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# Growth of soybean at future tropospheric ozone concentrations decreases canopy evapotranspiration and soil water depletion

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# ABSTRACT

Tropospheric ozone is increasing in many agricultural regions resulting in decreased stomatal conductance and overall biomass of sensitive crop species. These physiological effects of ozone forecast changes in evapotranspiration and thus in the terrestrial hydrological cycle, particularly in intercontinental interiors. Soybean plots were fumigated with ozone to achieve concentrations above ambient levels over five growing seasons in open-air field conditions. Mean season increases in ozone concentrations ( $[O_3]$ ) varied between growing seasons from 22 to 37% above background concentrations. The objective of this experiment was to examine the effects of future  $[O_3]$  on crop ecosystem energy fluxes and water use. Elevated  $[O_3]$  caused decreases in canopy evapotranspiration resulting in decreased water use by as much as 15% in high ozone years and decreased soil water removal. In addition, ozone treatment resulted in increased sensible heat flux in all years indicative of day-time increase in canopy temperature of up to  $0.7 \,^\circ$ C.

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# 1. Introduction

Of the tropospheric trace gases increasing in concentration from anthropogenic activities, tropospheric ozone ([O<sub>3</sub>]) is, in addition to being the third most significant greenhouse gas (IPCC, 2007), the most damaging to vegetation (Heagle, 1989; IPCC, 2007). The main drivers for increasing  $[O_3]$  in the troposphere are higher concentrations of volatile organic compounds (VOCs) and of nitrogen oxides  $(NO_x)$  which react in the presence of sunlight to form  $O_3$ (Fowler et al., 1999). Understanding the responses of vegetation to increasing [O<sub>3</sub>] has important implications for assessing future productivity, ecosystem functioning and climate feedbacks. Emissions of these precursors have already led to  $[O_3]$  that have negatively impacted vegetation (US-EPA, 1986; Fiscus et al., 1997; Mauzerall and Wang, 2001: Fiscus et al., 2005). Continued and predicted increases in emissions of the precursors to O<sub>3</sub> formation (IPCC, 2007) will likely exacerbate damage to plants well into the future (Morgan et al., 2003; Van Dingenen et al., 2008).

Growth at elevated  $[O_3]$  consistently impairs growth and biomass accumulation in many species of all functional groups and

\* Corresponding author. E-mail address: bernacch@illinois.edu (C.J. Bernacchi). reduces yield for agronomically important crops (Bergmann et al., 1999; Feng et al., 2003, 2011; Morgan et al., 2003; Timonen et al., 2004; Fiscus et al., 2005; Karlsson et al., 2005; Wang et al., 2007; Ainsworth, 2008; Betzelberger et al., 2010; Fishman et al., 2010). This is anticipated to significantly impact the global carbon cycle, with the O<sub>3</sub>-induced reduction in the terrestrial carbon sink driving increases in atmospheric [CO<sub>2</sub>] that result in at least as much radiative forcing as the direct radiative forcing arising from higher [O<sub>3</sub>] alone (Sitch et al., 2007). Undetermined, however, is the impact that elevated [O<sub>3</sub>] will have on the hydrologic cycle through its effect on evapotranspiration (ET), the combined evaporation and transpiration of water from an ecosystem. Through the plant's physiological control of transpiration and the impact that canopy architecture has on evaporation, ET can have a large impact on local and regional climate, particularly for continental interiors (Sellers et al., 1997). Thus, any ozone-induced changes in canopy ET will likely alter the hydrologic cycle in intercontinental areas with significant vegetation cover.

If growth at elevated  $[O_3]$  reduces ET, it would be predicted to slow depletion of soil moisture. This has the potential to feedback on plant performance by delaying the onset of stress during periods of drought (e.g., Bernacchi et al., 2007) and, in natural habitats, reduce the competitive fitness of the most ozone sensitive species. In addition, a large number of ecologically significant





biological and physical soil processes are influenced by soil water status, including litter decomposition, soil respiration, denitrification and invertebrate community composition. Therefore, to understand the full consequences of elevated [O<sub>3</sub>] for plant and ecosystem function, it is important to characterize changes in ET and soil water status.

The prediction that growth in elevated  $[O_3]$  would impact ET is based on observed physiological responses of plants under these conditions. Decreases in leaf-level stomatal conductance  $(g_s)$ resulting from growth in O<sub>3</sub>-enriched atmospheres are well documented (Morgan et al., 2003; Fiscus et al., 2005; Wittig et al., 2007), although this response was not evident in young soybean leaves (e.g., Bernacchi et al., 2006). Since g<sub>s</sub> and canopy ET are closely coupled in soybean (e.g., Bernacchi et al., 2007), O<sub>3</sub>-induced decreases in g<sub>s</sub> are expected to translate into proportional decreases in ET. However, canopy-scale control of ET might diminish the importance of g<sub>s</sub> (e.g., McNaughton and Jarvis, 1991), thereby minimizing the impact of decreased  $g_s$  in elevated  $[O_3]$ . A decrease in ET driven by  $g_s$  could be amplified in circumstances in which growth in O3-enriched atmospheres results in smaller plant canopies. Elevated [O<sub>3</sub>] can also decrease root development (Andersen, 2003), which could limit the amount of water that vegetation is able to acquire from the soil and thus act as an additional limitation to ET. While leaf control of  $g_s$  appears to slow when plants are grown in elevated  $[O_3]$ (Wilkinson and Davies, 2010), potentially making them more prone to damage during water stress, a lower rate of ET caused by elevated [O<sub>3</sub>] might lessen the frequency when these stress conditions occur since more moisture will be available in the soil profile.

Soybean (*Glycine max* (L.) Merr.) is one of the four most important agricultural species worldwide and, in annual rotation with corn (*Zea mays*), represents the largest ecosystem type in temperate North America. Changes in the water cycling by this expansive ecosystem are likely to have an important influence on Midwestern climate as current research suggests a strong coupling between biophysical processes, such as evapotranspiration, and regional climate. Estimates suggest that ca 35% of Central U.S. summer precipitation occurs as a result of transpiration from vegetation (Jiang et al., 2009). Modeled predictions suggest that reductions in  $g_s$  resulting from increases in atmospheric CO<sub>2</sub> from preindustrial concentrations to current levels have already decreased ET to the point that runoff into major rivers in the U.S. are higher (Gedney et al., 2006; Betts et al., 2007).

