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Early vigour in wheat: could it lead to more severe terminal drought stress under elevated atmospheric [CO_2] and semi-arid conditions?

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Abstract

Early vigour in wheat is a trait that has received attention for its benefits reducing evaporation from the soil surface early in the season. However, with the growth enhancement common to crops grown under elevated atmospheric CO_2 concentrations (e[CO_2]), there is a risk that too much early growth might deplete soil water and lead to more severe terminal drought stress in environments where production relies on stored soil water content. If this is the case, the incorporation of such a trait in wheat breeding programs might have unintended negative consequences in the future, especially in dry years. We used selected data from cultivars with proven expression of high and low early vigour from the Australian Grains Free Air CO_2 Enrichment (AGFACE) facility, and complemented this analysis with simulation results from two crop growth models which differ in the modelling of leaf area development and crop water use. Grain yield responses to e[CO_2] were lower in the high early vigour group compared to the low early vigour group, and although these differences were not significant, they were corroborated by simulation model results. However, the simulated lower response with high early vigour lines was not caused by an earlier or greater depletion of soil water under e[CO_2] and the mechanisms responsible appear to be related to an earlier saturation of the radiation intercepted. Whether this is the case in the field needs to be further investigated. In addition, there was some evidence that the timing of the drought stress during crop growth influenced the effect of e[CO_2] regardless of the early vigour trait. There is a need for FACE investigations of the value of traits for drought adaptation to be conducted under more severe drought conditions and variable timing of drought stress, a risky but necessary endeavour.

Keywords: physiological pre-breeding, climate change adaptation, model intercomparison, *Triticum aestivum*, drought adaptive traits.

Abbreviations: atmospheric carbon dioxide concentration ([CO_2]); elevated atmospheric carbon dioxide concentration (e[CO_2]); environment type (ET); genotype by environment by management interaction (GxExM); harvest index (HI); leaf area index (LAI); specific leaf area (SLA)

Introduction

Atmospheric CO_2 concentrations ($[CO_2]$) have been increasing from about 280 ppm to more than 400 ppm from the pre-industrial era until now. If global greenhouse gas emissions remain at the 2010 level, then atmospheric $[CO_2]$ will reach 550 ppm by 2050 (IPCC, 2014). For C3 crops like wheat, this considerable increase in the substrate of photosynthesis leads to higher growth rates, better water use efficiency and larger yields in the absence of temperature or rainfall changes (Leakey *et al.*, 2009). Because $e[CO_2]$ reduces stomatal conductance, and therefore increases transpiration efficiency, the response to elevated CO_2 ($e[CO_2]$) has often been reported to be greater under conditions of water stress and/or in semi-arid environments (Fitzgerald *et al.*, 2016; Leakey *et al.*, 2009; Gifford 1979). However, there are also recent reports that seem to indicate the contrary (Gray *et al.*, 2016) or that there are no consistent differences (van der Kooi *et al.*, 2016), leading some to suggest the response to $e[CO_2]$ might depend on the timing of the drought experienced by the crop (Bourgault *et al.*, 2017). The relationships between timing and extent of drought and trade-offs between biomass stimulation and leaf level water use efficiency gains have been shown to be especially important in highly variable, semi-arid or Mediterranean field conditions (Tausz-Posch *et al.*, 2019).

Early vigour, defined as a larger leaf area early in the season, is a trait that has recently received attention for its benefits under Mediterranean environments where intermittent, low rainfall conditions prevail (Rebetzke and Richards, 1999). The greater cover reduces evaporation from the soil surface and this water is then available for transpiration. Early vigour in wheat is associated with greater early root development, greater early nitrogen uptake and higher photosynthetic nitrogen use efficiency (Pang *et al.*, 2014), and appears to be partially achieved through a larger specific leaf area (SLA) early in the development of the plant (Rebetzke *et al.*, 2004). Multi-regional field trials have shown that the value of the trait is more important in areas that receive frequent small rainfall events during the growing season, but is not as useful in areas that depend on stored soil water (Wilson *et al*., 2015).

If crop traits affect the response to e[CO_2] in crops, it might be possible to take advantage of rising atmospheric [CO_2] by selecting for greater responsiveness in crop breeding programmes either directly or by selecting traits that are associated with a greater response (Ainsworth *et al.*, 2008; Ziska *et al.*, 2012; Tausz *et al.*, 2013). On the other hand, there could also be drought adaptive traits that, by modifying growth patterns in time, might negatively impact the response to e[CO_2] (Bourgault *et al.*, 2013). Considering crop breeding takes 10-20 years to release cultivars, it is important to evaluate future environmental conditions, such as increased [CO_2] on their relative performance. On the one hand, larger leaf area early in crop development leads to greater photosynthesis and lower evaporation from the soil surface. It is possible therefore that early vigour lines might benefit more than low vigour lines from the effects of e[CO_2], as they may carry an early benefit in both carbon gain and increased transpiration efficiency throughout the season. On the other hand, depending on the result of the competing effects of increased transpiration efficiency and lower evaporation compared to the higher water use of the larger leaf area development, too much growth early in the season could deplete the soil profile of water before grain filling. In environments where crops depend on stored water, this would lead to severe terminal drought stress (Nuttall *et al.*, 2012), analogue to the “haying-off” phenomenon observed in response to excess early N fertilizer application (van Herwaarden *et al.*, 1998). This could therefore negate some of the benefits from e[CO_2] under terminal drought but not under well-watered conditions or more moderate types of drought. We would thus expect that the benefits of this trait and its impact on the response to e[CO_2] might depend on the seasonal pattern of water availability.

Crop simulation models are particularly useful to investigate genotype by environment (GxE) interactions that are difficult to test experimentally and for climate scenarios that are not yet realised. Field and controlled environment experiments testing pairs of lines differing in traits such as tillering propensity, early vigour or water soluble carbohydrate accumulation in wheat have rarely been able to detect genotypic differences in the response to e[CO_2] (Bourgault *et al.*, 2013; Tausz-Posch *et al.*, 2015). This trait by [CO_2] interaction, if it exists, may be masked by large GxE variability related to environmental factors other than changes in [CO_2] and by background genetic differences not related to the trait being evaluated. This makes the occurrence of Type II statistical errors highly likely where true differences are not detected as significant. While this risk can be minimised by more elaborate and expensive experimental designs, complementing field data with simulation results can help explore GxE or, in this case, Gx[CO_2]xE interactions and can offer additional insights regarding likely interactive mechanisms between physiological traits and environmental factors, information that is useful to breeders. They can also deliver information about variables that are not typically observed and highlight further research questions to evaluate in the field. However, comparison against measured data is still fundamental to validate findings and improve crop models (Boote and Sinclair, 2006; Fischer, 2011).

Because drought as an environmental factor can have vastly different effects on crop growth based on its timing and intensity, more refined analyses of the E component of the GxE interaction might be needed. For example, Chenu *et al.* (2013) clustered wheat growing seasons for various locations across Australia into four environment types (ET) based on the ratio of crop water demand met by the soil water supply. The four environment types described four different drought patterns: no stress or negligible stress (ET1), moderate water stress post-anthesis (ET2), moderate to severe water stress beginning before anthesis but relieved during grain filling (ET3), or that continued through to maturity as a severe terminal drought (ET4). Because of the various feedback mechanisms between crop growth and the environment, such characterisation is more useful in exploring GxE interactions than aggregated meteorological data to understand the response to stressful conditions and evaluate the usefulness of adaptive traits (Chenu *et al.*, 2013).

The objective of this study was to evaluate the response to e[CO_2] of wheat lines that differed in early vigour under semi-arid conditions, and whether this was influenced by seasonal patterns of water availability. Specifically, we hypothesised that the greater increase in leaf area development in high early vigour lines grown under e[CO_2] would lead to more water use pre-anthesis and subsequently greater drought stress post-anthesis, therefore reducing the grain yield response to e[CO_2], but only in severe terminal stress conditions of ET3 and ET4 environments as characterised by Chenu *et al.* (2013). In order to do this, we identified a subset of lines grown in the Australian Grains Free Air CO_2 Enrichment (AGFACE) research programme which included lines with proven expression of high and low early vigour, and compare their response to e[CO_2]. We also extracted from the experimental data variables needed to modify early leaf area development and model the early vigour trait in two crop models: APSIM and SIMPLACE. We were then able to compare the simulated response to e[CO_2] between high and low early vigour lines, but also to investigate the water use dynamics between the two groups. We were thus able to answer the following questions:

1. Do simulations give us similar responses to e[CO_2] than the experimental data?
2. Does the response to e[CO_2] depend on the type of drought experienced by the crop?
3. Does e[CO_2] lead to more severe terminal drought stress in high vigour lines?

The experimental and simulation results showed that the response to e[CO_2] of high early vigour lines was indeed lower, but greater water use pre-anthesis under e[CO_2] was not associated with the early vigour trait. Therefore, we further asked:

4. How else could we explain the lower response to e[CO_2] in high early vigour lines?

Materials and Methods

AGFACE Experiment

The Australian Grains Free Air CO_2 Enrichment (AGFACE) facility was located near Horsham, Victoria (36°45’07”S 142°06’52”E, 127 m above sea level). Long term average (based on 1981-2010 period) annual rainfall is 435 mm, with approximately 320 mm falling during the winter growing season (from May to November inclusively). Average maximum and minimum temperatures are 17.6°C and 5.3°C respectively during the season, with July being the coldest month (Bureau of Meteorology, 2016). Elevated CO_2 levels (target 550 mmol mol^{-1} air) were maintained during daylight hours by injecting pure CO_2 into the air on the upwind side from horizontal stainless-steel tubes so the gas would be carried across the ring. Concentrations were maintained within 90% target (495-605 mmol mol^{-1} air) for 93-98% of the time. More details on the site and the CO_2 exposure equipment are given in Mollah *et al.* (2009). Details about the general management of the experiment are also given in O'Leary *et al.* (2015) and Fitzgerald *et al.* (2016).

