# CO<sub>2</sub>-induced microclimate changes help wheat to adapt to future climates

Markus Löw<sup>1</sup>, Shihab Uddin<sup>1,2</sup>, Glenn Fitzgerald<sup>1,3</sup> and Michael Tausz<sup>4,5</sup>

#### **Abstract**

Optimising water use is a central element of maintaining crop growth and yield in less favourable, drier climates. Rising  $CO_2$  levels alter responses of plants. Some of these responses can be used to help mitigate the effect of rising temperature, less frequent precipitation and prolonged drought. We speculate that closure of stomata under elevated  $CO_2$  (e $CO_2$ ) allows the plant not only to take advantage of higher water use, but also changes the microclimate within the canopy, around the plant. Increased water use efficiency under e $CO_2$  is well known, but does this allow wheat in a dryland agriculture system to lower its in-canopy air-temperature and vapour pressure deficit (VPD) which will have positive effects on sap flow, and leaf water status?

### **Keywords**

Climate change, water use, microclimate, FACE, VPD.

### Introduction

Adapting crops to future climates is a challenge in agriculture. Increasing temperature and uncertainties about precipitation events (CSIRO and BOM 2016) call for wheat cultivars that can survive in changing conditions (Porter and Semenov 2005). However, plants are a biological link between the soil and the atmosphere and can influence their environment to a certain extent. Elevated CO<sub>2</sub> (eCO<sub>2</sub>) allows plants to close stomata further, limiting water loss and therefore increasing their water use efficiency (Kimball 2016). At the same time, eCO<sub>2</sub> stimulates biomass and leaf area and lowers soil evapotranspiration due to shading from greater early season growth. This should leave more water in the soil during grain filling of wheat in Australian dryland agriculture (O'Leary et al. 2015). In addition, a cultivar with a genetically improved water use efficiency and early vigour (e.g., cv. Scout) can potentially take further advantage of eCO<sub>2</sub> (cf. Tausz-Posch et al. 2012). With more soil water available, plants are able to transpire more which will potentially lower the air-temperature in their surrounding canopy which then lowers the vapour pressure deficit (VPD), a driver of stomatal conductance, to less demanding values (Kimball et al. 1995; Franzaring et al. 2010). This additional available water should show up as increased sap-flow through the stem and allow the plants to maintain a favourable leaf turgor pressure. As a consequence, plants may be able to counteract some of the predicted increased temperatures in future climates, although perhaps to only fractions of a degree. Still, a more favourable microclimate surrounding the plant should lead to improved yield under eCO<sub>2</sub>. This study compares two wheat cultivars (one cultivar with increased water use efficiency) under current and future CO<sub>2</sub> concentrations (400 and 550 mol mol-1) using Free Air CO<sub>2</sub> Enrichment (FACE) to test if plants can 'do more with less'. We expect that under eCO<sub>2</sub> there will be higher soil moisture, increased sap flow, lower leaf turgor pressure, lower air-temperature, lower in-canopy VPD, higher yield and possibly harvest index. These responses should be larger for a water-use-efficient cultivar.

## Methods

Plant material, harvests and site

The wheat cultivars Scout and Yitpi have contrasting water use efficiencies and vigour. Scout contains a gene that improves WUE compared to Yitpi and has good early vigour (PacificSeeds 2010). Destructive harvests took place at physiological maturity to determine grain yield, above-ground biomass and harvest index. The experiment took place near Horsham, VIC, Australia, at the Australian Grains Free Air CO<sub>2</sub> Enrichment (AGFACE) facility during the growing seasons 2014 and 2015. AGFACE is set up as a randomised block design with eight 'rings' as statistical blocks. Four of the rings were equipped with FACE

<sup>&</sup>lt;sup>1</sup> Faculty of Veterinary and Agricultural Sciences, The University of Melbourne, 4 Water Street, Creswick, VIC 3363, m.loew@unimelb.edu.au.