The main objectives of this study were to assess in soybean the impact that rising ozone has on: (1) the major ecosystem energy fluxes over diurnal and seasonal time scales, (2) the seasonal crop water use, and (3) soil water content. This work was conducted on soybean grown in elevated [O3] at the Soybean Free Air gas Concentration Enrichment (SoyFACE) research facility. Soybean is one of the most widely studied crop species with regard to responses to elevated  $[O_3]$  (e.g., Morgan et al., 2003), however, the response of field-grown soybean canopy ET to growth in elevated [O<sub>3</sub>] is not known. We are not aware of any published data characterizing the response of ET for any field-grown plant to elevated [O<sub>3</sub>]. Using the residual energy balance approach, we present estimates of ET along with soil water content measurements for soybean grown under ambient 'background' [O<sub>3</sub>] and elevated (ambient plus  $\sim 25\%$ ) [O<sub>3</sub>] from five consecutive growing seasons (2002–2006) representing a range of environmental conditions.

## 2. Methods

#### 2.1. Site description

The SoyFACE facility is situated in a 32 ha (80 acre) field at the University of Illinois at Urbana-Champaign (40°03'21.3"N, 88°12'3.4"W, 230 m elevation). Soybean (*G. max* (L.) Merr. cv. Pioneer 93B15) and maize (*Z. mays*) each occupied half

of the field. The agricultural practices at SoyFACE followed those typical for Illinois rain-fed agriculture, including yearly crop rotations, and have been described previously (Ainsworth et al., 2004; Rogers et al., 2004; Bernacchi et al., 2006). A complete description of the SoyFACE experiment is given elsewhere (Rogers et al., 2004; Bernacchi et al., 2005; Morgan et al., 2005).

The experimental design consisted of four replicated plots (n of 4) for the control and elevated ozone treatment. All plots were instrumented with micrometeorological sensors to measure sensible and soil heat fluxes and net radiation, with the exception of 2002 when only 3 of the 4 blocks were instrumented. The target concentration for the elevated  $[O_3]$  plots was an instantaneous increase of 50% above ambient concentrations, which were measured at a single location at the west end of the SoyFACE facility. Background 'control' [O3] was monitored and increases in the elevated [O<sub>3</sub>] plots were targeted as a set value above the control. Concentrations of ozone in the control and elevated  $[O_3]$  plots were monitored continuously throughout the season (model 49C O3 analyzer; Thermo Environmental Instruments, Franklin, MA, USA; Morgan et al., 2004). The ozone analyzers were calibrated yearly (calibration USA EPA Equivalent Method EQQA-0880-047, range 0-0.05-1.0 ppm; Morgan et al., 2006). Fumigation occurred during daylight hours and only when the leaves were dry, which resulted in seasonal increases in daily 8hr average ozone of between 22 and 37%, depending on plot and season. The variability in ozone fumigation is consistent with high temporal variability of the ambient concentrations and the increases in ozone concentrations were based on projected future mean tropospheric increases predicted for 2050 (Prather et al., 2001, 2003). Daily 8 h mean [O<sub>3</sub>] along with the AOT40 (exposure accumulated above 40 ppb) and SUM06 (exposure accumulated when fluxes are greater than 60 ppb) O<sub>3</sub> flux indices (Mauzerall and Wang, 2001) are presented in Fig. 1.

#### 2.2. Micrometeorological measurements

A residual energy balance approach was used to determine ET based on measurements of sensible heat flux, soil heat flux and net radiation from individual plots as described previously for control and elevated CO<sub>2</sub> plots at SOyFACE (Bernacchi et al., 2006) using methods developed and validated previously (Huband and Monteith, 1986; Jackson et al., 1987; Kimball et al., 1994, 1995, 1999; Triggs et al., 2004). Values of ET are solved according to the energy balance equation:

$$\lambda ET = R_n - G_0 - H \tag{1}$$

where  $\lambda$  is latent heat of vaporization (J kg<sup>-1</sup>), ET is evapotranspiration (kg m<sup>-2</sup> s<sup>-1</sup>; positive upward),  $R_n$  is net radiation (W m<sup>-2</sup>, positive downward),  $G_0$  is soil surface heat flux (W m<sup>-2</sup>, positive downward), and *H* is sensible heat flux (W m<sup>-2</sup>, positive upward).

The residual energy balance approach to determine ET is effective in obtaining quantitative estimates of ET (Kimball et al., 1999) and is the only available technique suitable for the scale of FACE experiments that does not include enclosing portions of a plant canopy in a chamber. Techniques such as eddy covariance or flux gradient analysis, while more precise (Baldocchi et al., 1988), require a substantially larger fetch than can be achieved within the constraints of FACE technology (Kimball et al., 1999; Triggs et al., 2004; Bernacchi et al., 2007). While a full energy audit would include energy fluxes not represented in Eq. (1), e.g., photosynthesis, respiration and heat storage within the canopy, these additional fluxes represent a small component of incoming radiation and thus can be safely ignored (Meyers and Hollinger, 2004).

Three of four control and elevated ozone plots in year 2002 and all four plots in years 2003–2006 contained a micrometeorological station equipped to measure each of the three major energy flux terms on the right side of Eq. (1). Measurements were made in 10 s intervals and averaged over 10 min throughout the growing season from planting until harvest. A complete description of the experimental setup was provided previously (Bernacchi et al., 2007).

Net radiation  $(R_n)$  was measured using single-channel net radiometers (Models Q\*6 or Q\*7; Radiation and Energy Balance Systems (REBS), Inc., Seattle, WA USA), which were maintained at 1 m above the crop surface as the crop canopy grew. A cross-calibration was performed prior to, or immediately after, each growing season as described previously (Bernacchi et al., 2007). The replication of soil heat flux measurements differed throughout the experiment depending on the number of instruments available. These measurements were collected in one block in 2002 and three blocks in 2003-2006. Soil heat flux plates (Model HFT-3, REBS, Inc.) were buried at 10 mm depths between and within planting rows. The total number of heat flux plates in each replicate was four in 2002 and two in 2003–2006 and they were arranged perpendicular to the direction of planting such that equal numbers of measurements were made within and between rows. During the 2002 growing season only one replicate plot each for the control and elevated [O<sub>3</sub>] included measurements of  $G_0$ . Measurements of  $G_0$  also included the heat storage in soil above each heat flux plate as previously described (Kimball et al., 1994). Total soil heat flux represented <7% of seasonal  $R_n$ .