The experimental design was a randomized complete block split-split-plot design with 4 ambient and 4 elevated CO_2 octagonal wheat plots organised in blocks each containing one ambient (a[CO_2]) and one e[CO_2] main plot. In each year, a plus/minus supplemental irrigation treatment was usually nested within each main plot, except in 2007 when time of sowing was nested as sub-plots, and supplemental irrigation treatments were on separate main plots. There were two or more cultivars grown inside each sub-plot. More details are available on the design and treatments in O'Leary *et al*. (2015) and Fitzgerald *et al.* (2016) for 2007 to 2009, from Tausz-Posch *et al.* (2012, 2013, 2015) for 2009 to 2010 and in Houshmandhar *et al.* (2015) for 2013.

Initial soil water data was obtained from field samples collected before sowing in each main plot using a hydraulic soil corer and the gravimetric method at 0-10, 10-20 and in 20 cm increments thereafter up to 1.2 to 1.8 m depending on the year. These samples were averaged over the entire site from 2007 to 2010 as main plots were moved, whereas from 2011 to 2013, these were averaged over irrigation treatments as the main plots were rotated with field pea plots that were also subject to a plus/minus supplemental irrigation treatment. Bulk density was calculated for each layer based on the site average and assumed constant throughout the experiment. Initial soil mineral nitrate concentrations were also obtained from field samples at depths of 0-10, 10-20, 20-60 cm on separate samples and averaged over the entire site in each year.

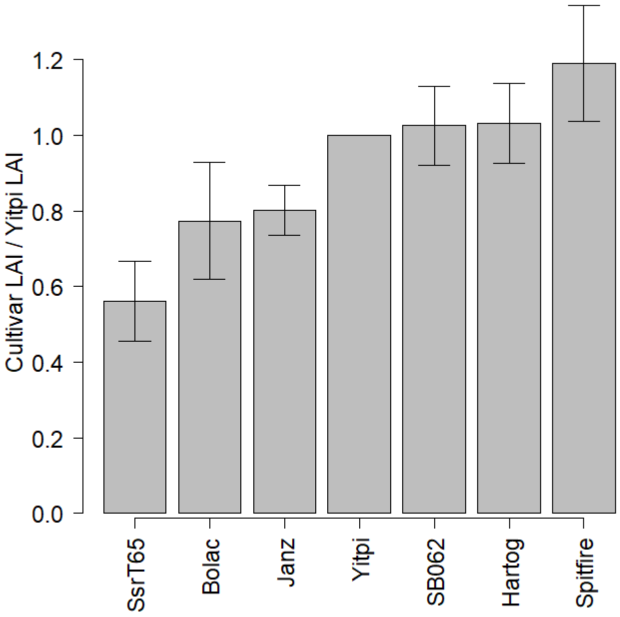
Destructive harvests were performed at the first node stage, anthesis and maturity (DC31, DC65 and DC90 according to the scale by Zadoks *et al*. (1974); see Table 1 for dates) on pre-determined areas of 50 to 75 cm in length (depending on year and sampling) across 4 rows excluding edge rows (from 2007 to 2009) or 5 rows including edge rows (from 2010-2013). The inclusion of edge rows is a source of increased experimental error, both in terms of increased variability and overestimation of treatment means (Rebetzke et al., 2013), but we assumed that DC31 data, the calculated specific leaf area data, and mean biomass and yield responses to e[CO_2] were not affected. At DC31 and DC65, a subsample was taken from the biomass sample and separated into green leaves, dead leaves, stems and heads (if applicable), and leaf area was determined on this subsample (LI-3100C or LI-3000C with conveyor belt, LI-COR Biosciences, Lincoln, NE, USA). Leaf area of the quadrat harvested was extrapolated based on the ratio of the subsample biomass to the total quadrat biomass, and leaf area index (LAI) was calculated by dividing this by the quadrat area. Specific leaf area (SLA) was calculated by dividing the subsample leaf area by the subsample green leaf biomass. At DC90, samples were dried at 40 or 70°C depending on the year, but biomass and grain yield were calculated and presented on a 0% water content basis. Sampling dates are given for each cultivar and each year in Table 1.

Table 1: Cultivars selected from AGFACE 2007-2013 with planting date, date of first node development (DC31), anthesis (DC65) and final harvest (DC90) samplings.

Cultivar	Vigour classification (see Figure 1)	Planting date	Stem elongation (DC31) sampling date	Anthesis (DC65) sampling date	Final harvest (DC90) sampling date
2007					
Janz	Low	18 Jun	6 Sept	29 Oct	12 Dec
Yitpi	Reference	18 Jun	6 Sept	29 Oct	12 Dec
2008					
Janz	Low	4 Jun	20 Aug	20 Oct	8 Dec
Yitpi	Reference	4 Jun	20 Aug	20 Oct	8 Dec
2009					
Hartog	High	23 Jun	2 Sept	27 Oct	4 Dec
Janz	Low	23 Jun	2 Sept	27 Oct	4 Dec
Yitpi	Reference	23 Jun	2 Sept	27 Oct	4 Dec
2010					
Hartog	High	27 May	5 Aug	18 Oct	9 Dec
Janz	Low	27 May	5 Aug	18 Oct	9 Dec
Yitpi	Reference	27 May	5 Aug	22 Oct	9 Dec
2011					
Janz	Low	25 May	1 Aug	12 Oct	5 Dec
SB062	High	25 May	1 Aug	12 Oct	5 Dec
SsrT65	Low	25 May	1 Aug	7 Oct	5 Dec
Yitpi	Reference	25 May	1 Aug	17 Oct	5 Dec
2012					
Janz	Low	30 May	15 Aug	18 Oct	5 Dec
SB062	High	30 May	15 Aug	15 Oct	5 Dec
SsrT65	Low	30 May	7 Aug	8 Oct	5 Dec
Yitpi	Reference	30 May	15 Aug	18 Oct	5 Dec
2013					
Bolac	Low	29 May	13 Aug	21 Oct	6 Dec
Spitfire	High	29 May	13 Aug	8 Oct	2 Dec
Yitpi	Reference	29 May	13 Aug	16 Oct	6 Dec

We restricted our analysis to include the reference cultivar “Yitpi” which was grown in every year from 2007 to 2013, as well as three cultivars which expressed high early vigour and three cultivars with low early vigour, based on LAI data collected at DC31 in AGFACE. In order to remove the effect of the year on LAI, we calculated the ratio of each cultivar LAI to the reference cultivar “Yitpi” under a[CO_2] for each year, and averaged this ratio across years if the cultivar was grown in more than one year (Figure 1). Efforts were made to select cultivar combinations that would span as many years as possible in the experimental data. The list of selected cultivars and years in which they were grown is given in Table 1. The normalized LAI (or ratio to reference cultivar "Yitpi") at DC31 was found to be significantly different between the two groups ($p<0.001$).

Figure 1: Ranking of cultivars for early vigour based on comparison with reference cultivar Yitpi leaf area index (LAI) at the first node stage (DC31) in AGFACE 2007-2013 based on subsequent grouping into low (<1), and high (>1) early vigour groups. Bars represent the average ratio of the cultivar leaf area index (LAI) to Yitpi LAI across the years grown and are presented ± standard errors.



Statistical Analyses

Given the design was slightly different across years, analyses were conducted separately for each year with ASREML-R 3 (Butler *et al.*, 2009; R Core Team, 2017). The effects of [CO_2], plus/minus supplemental irrigation, and early vigour groups (or cultivars) were treated as fixed and Wald tests were performed. Blocks, main plot number (nested within blocks), subplot number (nested within plots) and sub-subplot number (nested within subplots) were considered random effects. Predicted means were obtained for leaf area index at DC31 with the complete data set and used to select cultivars that expressed high and low early vigour, as described above. Analyses of the response to e[CO_2] were performed using early vigour groups or cultivars depending on the question being investigated.

Simulation models

APSIM-Wheat

The Agricultural Production Systems Simulator (APSIM version 7.8) is a modular modelling simulation framework that employs a daily time step and has been extensively tested for wheat in Australian conditions. APSIM modules consist of plant, soil and management modules that simulate crop growth, soil water and nitrogen dynamics and various management controls (Holzworth *et al.*, 2014).

Growth is conceptualised as biomass accumulation from radiation intercepted, limited by water deficit (if any) and modified by elevated [CO_2] (if applicable) and stress factors such as temperature and nitrogen deficiency. The [CO_2] factor is calculated from the atmospheric CO2 concentration and temperature as published in Reyenga *et al.,* (1999). This growth is partitioned into roots, stems, leaves and reproductive structures according to plant developmental stage. Wheat phenology is based on thermal time accumulation for each of 11 predefined stages of development and modified for cultivar-specific differences with photoperiod sensitivity and vernalisation sensitivity parameters. Plant organ biomass and leaf area are initialised at emergence. The default value of the initial leaf area is set at 200 mm^2 plant^{-1}. The daily increase in carbon-limited leaf area is calculated from the increase in leaf dry weight and maximum specific leaf area which varies from 27000 to 22000 mm^2 g^{-1} depending on leaf area index. The actual daily increase in leaf area is the minimum between the carbon-limited leaf area and the stressed leaf area, which is a function of the potential increase in leaf number and leaf size and modified by stress factors related to nitrogen deficiency and soil water deficit effects on canopy expansion. Crop water demand is modelled as a function of the potential daily biomass accumulation divided by the transpiration efficiency, which in turn is a function of the average daylight vapour pressure deficit (VPD) and [CO_2], but capped at a factor of 1.5 of the Priestly-Taylor potential evapotranspiration (Eo). The [CO_2] factor is calculated from a linear function and is approximately 1.05 at 400 ppm, and 1.21 at 550 ppm. The VPD is estimated using daily maximum and minimum temperatures according to Tanner and Sinclair (1983). The crop water supply (or potential soil water uptake) is based on the approach by Monteith (1986) as the sum of potential root water uptake from each profile layer where the rate of water extraction is determined by the KL factor which varies by layer with crop species and soil type (Zheng *et al.*, 2015).

