively (Morales et al., 2014). After fruit set, and coincident with the start of the treatments, plants were moved to the second greenhouses, i.e., the temperature-gradient greenhouses (TGGs). Table 1 summarizes temperature and RH data within the TGGs in 2013, 2014 and 2015.

Fruit-bearing cuttings were obtained according to Mullins (1966), as described by Kizildeniz et al. (2015). Briefly, cuttings were treated with indole butyric acid ( $300 \text{ mg L}^{-1}$ ) in a heated rockwool moist-bed (25–27 °C) kept in a cool chamber (5 °C). One month later, the rooted-cuttings were planted in 0.8 L plastic pots containing a mixture of sand, perlite and vermiculite (1:1:1, in volume) and transferred to the preculture greenhouse. Only a single flowering stem was allowed to develop on each plant, in order to get only one berry bunch per plant. Until fruit set, vegetation was controlled and only 4 leaves per plant was allowed to grow. At fruit-set, the cuttings were transplanted to 13 L plastic pots including a peat and perlite (2:1, v/v) mixture.

Field capacity, permanent wilting point and available water content of the peat:perlite mixture were determined using a soil moisture equipment with ceramic plates of 33 and 1500 kPa (Richards, 1941). Bulk density was calculated weighing the dry substrate contained in a given volume. The characteristics of the peat-perlite mixture used with

#### Table 1

Temperature and relative humidity recorded in the temperature gradient greenhouse experiments with red and white Tempranillo grapevine. T, ambient temperature and T + 4 °C, ambient temperature + 4 °C.

| Year   | 2013              |                    | 2014              |                    | 2015               |                    |
|--|-------------------|--------------------|-------------------|--------------------|--------------------|--------------------|
| Temperature regime   | Т                 | T + 4 °C           | Т                 | T + 4 °C           | Т                  | T + 4 °C           |
| Daily mean temp. (°C)<br>Days above 35 °C<br>Daily mean RH (%) | 21.2<br>2<br>64.9 | 24.2<br>36<br>57.0 | 22.1<br>8<br>65.4 | 26.5<br>42<br>52.1 | 22.9<br>19<br>63.7 | 27.1<br>50<br>52.7 |

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