Sensible heat flux determination relied on a number of sensors and was calculated as:

$$H = \rho_a c_p \frac{T_s - T_a}{r_a} \tag{2}$$



**Fig. 1.** (Top panels) Ozone treatment for the 2002–2006 growing seasons at the SoyFACE Facility showing daily 8 h mean ozone concentration; the black line shows the ambient treatment and the grey line the elevated  $O_3$  treatment. (Middle Panels) AOT40 (solid lines) and SUM06 (dashed lines) ozone index, calculated according to Mauzerall and Wang (2001). (Bottom panels) Daily maximum and minimum temperatures (black lines), vapor pressure deficit (grey lines), and daily total precipitation (bars) for the 2002–2006 growing seasons.

where  $\rho_a$  is air density (kg m<sup>-3</sup>),  $C_p$  the heat capacity of air (I kg<sup>-1</sup> °C<sup>-1</sup>),  $T_s$  and  $T_a$ the surface and air temperatures (°C), and  $r_a$  the aerodynamic resistance (s m<sup>-1</sup>). For the 2002-2005 growing seasons, air temperature was measured using custom-built aspirated psychrometers (Peresta et al., 1991) and in 2006 measurements were made using a thermistor (Model 107, Campbell Scientific, Inc.) with a custom aspirated heat shield employing PVC pipe, an axial fan, and aluminum tape. Surface temperatures, T<sub>s</sub>, were measured using infrared thermometers (IRT; IRT-P, Apogee Instruments, Inc., Logan, UT, USA), which were calibrated before each growing season (Triggs et al., 2004). Aerodynamic resistance was calculated based on a previously described model (lackson et al., 1987; Kimball et al., 1994, 1999; Triggs et al., 2004). This model relies on wind speed (Model 12102D, R.M. Young Company, Traverse City, Michigan, USA), Ta, Ts, dew point temperatures  $(T_d)$ , and canopy height. Canopy height was measured in weekly intervals throughout each season. Upon the completion of each growing season, height data fitted to an equation representing the sigmoidal increase in height throughout the season was incorporated into the appropriate equations. Meteorological measurements were collected at the SoyFACE facility as described previously (Bernacchi et al., 2007).

#### 2.3. Soil moisture

Access tubes were installed to allow measurement of soil moisture content  $(H_2O_{v/v\%})$  with a capacitance probe (Diviner-2000, Sentek Sensor Technologies) when soybean was grown on the eastern half of the field site in 2004 and 2006. Within each plot, two access tubes were positioned within crop rows and two access tubes were positioned between crop rows. Measurements were taken in 10-cm increments between depths of 5 and 105 cm on 19 dates in 2004 and 28 dates in 2006. Raw data from the probe were calibrated against gravimetric data using the method of Paltineanu and Starr (1997).

#### 2.4. Data analysis

#### 2.4.1. Seasonal patterns of micrometeorology

The 10-min data were averaged over each diurnal time period to provide daily mean values for each of the four fluxes (W  $m^{-2}$ ). These data were analyzed using

a complete block analysis of variance with day of year as the repeated measure, treatment as a main effect and block as a random factor (Mixed Procedure, SAS 9.1, The SAS Institute, Raleigh, NC, USA). The analyses were conducted separately for each year because of the annual rotation of the experiment to different sides of the field.

### 2.4.2. Diurnal patterns of micrometeorology

Within a growing season, the 10-min data were statistically analyzed using a repeated measures analysis of variance with block as random factors, treatment as a main effect and time of day as a repeated measure. The goal of this analysis was to determine the effect of ozone on the fluxes throughout the 24-hour time course so each measurement date within a growing season was treated as a subsample for each replicate plot. Therefore, all the data from each 10-min recorded time period throughout the 24-hour time course were averaged across each growing season. Thus, each growing season yielded an *n* of 4 (*n* of 3 in 2002) for the control and elevated  $[O_3]$  plots for each 10-min time period throughout the day.

#### 2.4.3. Seasonal mean values and water use

The daily means of each of the fluxes were calculated and the mean  $\lambda$ ET were accumulated over each growing season and converted to total crop water use (mm season<sup>-1</sup>) using the latent heat of vaporization for water. This analysis yielded one seasonal mean value for each of the four fluxes and total seasonal crop water use for each replicate plot (n = 3 in 2002 and n = 4 in 2003–2006). Differences for the mean fluxes and total season water use between control and elevated [O<sub>3</sub>] plots were tested using a complete block analysis of variance with treatment and year as main effects and block as a random factor. Since the number of measurement days varied for each season, the total crop water use for these comparisons was normalized to the mean number of measurement days across all four growing seasons, which worked out to be 76 days.

### 2.4.4. Soil moisture content

Average values of soil  $H_2O_{V/\%}$  were calculated for three soil layers (5–25 cm, 25–55 cm, 55–105 cm) in each plot. For each growing season and soil layer, these data were analyzed using a complete block analysis of covariance with day of year as the repeated measure, ozone treatment as a main effect, block as a random factor

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and saturated soil  $H_2O_{V/V\%}$  in each plot at the beginning of the growing season as the covariate (Mixed Procedure, SAS 9.1, The SAS Institute, Raleigh, NC, USA). Treatment effects on specific measurement dates were assessed with pairwise comparisons using the pdiff option in the Mixed Procedure.

# 3. Results

Based on the seasonal average eight-hour maximums, soybeans were exposed to between a low of 22% (2002 growing season) and a high of 37% (2006 growing season) greater [O<sub>3</sub>] in the elevated [O<sub>3</sub>] plots compared with the control plots (Table 1). Whereas the mean 8-hour maximum concentrations show fluctuations from day to day, the AOT40 and Sum06 ozone indices reflect the cumulative exposure to ozone throughout the growing season for the ambient and elevated [O<sub>3</sub>] plots (Fig. 1), and are presented here to allow the treatment effects on ET and soil moisture to be related to previous studies as well as air quality standards in North America and Europe. Based on both indices, the 2002 season experienced the highest ozone concentrations and the 2004 and 2006 growing seasons the lowest (Fig. 1; Table 1).

Meteorological conditions varied across the five growing seasons (Fig. 1) although none of the growing seasons deviated strongly from typical conditions for the region. The Palmer Crop Moisture Index (PCMI) showed that 2005 was the only growing season that had persistently dry conditions as indicated by PCMI values lower than -1 (Fig. 2). The only other year that experienced dry conditions was 2002 when the PCMI dropped below -1 for one week during midseason.