SIMPLACE

The SIMPLACE modeling framework (Gaiser *et al.*, 2013) was used to bring together the aboveground growth and development aspects of the Lintul-5 crop growth model (Wolf, 2012), the SlimWater3 water balance model based on SLIM (Addiscott and Whitmore, 1991), and modified to use evaporation and crop water extraction functions from the FAO-56 ET0 manual (Allen *et al.*, 1998), a heat stress reduction function (Gabaldón-Leal *et al.*, 2016) and a canopy temperature model (Webber *et al.*, 2016). The resulting model configuration, SIMPLACE<Lintul5, SlimWater3, CanopyT, HeatStressHourly> is further referred to as SIMPLACE.

In SIMPLACE, intercepted photosynthetically active radiation is converted to crop biomass by multiplication with radiation use efficiency (RUE), which varies with phenological stage, mean temperature, water and nutrient stress as well as atmospheric CO_2 concentration. At 400 ppm, the [CO_2] factor is approximately 1.04 and 1.18 at 550 ppm. Daily biomass production is allocated to roots, leaves, stems and grain yield with water stress increasing partitioning to roots. Phenological development rates are driven by 24-hour mean temperatures, photoperiod and vernalization requirements. Leaf area growth is initially exponential and later governed by the product of biomass expansion and specific leaf area, which can vary with the phenological stage. Water stress is conceptualized as the ratio of actual transpiration to potential transpiration, which is estimated as the product of a reference crop evapotranspiration and the transpiration crop coefficient following the FAO-56 dual crop coefficient methodology (Allen *et al.*, 1998). This is denoted TRANRF in SIMPLACE outputs. Transpiration is also reduced linearly by 0.336% per ppm as ambient CO_2 concentrations increases (Zhao *et al.*, 2015). Daily crop water use is calculated as the minimum of crop available water in the soil and potential transpiration.

Cultivar-specific calibration

In APSIM, cultivar values were available to determine time to anthesis and time to maturity for all but two genotypes (SsrT65 and SB062). SsrT65 was given values of Silverstar based on its genetic background and previous observations of similar phenology within AGFACE and elsewhere (Bourgault *et al.*, 2013; Mitchell *et al.*, 2012). SB062 was given values identical to Yitpi based again on observed values within AGFACE. In addition, the cultivar Bolac had additional parameters associated with thermal time to flower initiation, flowering and grain filling. These were set to the base cultivar values for a better fit with the experimental data. The values for the other genotypes were checked against experimental data and deemed acceptable without modifications (Table 2; Figure S1). The APSIM model was run without further calibration apart from modifications to parameters to define the early vigour groups (see below). In SIMPLACE, thermal time to anthesis and thermal time from anthesis to maturity were estimated from the experimental data (provided in Table 2; see also Figure S1). The default values for biomass partitioning were also checked against experimental data and deemed acceptable without modifications.

Table 2: Crop model phenological parameters for the cultivars used in this study.

	APSIM		SIMPLACE/Lintul5	
	photo_sens*	vern_sens*	TSUM1**	TSUM2**
Yitpi	3.0	1.5	850	750
Janz	2.2	2.9	800	750
SsrT65	1.5	1.5	750	750
Bolac	3.1	3.1	925	750
Hartog	3.0	1.5	850	750
SB062	3.0	1.5	850	750
Spitfire	3.0	1.5	850	750

* photo_sens and vern_sens are parameters in the APSIM wheat module that relate to photoperiod and vernalisation sensitivity, respectively. These modify thermal time parameters to extend or contract thermal time in early phases of crop development.

** TSUM1 and TSUM2 in SIMPLACE-Lintul5 specify the thermal time requirements from emergence to anthesis and from anthesis to maturity, respectively, and are modified by both a response to photoperiod and a small vernalisation response.

Modification of early leaf area development and vigour groups

Early vigour was modified in APSIM by modifying the relationship between SLA and LAI. To obtain regression coefficients of the maximum SLA as a function of LAI as observed in AGFACE and compare this to APSIM model parameters, we fitted boundary lines on experimental data with quantile regression estimate of 0.95 using the “quantreg” package in R (Koenker, 2017). SIMPLACE uses SLA values as a function of crop development and these were therefore modified according to approximations of experimental data from AGFACE (Table 3). SLA values used by SIMPLACE are for newly expanded leaves and therefore some calibration was needed to arrive at the integrated SLA value at DC65. The differences in SLA at the DC31 and DC65 growth stages were in general well simulated by APSIM and SIMPLACE with the modifications we imposed (Figure S2). The modification of this one parameter in the models was sufficient to bring about the changes in leaf area index at DC31 and at DC65 that we observed in the experimental data (Figures S3 and S4).

Table 3: Specific Leaf Area (SLA, in cm^2 g^{-1}) differences between early vigour groups from selected cultivars in AGFACE (2007-2013) and translation of values into model inputs.

<table>
<tr><th></th><th colspan="2"><i>APSIM</i></th><th colspan="3"><i>SIMPLACE</i></th></tr>
<tr><th>Cultivar/Group</th><th>Calculated slope and intercept for 95 percentile of SLA (cm^2 g^{-1}) as a function of LAI</th><th>max SLA</th><th>SLA (cm^2 g^{-1}) at DC31</th><th>SLA (cm^2 g^{-1}) at DC65</th><th>SLATB (in m^2 g^{-1})</th></tr>
<tr><td>High vigour group</td><td>300 – 7x</td><td><table><tr><td>LAI</td><td>Max SLA (mm^2 g^{-1})</td></tr><tr><td>0.0</td><td>30000</td></tr><tr><td>5.0</td><td>26000</td></tr></table></td><td>224 ± 14</td><td>174 ± 12</td><td><table><tr><td>DVS</td><td>SLA</td></tr><tr><td>0.0</td><td>0.022</td></tr><tr><td>0.4</td><td>0.022</td></tr><tr><td>1.0</td><td>0.016</td></tr><tr><td>2.0</td><td>0.016</td></tr></table></td></tr>
<tr><td>Low vigour group</td><td>250 – 5x</td><td><table><tr><td>LAI</td><td>Max SLA (mm^2 g^{-1})</td></tr><tr><td>0.0</td><td>25000</td></tr><tr><td>5.0</td><td>22000</td></tr></table></td><td>191 ± 14</td><td>152 ± 12</td><td><table><tr><td>DVS</td><td>SLA</td></tr><tr><td>0.0</td><td>0.018</td></tr><tr><td>0.4</td><td>0.018</td></tr><tr><td>1.0</td><td>0.011</td></tr><tr><td>2.0</td><td>0.011</td></tr></table></td></tr>
<tr><td>Yitpi</td><td>256 – 5x</td><td><table><tr><td>LAI</td><td>Max SLA (mm^2 g^{-1})</td></tr><tr><td>0.0</td><td>27000</td></tr><tr><td>5.0</td><td>22000</td></tr></table>(Default values)</td><td>196 ± 14</td><td>155 ± 12</td><td><table><tr><td>DVS</td><td>SLA</td></tr><tr><td>0.0</td><td>0.019</td></tr><tr><td>0.4</td><td>0.019</td></tr><tr><td>1.0</td><td>0.012</td></tr><tr><td>2.0</td><td>0.012</td></tr></table>(Default values)</td></tr>
</table>

In APSIM, the maximum SLA parameter value is a function of the LAI, whereas in SIMPLACE, the SLA value (within the SLATB parameter) is a function of the developmental stage (DVS), which varies from 0 to 2 (maturity), with 1 being anthesis.

Simulation of AGFACE results

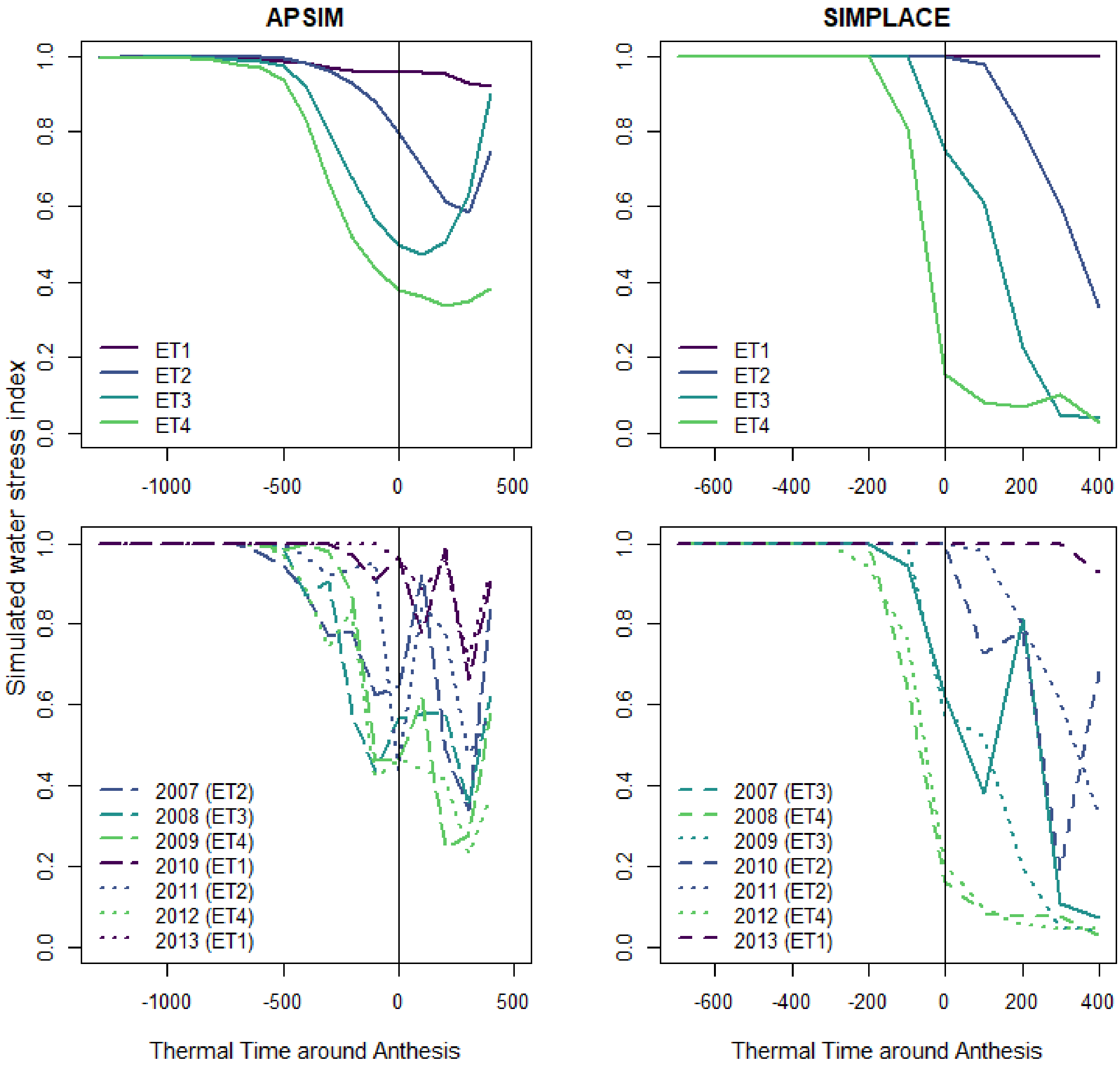
Simulations were performed with all seven cultivars in the seven years investigated (2007-2013) using weather data obtained on-site (MEA Premium Weather Station, Measurement Engineering Australia, Magill, SA, Australia). Soil parameters related to the soil water lower limit, drained upper limit and saturated water content, as well as other soil parameters necessary for running the two models were taken from O'Leary *et al*. (2015) (Table S1). Four treatments were simulated (plus/minus supplemental irrigation, a[CO_2]/e[CO_2] treatments) using the experimental planting dates (Table 1), initial soil water content (Table S2), nitrate content (Table S3), and supplemental irrigation water used in AGFACE (Table S4). For comparisons with experimental data, only simulations with the cultivars grown in specific years were used. In addition, for DC31 data, comparisons were conducted based on the date of sampling rather than the simulated growth stage. To explore differences in simulated water use, all combinations were used to provide a balanced comparison between high and low early vigour lines.