<sup>&</sup>lt;sup>2</sup> Department of Agronomy, Bangladesh Agricultural University, Bangladesh

<sup>&</sup>lt;sup>3</sup> Department of Economic Development, Jobs, Transport and Resources, Private Bag 260, Horsham, VIC 3401

<sup>&</sup>lt;sup>4</sup> School of Ecosystem and Forest Sciences, The University of Melbourne, 4 Water Street, Creswick, VIC 3363

<sup>&</sup>lt;sup>5</sup> Birmingham Institute of Forest Research, School of Biosciences, University of Birmingham, Edgbaston, UK

technology that allows increasing the prevailing day-time  $CO_2$  concentration to ~550 ppm (e $CO_2$ ) to mimic future climates as compared to the current ambient  $CO_2$  concentration of ~400 ppm (a $CO_2$ ). Details on the FACE system are published by Mollah et al. 2009. Within each ring, 4 m long plots were established to grow a variety of wheat cultivars. Scout and Yitpi were planted in two plots per ring. One plot received regular irrigation to create an unlimited water supply (wet) with growing season rain (GSR) + irrigation of 449 mm and 429 for 2014 and 2015, respectively. The other plot should have received rainfall water only (rainfed). But due to low seasonal rainfall in both growing seasons, rainfed plots received some supplemental irrigation (GSR including minor irrigation = 261 mm and 257 mm for 2014 and 2015) to allow the plants to reach maturity.

## Volumetric soil water content

Volumetric soil water content (vSWC) was measured weekly with a PR2 soil moisture profile probe (with hand-held reader HH2, Delta-T Devices, Burwell, UK). An access tube for the soil moisture probe was installed in each plot after sowing. A 1 m long sensor rod was inserted into the access tubes and vSWC was recorded at six depths at 100, 200, 300, 400, 600, and 1000 mm below the surface. Physical vSWC samples (soil cores) were taken before, during and after the growing seasons to calibrate the sensor to the local soil type and each depth.

## Sap flow

After flowering, sap flow sensors (based on stem heat balance principles, Dynagage SGA2, Dynamax Inc, Houston, USA), were installed on the stems. To maximise the leaf area above the sensor, they were installed as low as possible on the stem where the stem diameter allowed. There were two leaves above the sensor. Several layers of insulation were installed to minimise the effects of radiation and temperature on the stem heat balance. Leaf area above the sensor was calculated from length and width of the leaf blades. Relative sap flow is calculated as sap flow per leaf area above the sensor.

## In-canopy microclimate

Small, self-built air-temperature and relative humidity sensors were installed within the canopy after stem elongation. The sensors were in the vicinity of the sap flow sensor (see above), but higher up in the canopy to represent the microclimate surrounding the leaves that contribute to sap flow. Data was collected by a CR1000 data logger system (Campbell Scientific, Garbutt, Australia) located in each ring.

### Leaf water status, turgor pressure

Leaf patch clamp pressure probes (ZIM-sensor, YARA ZIM Plant Technology GmbH, Henningsdorf, Germany) were used to continuously and non-destructively measure leaf water status. The sensor consists of two magnets that clamp on the leaf blade from the top and bottom. One of the magnets has an embedded pressure sensor touching the leaf. The variable turgor pressure in the leaf (Pp) opposes the magnetic pressure and changes depending on the leaf water status. Pp and the magnetic pressure are inversely coupled (Zimmermann et al. 2013), and with the pressure exerted by the magnets on the leaf being constant, the leaf turgor pressure Pp can be measured. The measurement principle is described in detail by Zimmermann et al. (2013). The diurnal course of Pp is used to investigate changing Pp between times when stomata are open (mostly in the morning) and when they are closed, usually after 14:00 hrs. The diurnal pattern of Pp indicates the ability of a plant to adjust to changing microclimatic conditions and water status.

## Statistical analysis

The physical design of AGFACE was closely replicated in the statistical design. Linear mixed-effect models were used in a variety of combinations depending on measured parameter. Fixed factors were either CO<sub>2</sub>, cultivar, irrigation, or all, with corresponding random effects (ring, depth, time) as required. The variance structure of the models was adapted to accommodate homogeneity of variances based on model selection criteria.

#### Results

#### Soil water content vSWC

No difference in vSWC between the two cultivars was found (Figure 1). Small differences of about 0.02 m<sup>3</sup> m<sup>3</sup>, especially under eCO<sub>2</sub>, were not statistically significant. However, overall vSWC responded to eCO<sub>2</sub> (p

< 0.001), but the direction and magnitude of the effect depended on season, growth stage, and depth. Compared to aCO<sub>2</sub>, most water was saved in the lower soil layers (600 and 1000 mm depth) under eCO<sub>2</sub> towards the end of the season in both years. In 2014 vSWC under eCO<sub>2</sub> was increased from before flowering to maturity (data not shown) and in 2015 for most of the season. No water was saved under eCO<sub>2</sub> at the soil layers between 100-600 mm depth.