Two measurement days were selected to illustrate micrometeorological fluxes for a clear (day of year 234, 2004) and an overcast (day of year 244, 2003) day (Fig. 3). There were small but noticeable differences between the control and elevated  $[O_3]$  plots for  $R_n$ (Fig. 3). Differences between control and elevated  $[O_3]$  plots were more pronounced for *H* and ET, although the response to elevated  $[O_3]$  was more consistent on the clear compared with the overcast day (Fig. 3). The differences in  $G_0$  between the control and elevated  $[O_3]$  plots were negligible.

#### Table 1

Mean seasonal values for three ozone indices for the control and elevated ozone plots at SoyFACE.

Year	8-hour maximum (ppb)		% of days with	% Increase with				
	Control	Elevated	[O <sub>3</sub> ] fumigation <sup>a</sup>	fumigation				
2002	$55.3\pm0.84$	$\textbf{67.9} \pm \textbf{0.84}$	64.1%	37.0%				
2003	$45.5\pm0.73$	$56.9\pm0.73$	71.8%	33.2%				
2004	$\textbf{37.2} \pm \textbf{0.73}$	$46.1 \pm 0.76$	65.9%	36.2%				
2005	$44.3 \pm 0.73$	$59\pm0.73$	71.6%	42.0%				
2006	$\textbf{35.4} \pm \textbf{0.73}$	$\textbf{48.6} \pm \textbf{0.73}$	83.3%	41.6%				
Year	AOT40 (ppmh)							
			Elevated					
2002		35.3 ± 0.	14	$64.7\pm0.14$				
2003		$22.2\pm0.$	12	$50.8\pm0.12$				
2004		$3.3 \pm 0.$	12	$25\pm0.12$				
2005		$44\pm0.12$						
2006		12	$26.8\pm0.12$					
Year	Sum06 (ppmh)							
		Control		Elevated				
2002		$21.4 \pm 0.$	07	$38.6\pm0.07$				
2003		$13.9\pm0.$	06	$29.2\pm0.06$				
2004		$5.2 \pm 0.$	06	$14.8\pm0.06$				
2005		$10.4 \pm 0.$	06	$26\pm0.06$				
2006		$4\pm 0.$	06	$16\pm0.06$				

 $^{a}\,$  Represents the percentage of days when 8-hour maximum  $\left[ O_{3}\right]$  was at least 20% greater than control.

 $^{\rm b}$  Represents the mean percentage increase in 8-hour maximum  $\left[O_3\right]$  for these days.



**Fig. 2.** Weekly mean Palmer Crop Moisture Index (PCMI; Palmer, 1968) and the mean 30-yr average PCMI value (dotted line) for Illinois Climate Division 5, which includes SoyFACE.

Mean seasonal values for each of the four fluxes were computed for each 10-min time point throughout the day (Fig. 4). All fluxes showed statistically significant differences (p < 0.05) for the treatment by time of day interaction. The differences between control and elevated  $[O_3]$  for  $R_n$  and for  $G_0$  occurred during daylight hours, showed interannual variation over the five growing seasons. and were relatively small compared with the treatment effect observed with H and ET (Fig. 4). The 2004 and 2006 growing seasons showed higher  $R_n$  for the first part of the day for the control relative to the elevated  $[O_3]$  treatments (Fig. 4). The elevated  $[O_3]$ plots had consistently higher H over all growing seasons and, as expected, the differences were more pronounced during daylight hours except for 2004 when the differences in H between the control and elevated [O<sub>3</sub>] plots were consistent over the diel time course (Fig. 4). Four of the five years showed substantially higher H during daylight hours with peak differences of  $\sim 40 \text{ W/m}^2$  in 2002, 2003, and 2005 and  $\sim 20 \text{ W/m}^2$  in 2006. The difference in H between the control and elevated [O<sub>3</sub>] plots during the 2004 growing season was small.

Daily mean values for each of the four fluxes were computed over each growing season (Fig. 5) and, as with the data presented over the diurnal time course, the large dataset resulted in statistically significant differences between the control and elevated  $[O_3]$ plots for most fluxes over most years. The differences in seasonal means were generally less than ca 4.5 W/m<sup>2</sup> for either  $R_n$  or  $G_0$ (Table 2). Sensible heat fluxes were consistently higher in elevated [O<sub>3</sub>] plots throughout each of the growing seasons except 2004 (Fig. 5). Latent heat flux followed a pattern inverse to H over the growing season with the elevated  $[O_3]$  plots having lower values relative to the controls (Fig. 5). This difference, although variable from day to day, was consistent over the growing seasons (Table 2). Overall, the elevated  $[O_3]$  plots had significantly higher H and significantly lower  $\lambda$ ET when all years were analyzed together (Table 2). However, pairwise statistical comparisons shows that in 2004 there were no statistical differences in *H* or  $\lambda$ ET between the control and elevated [O<sub>3</sub>] plots. Computed total water use by the plant canopies was consistently higher for the control compared with the elevated [O<sub>3</sub>] plots when all years were statistically analyzed together (Table 2) but posterior pairwise analysis again showed no differences in total water use between the control and elevated [O<sub>3</sub>] plots in 2004. In all years excluding 2004, the reduction in total water use by the elevated [O<sub>3</sub>] grown plants varied with percent decreases ranging from11% in 2006 to ca 14% in 2003.



**Fig. 3.** Net radiation ( $R_n$ ), sensible heat flux (H), soil heat flux ( $G_0$ ), and latent heat flux ( $\lambda$ ET) for control (black circles) and elevated [CO2] (grey circles) over an example sunny (day of year 230, 2003; top panels) and overcast (day of year 244, 2003; bottom panels) day. Each symbol represents a mean of four replicate blocks. Error bars represent one standard error around the mean.

Canopy temperatures were consistently warmer over the entire diel time course for the canopies grown in elevated  $[O_3]$  (Fig. 6). For all growing seasons except 2004, the increase in canopy temperature between the control and elevated  $[O_3]$  plots was greatest

during the day. However, in 2004 the canopy temperature differences were greatest during the night and the canopy temperatures were substantially warmer at night relative to the other growing seasons.