Environmental characterisation

In APSIM, for each simulation, the proportion of the crop water demand satisfied by the soil water supply was calculated as a daily water stress index. This value was constrained between 0 and 1 where 1 represents 100% of the water demand satisfied by the available soil water content. The daily water stress index value was aggregated over periods of 100°Cd (thermal time) centred at anthesis, as presented in Chenu *et al.* (2013). Classification of each year by irrigation treatment by [CO_2] by cultivar combination into an environment type was done by minimising the sums of squared differences between the aggregated water stress index curve for each simulation and the water stress index curves of the four environment types from Chenu *et al.* (2013)(Figure 2).

In SIMPLACE, the parameter TRANRF, which represents the ratio of actual transpiration to potential transpiration, was similarly aggregated over 100°Cd and centred at anthesis. Since environmental type curves were not easily transferable between the two models, we used the partitioning clustering function *pam* from the *cluster* package in R (R Core Team, 2017) to define four environmental types (adapted from Chenu *et al.*, 2013) based on the full set of simulations (i.e. combinations of 7 cultivars, 7 years, a[CO_2]/e[CO_2] and plus/minus supplemental irrigation for a total of 198 simulations). The results of the environment type clustering for both models (including curves reproduced from Chenu *et al.*, 2013) are presented in Figure 2 (top row). Examples of the environmental characterisation based on the reference cultivar Yitpi under a[CO_2] and rainfed conditions are also presented (Figure 2; bottom row).

Figure 2: Environmental characterisation from modelling outputs. Top row refers to clusters identifying environmental types, while bottom row shows patterns of simulated water stress for the reference cultivar Yitpi under ambient [CO_2] and rainfed conditions. This water stress index was calculated in APSIM (left) from the ratio of the soil water supply to crop water demand (with an upper bound of 1), while the water stress index in SIMPLACE (right) is the ratio of actual transpiration to potential transpiration (TRANRF parameter). A water stress index of 1 represents no stress, while a value of 0 represents a situation where no soil water is available to the crop. Daily ratios were aggregated over periods of 100°Cd thermal time centred at anthesis. The top left panel is taken from Chenu *et al.,* 2013.

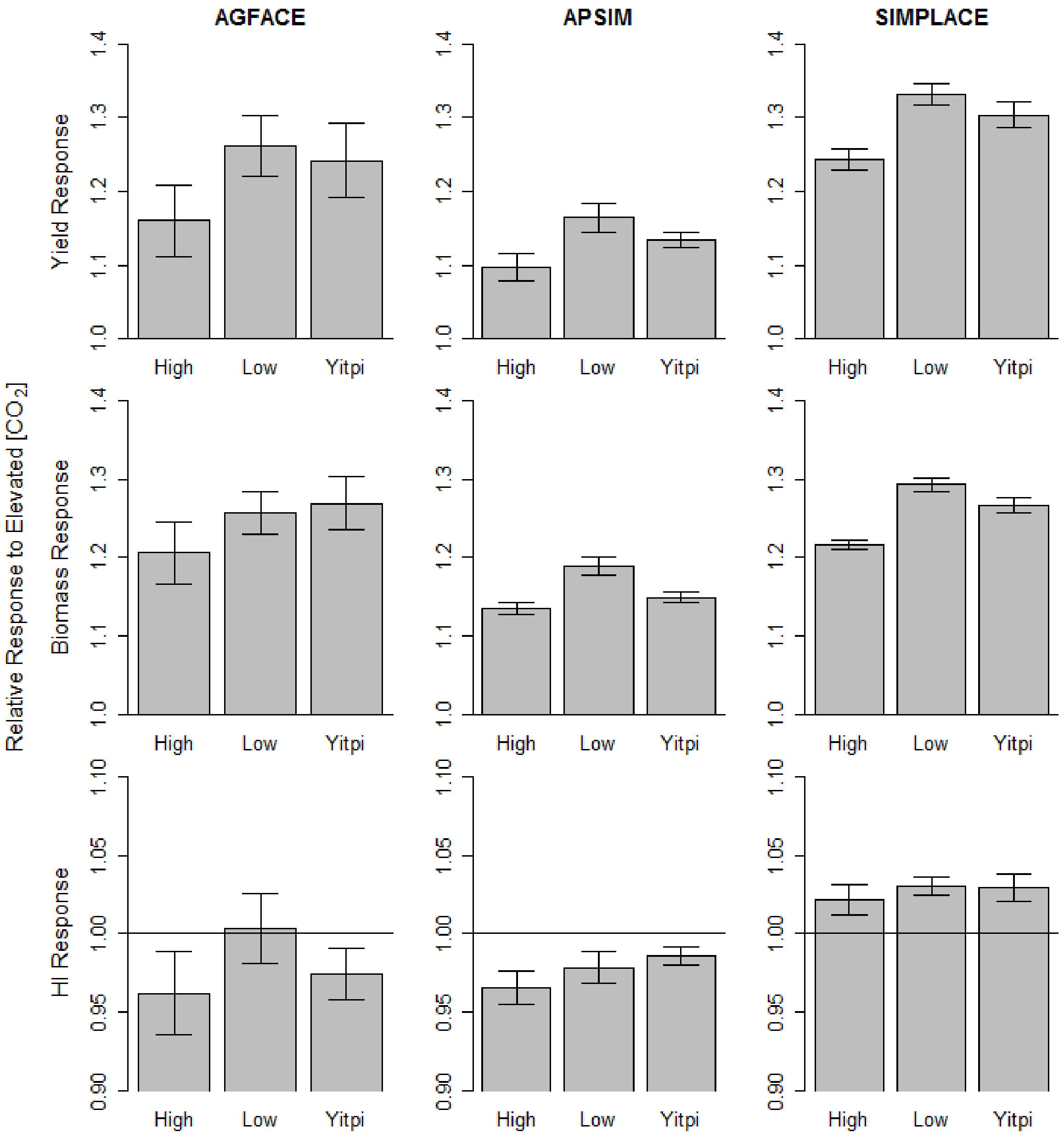


Results

Do model simulations give us similar responses to e[CO_2] than the experimental data?

Experimental results suggested that early vigour decreased the average grain yield response to e[CO_2] with high early vigour lines showing a 16% mean increase compared to 26% increase for low early vigour lines, although vigour group effects on the response were not statistically significant ($p=0.2879$; Figure 3). The biomass response at maturity was similar to the grain yield response, although with slightly higher mean values: 30% versus 39% for high and low early vigour lines, respectively ($p=0.4375$; Figure 3). For the harvest index (HI), there is a small non-significant decrease of 4% for high early vigour lines, but no change in low vigour lines ($p=0.1644$; Figure 3). Simulated results from both APSIM and SIMPLACE showed similar trends to the experimental data. In APSIM, the response to e[CO_2] was 10 and 16% for grain yield, and 13 and 19% for biomass at maturity for high and low vigour, respectively. Results from SIMPLACE showed slightly higher values but similar differences: 24 and 33% for grain yield, 22 and 29% for biomass at maturity (Figure 3). Although SIMPLACE generally showed a slight increase in HI as a response to e[CO_2], rather than the slight decrease observed in the experimental data and in APSIM simulation results, both models simulated the small differences in the response of HI between high and low early vigour lines accurately (Figure 3).

Figure 3: Responses to elevated [CO_2] according to early vigour group (high, low, and the cultivar Yitpi as reference) for yield, above-ground biomass at maturity (DC90) and harvest index (HI) in the Australian Grains Free Air CO_2 Enrichment (AGFACE) facility in 2007-2013 (experimental data) and as simulated by APSIM and SIMPLACE. Results presented are response means ± standard error of means.