## *In-canopy microclimate*

In-canopy VPD at midday from about flowering to maturity (Figure 2A) was consistently lower under eCO2 (p < 0.01), however, a  $CO_2$  treatment x cultivar interaction was found. Indeed, VPD in a cv Scout canopy was on average about 0.2 kPa lower under eCO<sub>2</sub> as compared to eCO<sub>2</sub> whereas VPD within the cv Yitpi canopy increased slightly, but not statistically significantly.

## Sap flow

An example time course of relative sap flow of cv Yitpi is shown in Figure 2B. After a  $\sim$ 3 hour long lag phase following sunrise, sap flow was increased to a maximum level at about 13:00 hrs each day and declined afterwards. Relative sap flow was very small, however small differences between aCO<sub>2</sub> and eCO<sub>2</sub> appear (p < 0.001) between noon and 15:00 hrs on sunny days (Nov 7 and 9, Figure 2B) when sap flow under eCO<sub>2</sub> decreased more rapidly compared to aCO<sub>2</sub>. This difference disappeared by about 16:00 hrs.

## Leaf water status

Scout responded more dynamically to diurnal VPD-changes between morning and afternoon compared to Yitpi (p < 0.01, not shown). eCO<sub>2</sub> increases this plasticity for both cultivars (p < 0.05) , and, at the same time, lowered the range of the daily turgor adjustments (p < 0.01).

## Harvest

The harvest index was lower under eCO<sub>2</sub> (p < 0.05), but was less reduced for cv Scout as compared to cv Yitpi (p < 0.05, data not shown). Yield and aboveground biomass responses were the same for both cultivars across all treatments and years, eCO<sub>2</sub> increased biomass by 10% for Yitpi and 21% for Scout relative to aCO<sub>2</sub>. But this large difference in eCO<sub>2</sub> response was not statistically significant.

## Conclusion

The ecophysiological responses of cvs Scout and Yitpi to eCO<sub>2</sub> were variable and an increased yield was not found. Still, several of our hypotheses regarding microclimate changes were confirmed which leads to the conclusion that the tested cultivars are able to alter their environment in their favour which could be utilised to ameliorate the impact of increased temperatures, more variable precipitation and more frequent drought in future climates. The more water-use efficient cultivar Scout can utilise the available water better than Yitpi, however, most of the gained resources don't seem to go into increased biomass, and they didn't affect yield. Future studies have to look into the interaction of biomass and water use further to find traits that might allow benefiting from more water without increasing biomass, therefore increasing the harvest index. This might include optimising below-ground (root growth) processes, and nutrient use.

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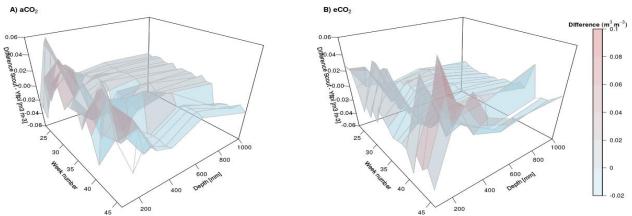


Figure 1. Difference between weekly volumetric soil moisture profiles of rainfed wheat cultivars Scout and Yitpi during 2015 under ambient (A) and elevated  $CO_2$  (B). Key growth stages were: stem elongation in calendar week 32, flowering in week 40, and maturity in week 46.

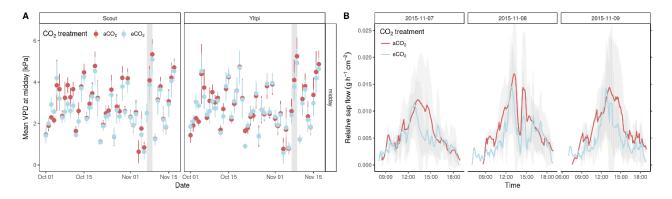


Figure 2. Microclimate and sap flow of wheat in AGFACE during October / November 2015. A) Mean in-canopy vapour pressure deficit (VPD) at midday (10:00 to 15:00) between flowering and maturity for wheat cultivars Scout and Yitpi. B) Mean diurnal course of relative sap flow of Yitpi during the three days that are shown as grey box in A. Error bars indicate standard deviation.