**Fig. 4.** The average diurnal course of the absolute differences (elevated  $O_3$  minus control) between elevated  $[O_3]$  and control plots for  $R_n$ , H, Gand  $\lambda$ ET. Each point represents a 10-minute interval of the day averaged across all measurement days and three to four replicate blocks, depending on the year. Negative represents higher values in the control plots and positive higher in the elevated  $[O_3]$ . Error bars are as in Fig. 3.



**Fig. 5.** The mean daily energy fluxes for  $R_n$ , H, G and  $\lambda$ ET for the control and elevated O<sub>3</sub> plots over each of the five growing seasons. Each point represents the daily mean flux of three or four replicate plots, depending on the year. Symbols and error bars are as in Fig. 3.

There was no difference in soil  $H_2O_{v/v_{\infty}}$  between treatments at the beginning of either the 2004 or 2006 (the two years in which data was collected) growing season (Fig. 7A–F). Cycles of soil  $H_2O_{v/v_{\infty}}$  depletion due to crop water use and subsequent replenishment by rain were greatest in magnitude and frequency at shallow depths. In 2004, a significant period of soil drying occurred from DOY ~200 to 235. During this time soil moisture was depleted more slowly under soybean exposed to elevated [O<sub>3</sub>], resulting in significantly wetter soil at depths of 5–25 cm and 25–55 cm (Fig. 7A,B). At depths of 55–105 cm there was almost no depletion of soil  $H_2O_{v/v_{\infty}}$  by crop water use, and consequently no effect of elevated [O<sub>3</sub>] (Fig. 7C). In 2006, frequent and heavy rain meant that crop water use rarely led to significant soil drying and, as a consequence, the difference in ET at ambient and elevated  $[O_3]$  had no significant effect on soil  $H_2O_{V/v\%}$  (Fig. 7D–F).

# 4. Discussion

This study demonstrated that elevated  $[O_3]$  reduced ET for soybean grown under field conditions at the SoyFACE research facility for four of five growing seasons and that this can lead to less soil water depletion. The five growing seasons represented a wide range of meteorological and climatic conditions as well as years with relatively high (AOT40 > 35 ppmh) and low (AOT40 < 4 ppmh) ambient  $[O_3]$ . All five years had statistically significant differences in all measured energy fluxes; however, these statistical

Table 2

Mean seasonal values of the four major energy fluxes ( $W/m^2$ ) associated with soybean from control and elevated  $O_3$ . Also shown is the conversion of  $\lambda$ ET to amount of water used during the growing season (mm). Statistical results using ANOVA are shown at the bottom.

Year	Treatment	R <sub>n</sub>	G <sub>0</sub>	Н	λΕΤ	Water use
2002	Control	$151.9\pm0.9$	$8.5\pm0.8$	25.3 ± 1.1	$119.2 \pm 2.8$	$379.5 \pm 8.9$
	Elevated O <sub>3</sub>	$152.7\pm0.9$	$\textbf{8.5}\pm\textbf{0.8}$	$40.5 \pm 1.1$	$104.6 \pm 2.8$	$\textbf{333.2} \pm \textbf{8.9}$
2003	Control	$145.6\pm2.2$	$4.4\pm0.7$	$\textbf{45.2} \pm \textbf{3.0}$	$96.0\pm2.3$	$305.6\pm7.2$
	Elevated O <sub>3</sub>	$145.0\pm2.2$	$\textbf{6.9} \pm \textbf{0.7}$	$55.4 \pm 3.0$	$\textbf{82.8} \pm \textbf{2.3}$	$263.5\pm7.2$
2004	Control	$137.3\pm1.2$	$\textbf{3.2}\pm\textbf{0.8}$	$29.9 \pm 4.4$	$104.3\pm4.0$	$332.1\pm12.9$
	Elevated O <sub>3</sub>	$135.6\pm1.0$	$2.2\pm0.7$	$30\pm3.8$	$103.5\pm3.5$	$329.5\pm11.1$
2005	Control	$145.7\pm1.4$	$\textbf{7.8} \pm \textbf{0.7}$	$20.5\pm5.2$	$117.3\pm5.7$	$\textbf{373.7} \pm \textbf{18.1}$
	Elevated O <sub>3</sub>	$146.2\pm1.4$	$\textbf{6.9} \pm \textbf{0.7}$	$37.6\pm5.2$	$101.7\pm5.7$	$\textbf{323.9} \pm \textbf{18.1}$
2006	Control	$152.0\pm 6.8$	$5.3\pm0.8$	$20.6\pm9.1$	$124.9\pm 6.5$	$397.6\pm20.5$
	Elevated O <sub>3</sub>	$141.4\pm6.8$	$\textbf{3.9} \pm \textbf{0.8}$	$\textbf{26.7} \pm \textbf{9.1}$	$111.0\pm6.5$	$353.5\pm20.6$
Year		**	**	**	**	**
Treatment		-	_	**	**	**
Year*Treatment		-	*	-	-	-

 $\overline{p} < 0.05$ , \*\*p < 0.01.



**Fig. 6.** The mean diurnal course of the absolute differences between elevated  $[O_3]$  and control plots for canopy temperature for the time period in each growing season when the canopies were completely closed but prior to the onset of senescence (DOY 190–240). Each line represents the mean of three or four replicates, depending on year. Standard errors are not graphed, but range from 0.21 °C to 0.32 °C depending on year.

differences are driven, in many cases, by the large number measurements included in the statistical model.

When the energy flux into ET was summed throughout each growing season to calculate seasonal water use, it was significantly lower under elevated  $[O_3]$  for four growing seasons (12% in 2002,

14% in 2003, 13% in 2005, 11% in 2006; Table 2). While there was no significant difference between the control and elevated  $[O_3]$ treatment in season mean H, ET, or water use in 2004, this season started off with higher ET in elevated [O<sub>3</sub>] for the first three weeks followed by a decline in water use for the remainder of the season (Fig. 5). In 2004, soil water content was significantly greater under elevated  $[O_3]$  than ambient  $[O_3]$  in the upper and middle layers of the soil profile (Fig. 7) during the period when elevated  $[O_3]$ showed lower ET relative to the control. This is important for two reasons. First, it corroborates the evidence from the micrometeorological data indicating growth of soybean at elevated [O<sub>3</sub>] reduces ET. Second, it demonstrates that reductions in ET at elevated  $[O_3]$ can potentially feedback to impact crop drought stress as well as soil processes that are regulated by soil moisture. This suggests that elevated [O<sub>3</sub>] induced changes in ET may need to be accurately simulated in models of plant and ecosystem function if they are to effectively predict future changes in important fluxes of carbon and water

The impact of reduced ET at elevated  $[O_3]$  on soil moisture was strongly dependent on the pattern of precipitation. Longer periods of low precipitation allowed small treatment effects on ET to accumulate into significant treatment effects on soil  $H_2O_{V/v}$ . The 2004 growing season exemplifies this scenario and also shows that within-season, temporal variation in ET under elevated  $[O_3]$  can be important even when there is no change in season-long water use. In contrast, when frequent rain events occurred, significant treatment effects on ET did not translate into significantly altered  $H_2O_{V/v}$ . The 2006 growing season exemplifies this scenario. While  $H_2O_{V/v}$ .