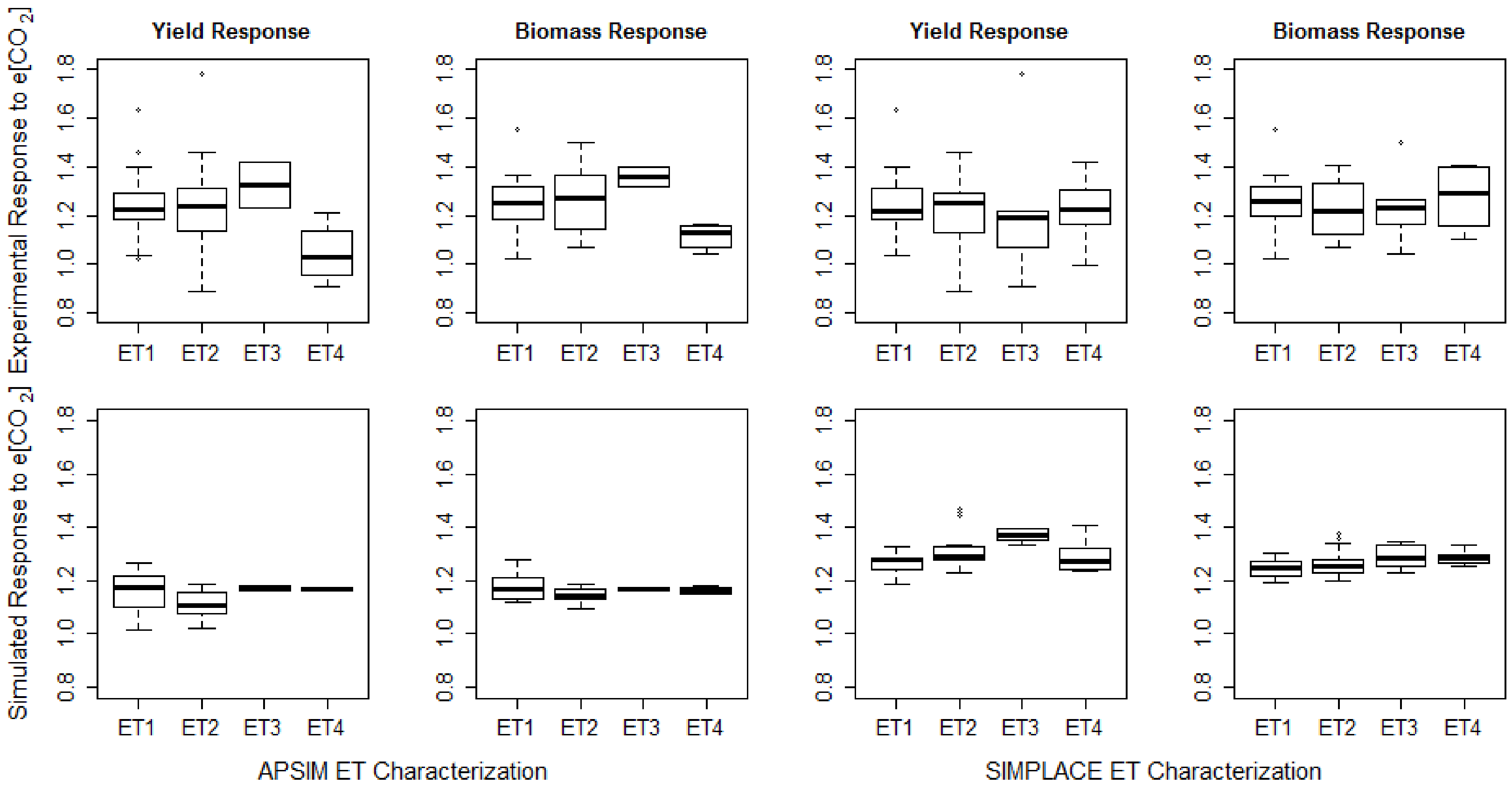
Does the response to e[CO_2] depend on the type of drought experienced by the crop?

As mentioned previously, the clustering of environment types was slightly different between the two models (Figure 2), and therefore the environmental characterization also varied between the two models. In general, however, both models followed the same trends, with SIMPLACE indicating more stress than APSIM in 29 instances out of 84 year-cultivar-irrigation-[CO_2] combinations (Table 4). Using the ET characterization from APSIM (under a[CO_2]), the experimental response to e[CO_2] for grain yield and biomass at maturity was smaller in the most severe terminal stress conditions (ET4; Figure 4, top left). This was not significant with the response in grain yield ($p=0.1485$), but was significant with the biomass response ($p=0.0296$). The vigour group by ET interactions for these two parameters were not statistically significant, but according to APSIM, the low stress environment types ET1 and ET2 represented the large majority of instances, while ET3 and ET4 were only experienced by Yitpi or high vigour lines (Table 4). Therefore, there were no experimental data for low vigour lines in ET3 and ET4 to compare to high early vigour lines in these same environments. By contrast, the ET characterization of experimental data with SIMPLACE did show instances of low early vigour lines experiencing ET3 and ET4 environment types in the experimental data set (Table 4). However, the responses to e[CO_2] in grain yield and final biomass showed no differences between the environment types (Figure 4 top right; $p=0.9710$ and $p=0.7992$ respectively). Similarly, there were no early vigour group by ET interactions, so there were no significant differences in response between high and low vigour lines in the more stressful environments.

Table 4: Environmental characterisation of AGFACE experimental data with APSIM and SIMPLACE simulations outputs, according to methodology by Chenu *et al.* (2013). The four environment types describe four different drought patterns: no stress or negligible stress (ET1), moderate water stress post-anthesis (ET2), moderate to severe water stress beginning before anthesis but relieved during grain filling (ET3), or that continued through to maturity as a severe terminal drought (ET4).

		APSIM Outputs				SIMPLACE Outputs			
		Rainfed		Supplemental		Rainfed		Supplemental	
Year	Cultivar	a[CO$_2$]	e [CO$_2$]	a[CO$_2$]	e [CO$_2$]	a[CO$_2$]	e [CO$_2$]	a[CO$_2$]	e [CO$_2$]
2007	Janz	ET2	ET2	ET2	ET2	ET2	ET2	ET2	ET2
	Yitpi	ET2	ET3	ET2	ET2	ET3	ET3	ET2	ET2
2008	Janz	ET2	ET2	ET2	ET2	ET4	ET4	ET3	ET3
	Yitpi	ET3	ET3	ET3	ET3	ET4	ET4	ET4	ET4
2009	Hartog	ET4	ET4	ET2	ET3	ET3	ET3	ET2	ET2
	Janz	ET2	ET2	ET2	ET2	ET3	ET3	ET2	ET2
	Yitpi	ET4	ET4	ET2	ET2	ET3	ET3	ET2	ET2
2010	Hartog	ET2	ET2	ET1	ET1	ET2	ET2	ET1	ET1
	Janz	ET1	ET1	ET1	ET1	ET2	ET2	ET1	ET1
	Yitpi	ET1	ET1	ET1	ET1	ET2	ET2	ET1	ET1
2011	Janz	ET2	ET2	ET1	ET1	ET2	ET2	ET1	ET1
	SB062	ET2	ET2	ET2	ET1	ET2	ET2	ET1	ET1
	SsrT65	ET1	ET1	ET1	ET1	ET1	ET1	ET1	ET1
	Yitpi	ET2	ET2	ET1	ET1	ET2	ET2	ET1	ET1
2012	Janz	ET2	ET2	ET1	ET2	ET4	ET3	ET2	ET2
	SB062	ET4	ET4	ET2	ET2	ET4	ET4	ET2	ET2
	SsrT65	ET1	ET1	ET1	ET1	ET3	ET3	ET1	ET1
	Yitpi	ET4	ET4	ET2	ET2	ET4	ET4	ET2	ET2
2013	Bolac	ET1	ET1	ET1	ET1	ET2	ET2	ET2	ET2
	Spitfire	ET1	ET1	ET1	ET1	ET1	ET1	ET1	ET1
	Yitpi	ET1	ET1	ET1	ET1	ET1	ET1	ET1	ET1

Figure 4: Experimental (top row) and simulated (bottom row) responses to elevated [CO_2] according to environment type (ET) for grain yield and above-ground biomass at maturity (DC90) in the Australian Grains Free Air CO_2 Enrichment (AGFACE) facility in 2007-2013 (experimental data) and as simulated by APSIM and SIMPLACE (with their respective ET characterization). Results are presented as boxplots, with the central line as the median, and the edges of the box as first and third quartiles.



The lower experimental response under ET4 with the APSIM ET characterization was not captured in simulated yield results (Figure 4 bottom row). Using the full set of simulations (i.e. all 7 cultivars in all 7 years, with a[CO_2]/e[CO_2], and plus/minus supplemental irrigation (196 instances)), we were able to simulate ET3 and ET4 environments experienced by low vigour lines (mostly with the longer-season cultivar Bolac). Simulated yield and biomass accumulation at maturity with APSIM did not show the lower response in ET4 (Figures S5 and S6 for yield and biomass response respectively) observed in the experimental data. On the other hand, in this balanced set of simulated data, both models simulated larger responses for low vigour lines compared to high vigour lines under ET1 and ET2 environments (Figures S5 and S6). Differences in response were quite consistent between the two models, despite using distinct clustering analyses.

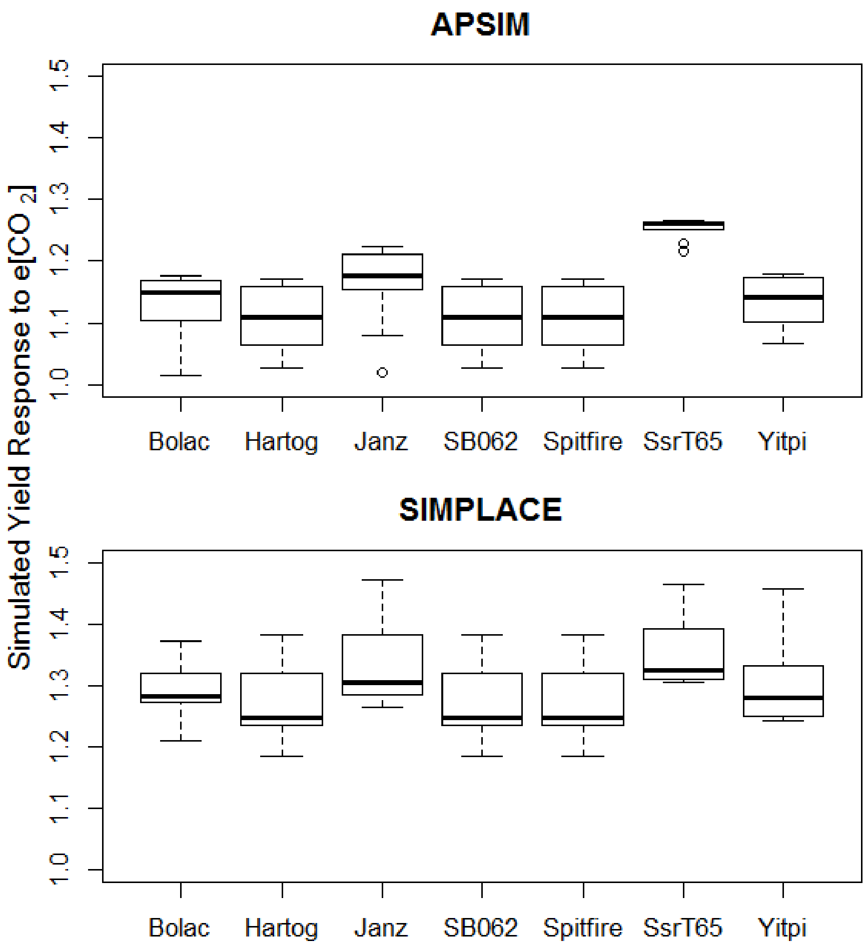
Does e[CO_2] lead to more severe terminal drought stress in high early vigour lines?

Simulation results with both models suggested that in most cases, the savings from greater transpiration efficiency and lower soil evaporation were more than enough to compensate for the greater transpiration from the larger leaf area, so that pre-anthesis water use was generally lower under e[CO_2]. When pre-anthesis water use was greater under e[CO_2], this was not necessarily associated with high early vigour lines or with greater water stress post-anthesis. In APSIM, the only instances (14 out of 98) when e[CO_2] led to greater water use pre-anthesis compared to growth at a[CO_2] involved low vigour lines (Janz and SsrT65). This led to greater water stress post-anthesis in the majority of cases (10/14 instances), but considering these all occurred in ET1 and ET2 environments, the stress was not important. For high vigour lines, e[CO_2] consistently led to more soil water availability in the profile at anthesis: although transpiration pre-anthesis was consistently greater with e[CO_2] due to the greater leaf area index, water savings from the lower soil surface evaporation and greater transpiration efficiency were more than enough to compensate. In SIMPLACE, pre-anthesis water use was greater in 16 instances in all vigour groups, but only 5 of these led to greater post-anthesis water stress: one with a low vigour line (Janz), one with Yitpi and three with high vigour lines (Hartog, SB062 and Spitfire). All these instances were characterized with SIMPLACE as ET4 environment types. This could indicate a greater propensity, in some cases, for e[CO_2] conditions to exacerbate drought stress in dry years, but the grain yield response to e[CO_2] did not seem affected (Figure 4).

How else could we explain the lower response to e[CO_2] in high early vigour lines?

Cultivar differences in simulations for this study were limited to phenology and early vigour. Out of the three selected low vigour cultivars in this study, SsrT65 and Janz are shorter-season than Yipti and high vigour lines, and phenology could therefore be a confounding factor: pre-anthesis and post-anthesis drought stress would occur under different conditions in the same season depending on when anthesis occurs. Based on the comparison between the three low vigour lines SsrT65, Janz and Bolac (which only differed in phenology in the models), time to anthesis did have a substantial effect on the simulated response to e[CO_2] (Figure 5). Both models were consistent in showing a higher response with a shorter time to anthesis. The comparison between high vigour lines and Yitpi (which all have the same phenology) confirmed that early vigour also reduced the response to e[CO_2]. In addition, the cultivar Bolac is a low vigour long-season cultivar which showed a larger grain yield response than high vigour lines despite its long phenology. Clearly, both traits have effects on the response to e[CO_2] according to simulated results.

Figure 5: Simulated Responses to elevated [CO_2] according to cultivars for grain yield in the Australian Grains Free Air CO_2 Enrichment (AGFACE) facility in 2007-2013, including 7 cultivars across 7 years, plus/minus supplemental irrigation (therefore including combinations that did not occur experimentally, but providing a balanced data set). Results are presented as boxplots, with the central line as the median, and the edges of the box as first and third quartiles.



We hypothesised high early vigour lines under e[CO_2] would respond with a greater increase in leaf area early in their development, which would lead to more water use pre-anthesis and greater drought stress post-anthesis, which would reduce the grain yield response to e[CO_2]. Models showed that a greater response in leaf area led to a greater increase in pre-anthesis water use and this led to more post-anthesis water stress with e[CO_2] in APSIM, but not in SIMPLACE (Figure S7). More post-anthesis water stress with e[CO_2] led to larger grain yield responses with APSIM, but smaller yield responses in SIMPLACE (Figure S8). In addition, models showed that low vigour lines, and not high vigour lines, responded with a larger pre-anthesis leaf area stimulation (Figure S7) and larger increases in radiation intercepted, and this was positively, rather than negatively, correlated with the grain yield response (Figure S8). The pre-anthesis response in radiation intercepted explained 63% of the response in grain yield in APSIM and 50% in SIMPLACE. The simulated higher grain yield responses in low vigour lines in ET1 and ET2 environments (Figures S5 and S6) further suggested a larger potential for growth increases in these lines under favourable conditions. This is consistent with a scenario where high vigour lines closed the canopy earlier in the crop development compared to low vigour lines and radiation becomes saturated and limiting growth.

Discussion

We hypothesised there might be a three-way interaction between vigour, [CO_2] and water availability post-anthesis, with high vigour lines under e[CO_2] being more prone to deplete soil water before grain filling, which would negate some of the benefits of e[CO_2]. Experimental data from AGFACE with a subset of cultivars with proven expression of high and low early vigour suggested a lower response from high early vigour lines but was not statistically significant. While simulations with both APSIM and SIMPLACE also suggested high vigour lines responded less to e[CO_2] than low vigour lines, this did not appear to be directly related to the scenario envisaged in our hypothesis. Instead, pre-anthesis growth stimulation, and in particular larger increases in radiation intercepted in low early vigour lines, explained differences in grain yield responses. The importance of radiation intercepted as illustrated by the models is in sharp contrast with the current literature: among environmental factors hypothesized to influence the response to e[CO_2], the scientific focus has rather been on investigating water and nutrient availabilities, sometimes with varying temperatures (see for example recent reviews by Kimball, 2016; Tausz-Posch *et al.*, 2019; van der Kooi *et al.*, 2016) while differences in radiation intercepted are not typically among the primary factors being considered. A notable connection, however, is the work by Shimono (2011; and Shimono *et al.* 2014, 2019) who suggested using the response to low density planting (in rice) as a surrogate for grain yield response to e[CO_2], although they also argue that the two responses would likely occur through different physiological mechanisms (Shimono *et al.*, 2019). To properly test the effect of radiation intercepted on the response to e[CO_2] in an experimental setting, data on the rate of canopy development and timing of canopy closure in addition to daily radiation would need to be collected.

The experimental yield and biomass response to e[CO_2], when characterized by drought type with APSIM, suggested that the response increased slightly with moderate water stress (in ET2 and ET3 environments), but decreased sharply in severe terminal drought stress (ET4 environment). These results are consistent with the study of Gray *et al.* (2016) with 8-year of soybean experimentation, which showed that severe drought led to a complete lack of response to e[CO_2]. This might also explain why the response to e[CO_2] is more variable with water stress as shown in a recent meta-analysis by van de Kooi *et al.* (2016). This study highlighted a gap in knowledge of the response to e[CO_2] under severe water stress scenarios. While the environmental characterisation with APSIM showed a tendency towards a lower response in the strongest terminal stress environment (ET4) with the experimental data (Figure 4), we still do not have many replicates of each ETs even with 7 years of data. In fact, by adding vigour as an additional factor, we are completely missing representations of low early vigour lines in ET3 and ET4 seasons, partly because of confounding effects of short phenology in 2 of the 3 low early vigour lines. While advanced statistical models such as mixed models used here can handle some missing data (Smith *et al.,* 2005), the absence of treatment combinations limits our confidence in results. In particular, the environmental characterization with SIMPLACE did not lead to the same pattern of effects on the response (Figure 4), which might be partially explained by some cultivar-season combinations being characterized differently based on the model used. However, the clustering analysis for SIMPLACE was also based on a much smaller data set than the APSIM clustering.

The lack of response to e[CO_2] under severe water stress as reported here and by Gray *et al.*, (2016) suggests there are still real gaps in knowledge about interactions between [CO_2] and water stress above and beyond the relationships with primary productivity and stomatal conductance. As simulated responses did not show this pattern, such knowledge has not been incorporated in simulation models. For example, Gray *et al.* (2016) showed that soybean plants grown under e[CO_2] were more responsive to water stress than those grown under a[CO_2], showing a lower stomatal conductance for the same leaf-level ABA concentration. Whether, or to what extent, this might also be true for wheat has not been investigated. In addition, Tausz-Posch *et al.* (2013) observed that differences in stomatal conductance in wheat depended on the time of day or the measurement day, so there may be other factors that interact with [CO_2] on an hourly or minute scale that are not captured in the single TE modifier used by models. Other research pointed out differences in the root-to-shoot ratio under e[CO_2]: for example, Pandey *et al.* (2018) showed increased partitioning to roots with e[CO_2] and low phosphorus conditions for hydroponically grown wheat plants. These findings remain to be confirmed in field settings. Nie *et al.* (2013) in a meta-analysis with heavy reliance on perennial systems showed e[CO_2] not only led to increased root-to-shoot ratio, but also increased proportion of roots at depth and increased proportion of fine roots. Such effects of e[CO_2], if also present in wheat, would modify the ratios of water demand and water supply in the models, and therefore the water stress indices from simulation models, potentially leading to different environmental characterisations under e[CO_2] and/or better relationships between water stress indices and the grain response to e[CO_2].