**Fig. 7.** Soil H<sub>2</sub>O<sub>V/V<sup>±</sup></sub> at depths of 5–25 cm (A,D), 25–55 cm (B,E) and 55–105 cm (C,F) in plots of *G. max* growing under ambient (black symbols) and elevated O<sub>3</sub> (grey symbols) during the 2004 (A,B,C) and 2006 (D,E,F) growing seasons at SoyFACE. Each point is the mean ( $\pm$ SE) of the replicate plots measured at that time (n = 4). p-values indicate statistical significance of treatment effects in ANOVA. Statistically significant effects from pairwise comparison of treatments on a single date indicated by \*p < 0.1, \*\*p < 0.05, \*\*\*p < 0.01, \*\*\*p < 0.001.

was not altered by elevated [O<sub>3</sub>], mass balance dictates that the change in plant water use must drive changes in drainage and runoff under such meteorological conditions.

The measurements collected in this study show that increasing ozone concentrations beyond a threshold level will feedback on vegetation to induce a decrease in water use. This study, however, does not resolve the physiological mechanisms that induce this response. It is likely that a large component that drives the decrease in ET stems from a reduction in  $g_s$ , even though the reduction in  $g_s$ may be limited to older leaves in the canopy (Morgan et al., 2003; Bernacchi et al., 2006). Beyond the effect that elevated  $[O_3]$  has on water use due to reducing g<sub>s</sub>, elevated [O<sub>3</sub>] also decreases LAI in some years (Dermody et al., 2006), which will enhance decreases in ET. Elevated  $[O_3]$  has been shown to decrease root growth in many species (e.g., Andersen, 2003) including soybean (Blum and Tingey, 1977); indeed the impact of elevated  $[O_3]$  on plants may be more severe in roots than in above-ground organs (Andersen, 2003). Together, decreases in total biomass probably contribute to the lower water use by plants grown at elevated  $[O_3]$ .

The observed decline in water use associated with growth at elevated  $[O_3]$  is demonstrated by the higher canopy temperatures relative to the control. Interestingly, every year showed higher day-time as well as night-time canopy temperatures (Fig. 6). The effect of higher canopy temperatures at night have little or no impact on ET as under night-time conditions canopy conductance approaches zero (data not shown). However, this nocturnal warming could be driven by a combination of thinner canopies associated with growth in elevated  $[O_3]$  coupled with diaheliotropic leaf movements exhibited by soybean. Thus, at night it is likely that more of the warmer soil surface is observed by the infrared radiometers used to measure surface temperature.

Many factors will influence the rate in which water is evapotranspired by plant canopies and there exists a large degree of variability from year-to-year. The results from this study show that under most circumstances, the addition of  $[O_3]$  in plant canopies reduces ET. However, background [O<sub>3</sub>] varied from year-to-year (Fig. 1; Table 1) and it is likely that this variability is already influencing rates of ET. For example, it is shown that [O<sub>3</sub>] is already causing reduced yields in some crop species (Bergmann et al., 1999; Feng et al., 2003, 2011; Morgan et al., 2003; Timonen et al., 2004; Fiscus et al., 2005; Karlsson et al., 2005; Wang et al., 2007; Ainsworth, 2008; Betzelberger et al., 2010; Fishman et al., 2010). Given the high variability of climate from year-to-year, it is difficult to determine how much current rates of water use might be influenced by ambient [O<sub>3</sub>], but future analyses focusing on withinyear response surface ozone experiments or using ecosystem modeling coupled with known physiological parameterizations could help to resolve this.

Soybean, together with maize, makes up the largest continuous ecosystem type in temperate North America. This is the first experiment to show that growth in elevated [O<sub>3</sub>] under open-air conditions results in a decrease in ET. As described previously (e.g., Sellers et al., 1997), the impact of atmospheric change on the partitioning of energy between H and ET for soybean is likely to have consequences in regional climate and hydrology. In 2002, 2003 and 2005 elevated [O<sub>3</sub>] resulted in a greater than 12% decrease in seasonal water use by soybean and averaged over the 5 years of this experiment seasonal water use was 10% lower at elevated  $[O_3]$ . These results suggest that future increases in [O<sub>3</sub>] could lead to alterations in the local and regional hydrologic cycles where soybean, and perhaps other major crops, is grown. Since increases in *H* are likely to lead to higher air temperatures (e.g., Georgescu et al., 2009), the impact of elevated [O<sub>3</sub>] on soybean would augment the warming air temperatures caused by higher concentrations of greenhouse gases.