The simulation of leaf area curves assumes a bell-shaped growth, peaking shortly before anthesis, and simulation of early vigour therefore leads to a larger maximum leaf area. However, there are indications that in some populations bred for high early vigour this might not be the case: for example, Bourgault *et al.* (2013) demonstrated high early vigour lines had a greater leaf area for the first three leaves, but had fewer leaves, so their leaf area at anthesis was less than for the low early vigour parent. If we were to explore this in greater detail with such lines, the simulation of leaf area would need to be conceptualized in more detail and possibly with less reliance on SLA, which is assumed by models to be either fixed for a specific growth stage (SIMPLACE) or vary with LAI (APSIM), even though we know it is also sensitive to environmental factors and therefore a source of uncertainty in these models (Tardieu *et al.,* 1999; Setiyono *et al.,* 2008).

Then, how confident are we about the partitioning of ET into soil evaporation and transpiration in models? Although the LAI was increased approximately 30% with e[CO_2], cumulative differences between ambient and e[CO_2] simulations in both evaporation and transpiration were below 9 and 16 mm respectively. While it is expected that the higher transpiration demand from a greater LAI under

$e[CO_2]$ might be offset by savings through improved transpiration efficiency, are such small differences realistic for differences in soil evaporation? The time period during which differences in soil evaporation between high and low vigour lines is arguably quite short, and usually lies during the time of the year for which potential ET is low. Then perhaps the value of the early vigour trait in reducing evaporation might be secondary to other benefits, such as a more vigorous root system, which would allow for quicker uptake of soil water and nutrients early in the season (Palta *et al.*, 2011).

Finally, increasing atmospheric $[CO_2]$ will also cause increasing temperatures in most regions of the world. The net effect of the water savings from $e[CO_2]$ combined with the advanced crop development and higher water use from higher chronic temperatures and/or the effects of potential heat waves during grain filling with regards to biomass development and grain yield are highly uncertain. Although the inclusion of this fourth factor is not possible with this study, a proper assessment of the implications of genetic traits for climate change adaptation would require the evaluation of several scenarios in regards to temperature pattern changes. Previous research has shown that considering temperature and $[CO_2]$ effects separately resulted in very different findings compared to studies where the effects were combined, in particular with regards to crop water requirement and heat stress (Zhao et al., 2015).

Conclusion

Experimental data from AGFACE gave some indications that lines expressing high early vigour might respond less to $e[CO_2]$ than low vigour lines. Although the effect was not found to be statistically significant in the experimental data, two crop growth models supported this notion. We had hypothesized these observations could be explained by that high early vigour lines depleting available soil water earlier in the season when grown under $e[CO_2]$, and thus suffering from more severe terminal drought stress. Environmental characterizations of the seasonal pattern of drought stress in the experimental data and patterns of water use in the simulations however did not support this hypothesis. Rather, both simulation models suggested early canopy closure in high early vigour lines would limit the potential for increased growth and yield, especially in more favourable environments. Under less favourable conditions, the timing of the drought stress appears to influence the effect of e[CO_2]. This study highlighted the need to resolve whether there are differences in the response to e[CO_2] under drought types that differ in intensity and timing.

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The authors declare no conflict of interest.

The data that support the findings of this study are available from the corresponding author upon reasonable request and approval from the former AGFACE program leaders (M. Tausz and G. Fitzgerald).

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Supplemental information

Figure S1: Fit of phenological stages for APSIM and SIMPLACE against experimental data.

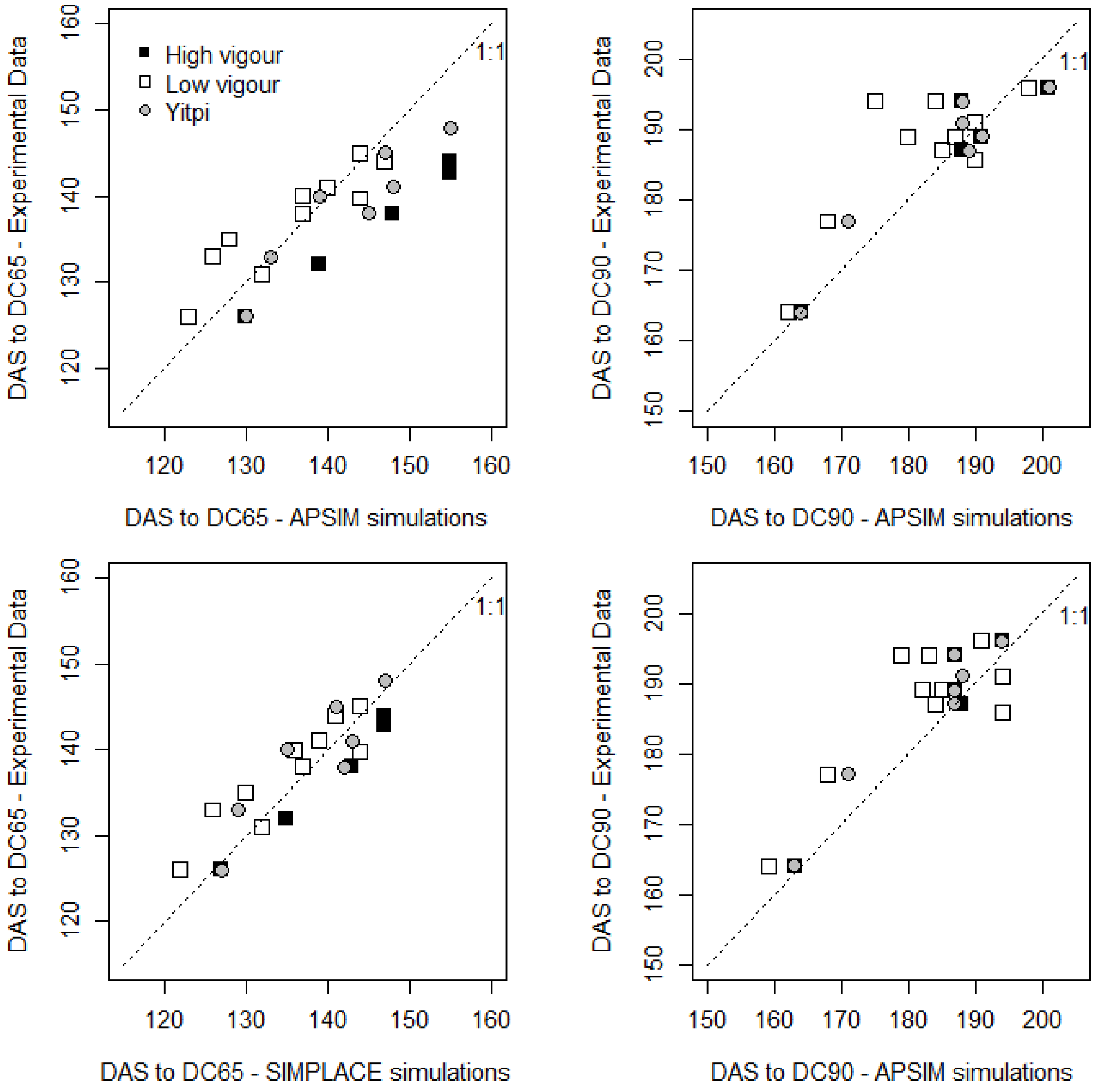
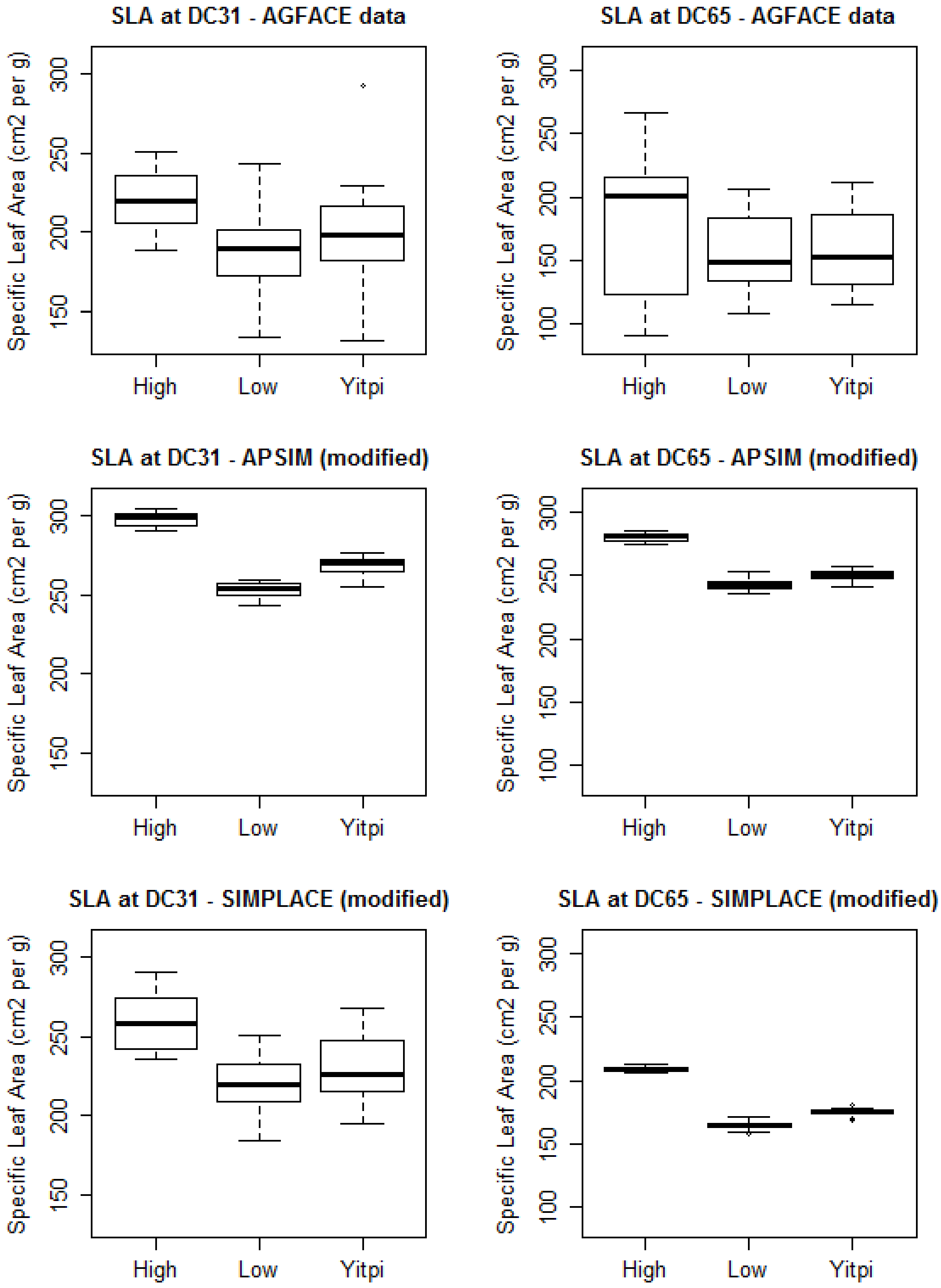


Figure S2: Range of specific leaf area (SLA) values of experimental data (AGFACE) and simulated data for both models with modifications for early vigour in the high and low vigour lines as described in Table 3.