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## References

- Ainsworth, E.A., 2008. Rice production in a changing climate: a meta-analysis of responses to elevated carbon dioxide and elevated ozone concentration. Global Change Biology 14, 1642–1650.
- Ainsworth, E.A., Rogers, A., Nelson, R.L., Long, S.P., 2004. Testing the "source-sink" hypothesis of down-regulation of photosynthesis in elevated [CO<sub>2</sub>] in the field with single gene substitutions in glycine max. Agricultural and Forest Meteorology 122, 85–94.
- Andersen, C.P., 2003. Source-sink balance and carbon allocation below ground in plants exposed to ozone. New Phytologist 157, 213–228.
- Baldocchi, D.D., Hicks, B.B., Meyers, T.P., 1988. Measuring biosphere-atmosphere exchanges of biologically related gases with micrometeorological methods. Ecology 69, 1331–1340.
- Bergmann, E., Bender, J., Weigel, H.J., 1999. Ozone threshold doses and exposureresponse relationships for the development of ozone injury symptoms in wild plant species. New Phytologist 144, 423–435.
- Bernacchi, C.J., Morgan, P.B., Ort, D.R., Long, S.P., 2005. The growth of soybean under free air [CO<sub>2</sub>] enrichment (FACE) stimulates photosynthesis while decreasing in vivo Rubisco capacity. Planta 220, 434–446.
- Bernacchi, C.J., Leakey, A.D.B., Heady, L.E., Morgan, P.B., Dohleman, F.J., McGrath, J.M., Gillespie, K.M., Wittig, V.E., Rogers, A., Long, S.P., Ort, D.R., 2006. Hourly and seasonal variation in photosynthesis and stomatal conductance of soybean grown at future CO<sub>2</sub> and ozone concentrations for 3 years under fully open-air field conditions. Plant Cell and Environment 29, 2077–2090.
- Bernacchi, C.J., Kimball, B.A., Quarles, D.R., Long, S.P., Ort, D.R., 2007. Decreases in stomatal conductance of soybean under open-air elevation of [CO<sub>2</sub>] are closely coupled with decreases in ecosystem evapotranspiration. Plant Physiology 143, 134–144.
- Betts, R.A., Boucher, O., Collins, M., Cox, P.M., Falloon, P.D., Gedney, N., Hemming, D.L., Huntingford, C., Jones, C.D., Sexton, D., Webb, M.J., 2007. Projected increase in continental runoff due to plant responses to increasing carbon dioxide. Nature 448, 1037–1041.
- Betzelberger, A.M., Gillespie, K.M., McGrath, J.M., Koester, R.P., Nelson, R.L., Ainsworth, E.A., 2010. Effects of chronic elevated ozone concentration on antioxidant capacity, photosynthesis and seed yield of 10 soybean cultivars. Plant, Cell and Environment 33 (9), 1569–1581.
- Blum, U., Tingey, D.T., 1977. A study of the potential ways in which ozone could reduce root growth and nodulation of soybean. Atmospheric Environment 11, 737–739.
- Dermody, O., Long, S.P., DeLucia, E.H., 2006. How does elevated CO<sub>2</sub> or ozone affect the leaf-area index of soybean when applied independently? New Phytologist 169, 145–155.
- Feng, Z., Jin, M., Zhang, F., Huang, Y., 2003. Effects of ground-level ozone (O<sub>3</sub>) pollution on the yields of rice and winter wheat in the Yangtze River Delta. Journal of Environmental Sciences-China 15, 360–362.
- Feng, Z., Pang, J., Kobayashi, K., Zhu, J., Ort, D.R., 2011. Differential responses in two varieties of winter wheat to elevated ozone concentration under fully open-air conditions. Global Change Biology 17, 580–591.
- Fiscus, E.L., Reid, C.D., Miller, J.E., Heagle, A.S., 1997. Elevated CO<sub>2</sub> reduces O<sub>3</sub> flux and O<sub>3</sub>-induced yield losses in soybeans: possible implications for elevated CO<sub>2</sub> studies. Journal of Experimental Botany 48, 307–313.
- Fiscus, E.L., Booker, F.L., Burkey, K.O., 2005. Crop responses to ozone: uptake, modes of action, carbon assimilation and partitioning. Plant Cell and Environment 28, 997–1011.
- Fishman, J., Creilson, J.K., Parker, P.A., Ainsworth, E.A., Vining, G.G., Szarka, J., Booker, F.L., Xu, X., 2010. An investigation of widespread ozone damage to the soybean crop in the upper midwest determined from ground-based and satellite measurements. Atmospheric Environment 44, 2248–2256.
- Fowler, D., Cape, J.N., Coyle, M., Smith, R.I., Hjellbrekke, A.-G., Simpson, D., Derwent, R.G., Johnson, C.E., 1999. Modelling photochemical oxidant formation, transport, deposition and exposure of terrestrial ecosystems. Environmental Pollution 100, 43–55.
- Gedney, N., Cox, P.M., Betts, R.A., Boucher, O., Huntingford, C., Stott, P., 2006. Detection of a direct carbon dioxide effect in continental river runoff records. Nature 439, 835–838.
- Georgescu, M., Lobell, D.B., Field, C.B., 2009. Potential impact of U.S. biofuels on regional climate. Geophysical Research Letters 36, L21806.
- Heagle, A.S., 1989. Ozone and crop yield. Annual Review of Phytopathology 27, 397–423.
- Huband, N.D.S., Monteith, J.L., 1986. Radiative surface-temperature and energybalance of a wheat canopy. 2. Estimating fluxes of sensible and latent-heat. Boundary-Layer Meteorology 36, 107–116.
- IPCC, 2007. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. In: Solomon, S., Qin, D., Manning, M., Chen, Z., Marquis, M., Averyt, K.B., Tignor, M., Miller, H.L. (Eds.),

Climate Change 2007: The Physical Science Basis. Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA 996 p.

- Jackson, R.D., Moran, M.S., Gay, L.W., Raymond, L.H., 1987. Evaluating evaporation from field crops using airborne radiometry and ground-based meteorological data. Irrigation Science 8, 81–90.
- Jiang, X., Niu, G.Y., Yang, Z.L., 2009. Impacts of vegetation and groundwater dynamics on warm season precipitation over the Central United States. Journal of Geophysical Research 114, D06109. doi:10.1029/2008JD010756.
- Karlsson, P.E., Pleijel, H., Belhaj, M., Danielsson, H., Dahlin, B., Andersson, M., Hansson, M., Munthe, J., Grennfelt, P., 2005. Economic assessment of the negative impacts of ozone on crop yields and forest production. A case study of the estate OstadsSateri in southwestern Sweden. AMBIO 34, 32–40.
- Kimball, B.A., Lamorte, R.L., Seay, R., Pinter, P.J., Rokey, R., Hunsaker, D.J., Dugas, W.A., Heuer, M.L., Mauney, J., Hendrey, G.R., Lewin, K.F., Nagy, J., 1994. Effects of freeair CO<sub>2</sub> enrichment on energy-balance and evapotranspiration of cotton. Agricultural and Forest Meteorology 70, 259–278.
- Kimball, B.A., Pinter, P.J., Garcia, R.L., LaMorte, R.L., Wall, G.W., Hunsaker, D.J., Wechsung, G., Wechsung, F., Kartschall, T., 1995. Productivity and water use of wheat under free-air CO<sub>2</sub> enrichment. Global Change Biology 1, 429–442.
  Kimball, B.A., LaMorte, R.L., Pinter, P.J., Wall, G.W., Hunsaker, D.J., Adamsen, F.J.,
- Kimball, B.A., LaMorte, R.L., Pinter, P.J., Wall, G.W., Hunsaker, D.J., Adamsen, F.J., Leavitt, S.W., Thompson, T.L., Matthias, A.W., Brooks, T.J., 1999. Free-air CO<sub>2</sub> enrichment and soil nitrogen effects on energy balance and evapotranspiration of wheat. Water Resources Research 35, 1179–1190.
- Mauzerall, D.L., Wang, X., 2001. Protecting agricultural crops from the effects of tropospheric ozone exposure: reconciling science and standard setting in the United States, Europe, and Asia. Annual Review of Energy and the Environment 26, 237–268. McNaughton, K.G., Jarvis, P., 1991. Effects of spatial scale on stomatal control of
- transpiration. Agricultural and Forest Meteorology 54, 279–301.
- Meyers, T.P., Hollinger, S.E., 2004. An assessment of storage terms in the surface energy balance of maize and soybean. Agricultural and Forest Meteorology 125, 105–115.
- Morgan, P.B., Ainsworth, E.A., Long, S.P., 2003. How does elevated ozone impact soybean? A meta-analysis of photosynthesis, growth and yield. Plant Cell and Environment 26, 1317–1328.
- Morgan, P.B., Bernacchi, C.J., Ort, D.R., Long, S.P., 2004. An in vivo analysis of the effect of season-long open-air elevation of ozone to anticipated 2050 levels on photosynthesis in soybean. Plant Physiology 135, 2348–2357.
- Morgan, P.B., Bollero, G.A., Nelson, R.L., Dohleman, F.G., Long, S.P., 2005. Smaller than predicted increase in aboveground net primary production and yield of field-grown soybean under fully open-air [CO<sub>2</sub>] elevation. Global Change Biology 11, 1856–1865.
- Morgan, P.B., Mies, T.A., Bollero, G.A., Nelson, R.L., Long, S.P., 2006. Season-long elevation of ozone concentration to projected 2050 levels under fully open-air conditions substantially decreases the growth and production of soybean. New Phytologist 170, 333–343.
- Paltineanu, I.C., Starr, J.L., 1997. Real-time soil water dynamics using multisensor capacitance probes: laboratory calibration. Soil Science Society of America Journal 61, 1576–1585.

- Peresta, G.J., Kimball, B.A., Johnson, S.M., 1991. WCL Report 18-Procedures for CO<sub>2</sub>-Enrichment Chamber Construction and Data Acquisition and Analysis. United States Water Conservation Laboratory, Pheonix, AZ, USA.
- Prather, M., Ehhalt, D., Dentener, F., Derwent, R., Dlugokencky, E., Holland, E., Isaksen, I., Katima, J., Kirchhoff, V., Matson, P., Midgley, P., Wang, M., 2001. Atmospheric chemistry and greenhouse gases. In: Houghton, J.T., Ding, Y., Griggs, D.J., Noguer, M., van der Linden, P.J., Dai, X., Maskell, K., Johnson, C.A. (Eds.), Climate Change 2001: The Scientific Basis. Cambridge University Press, Cambridge, UK, pp. 239–287.
- Prather, M., Gauss, M., Berntsen, T., Isaksen, I., Sundet, J., Bey, I., Brasseur, G., Dentener, F., Derwent, R., Stevenson, D., Grenfell, L., Hauglustaine, D., Horowitz, L., Jacob, D., Mickley, L., Lawrence, M., von Kuhlmann, R., Muller, J., Pitari, G., Rogers, H., Johnson, M., Pyle, J., Law, K., van Weele, M., Wild, O., 2003. Fresh air in the 21st century? Geophysical Research Letters 30 (2), 1100. doi:10.1029/2002GL016285.
- Rogers, A., Allen, D.J., Davey, P.A., Morgan, P.B., Ainsworth, E.A., Bernacchi, C.J., Cornic, G., Dermody, O., Dohleman, F.G., Heaton, E.A., Mahoney, J., Zhu, X.G., Delucia, E.H., Ort, D.R., Long, S.P., 2004. Leaf photosynthesis and carbohydrate dynamics of soybeans grown throughout their life-cycle under free-air carbon dioxide enrichment. Plant Cell and Environment 27, 449–458.
- Sellers, P.J., Dickinson, R.E., Randall, D.A., Betts, A.K., Hall, F.G., Berry, J.A., Collatz, G.J., Denning, A.S., Mooney, H.A., Nobre, C.A., Sato, N., Field, C.B., Henderson-Sellers, A., 1997. Modeling the exchanges of energy, water, and carbon between continents and the atmosphere. Science 275, 502–509.
- Sitch, S., Cox, P.M., Collins, W.J., Huntingford, C., 2007. Indirect radiative forcing of climate change through ozone effects on the land carbon sink. Nature 448, 791.
- Timonen, U., Huttunen, S., Manninen, S., 2004. Ozone sensitivity of wild field layer plant species of northern Europe. A review. Plant Ecology 172, 27–39.
- Triggs, J., Kimball, B.A., Pinter, P.J., Wall, G.W., Conley, M.M., Brooks, T.J., LaMorte, R.L., Adam, N.R., Ottman, M.J., Matthias, A.D., Leavitt, S.W., Cerveny, R.S., 2004. Free-air CO<sub>2</sub> enrichment effects on the energy balance and evapotranspiration of sorghum. Agricultural and Forest Meteorology 124, 63–79.
- U.S. EPA, 2006. Air Quality Criteria for Ozone and Related Photochemical Oxidants. U.S. Environmental Protection Agency, Washington, DC. EPA/600/R-05/004aFcF, 2006.
- Van Dingenen, R., Dentener, F.J., Raes, F., Krol, M.C., Emberson, L., Cogala, J., 2008. The global impact of ozone on agricultural crop yields under current and future air. Atmospheric Environment 43, 604–618.
- Wang, X., Manning, W., Feng, Z., Zhu, Y., 2007. Ground-level ozone in China:
- distribution and effects on crop yields. Environmental Pollution 147, 394–400. Wilkinson, S., Davies, W.J., 2010. Drought, ozone, ABA and ethylene: new insights from cell to plant to community. Plant Cell and Environment 33 (4),
- 510–525. Wittig, V.E., Ainsworth, E.A., Long, S.P., 2007. To what extent do current and projected increases in surface ozone affect photosynthesis and stomatal conductance of trees? A meta-analytic review of the last 3 decades of experiments. Plant Cell and Environment 30, 1150–1162.