SLA at DC31 - AGFACE data

SLA at DC65 - AGFACE data

SLA at DC31 - APSIM (modified)

SLA at DC65 - APSIM (modified)

SLA at DC31 - SIMPLACE (modified)

SLA at DC65 - SIMPLACE (modified)

Figure S3: Range of leaf area index values (LAI) values of experimental data (AGFACE) and simulated data for both models with modifications for early vigour in the high and low vigour lines as described in Table 3.

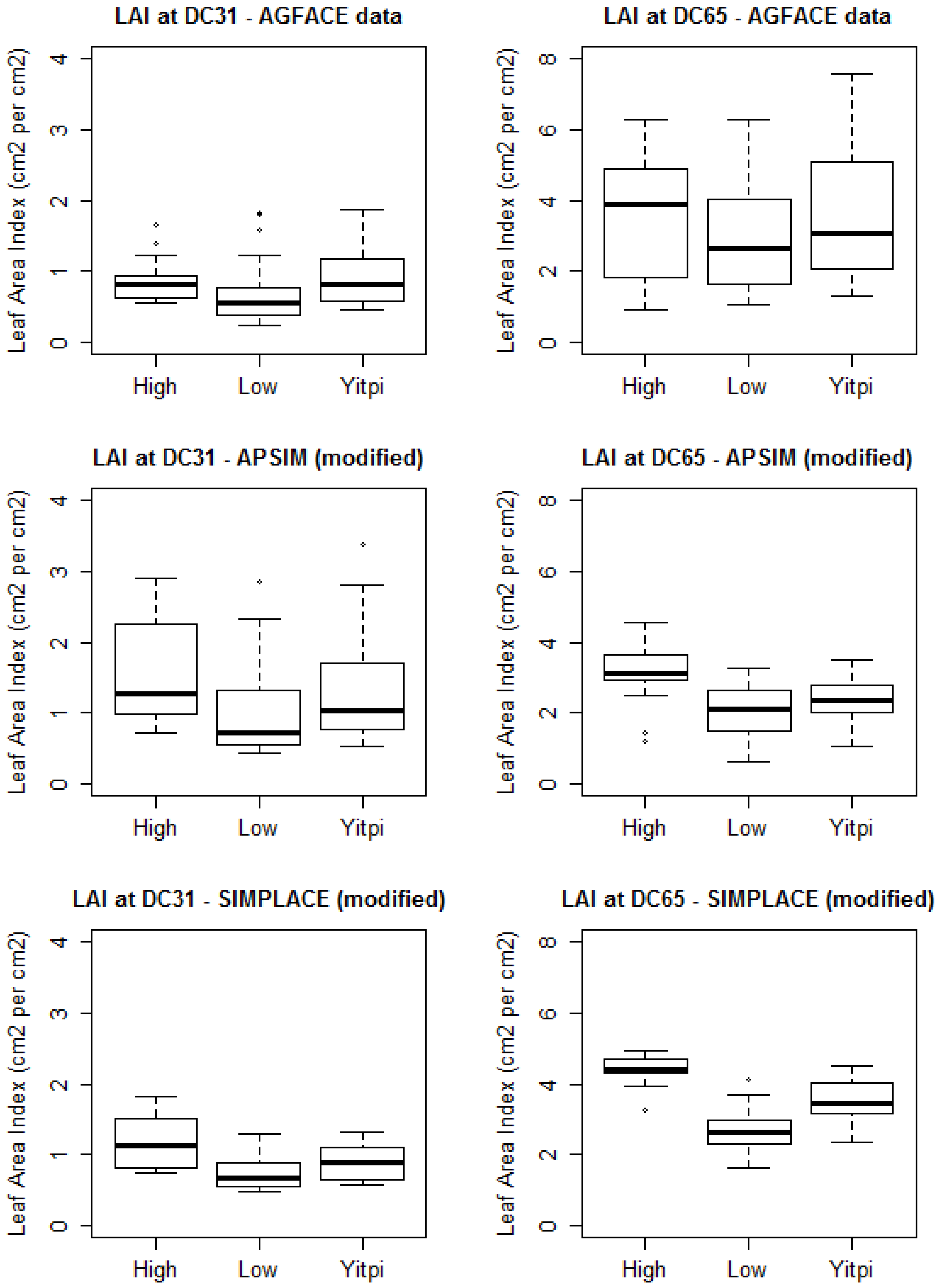


Figure S4: Fit of experimental AGFACE data to simulated results for leaf area index and above-ground biomass at stem elongation (DC31) for APSIM and SIMPLACE based on date of sampling. The group on the left higher than the 1:1 line is data from 2008, while the diffuse group underneath the 1:1 line with APSIM simulations is 2007 and 2009 (both planted late).

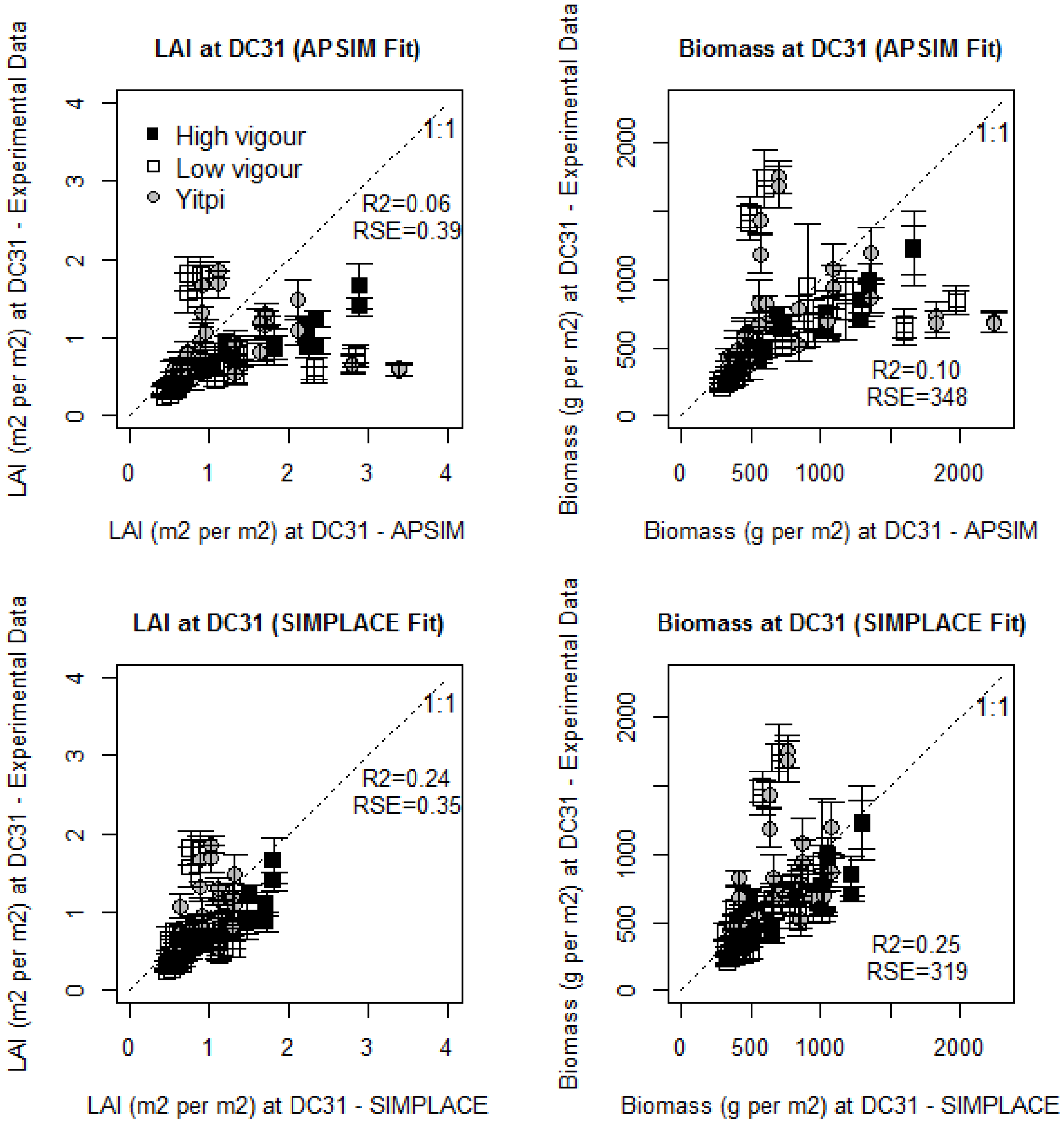


Figure S5: Simulated grain yield response to elevated [CO_2] according to vigour groups in different environment types (with characterization done with respective model) for the Australian Grains Free Air CO2 Enrichment (AGFACE) facility in 2007 to 2013. Simulated data includes all 7 cultivars in all 7 years, plus/minus irrigation (therefore including combinations that did not occur experimentally).

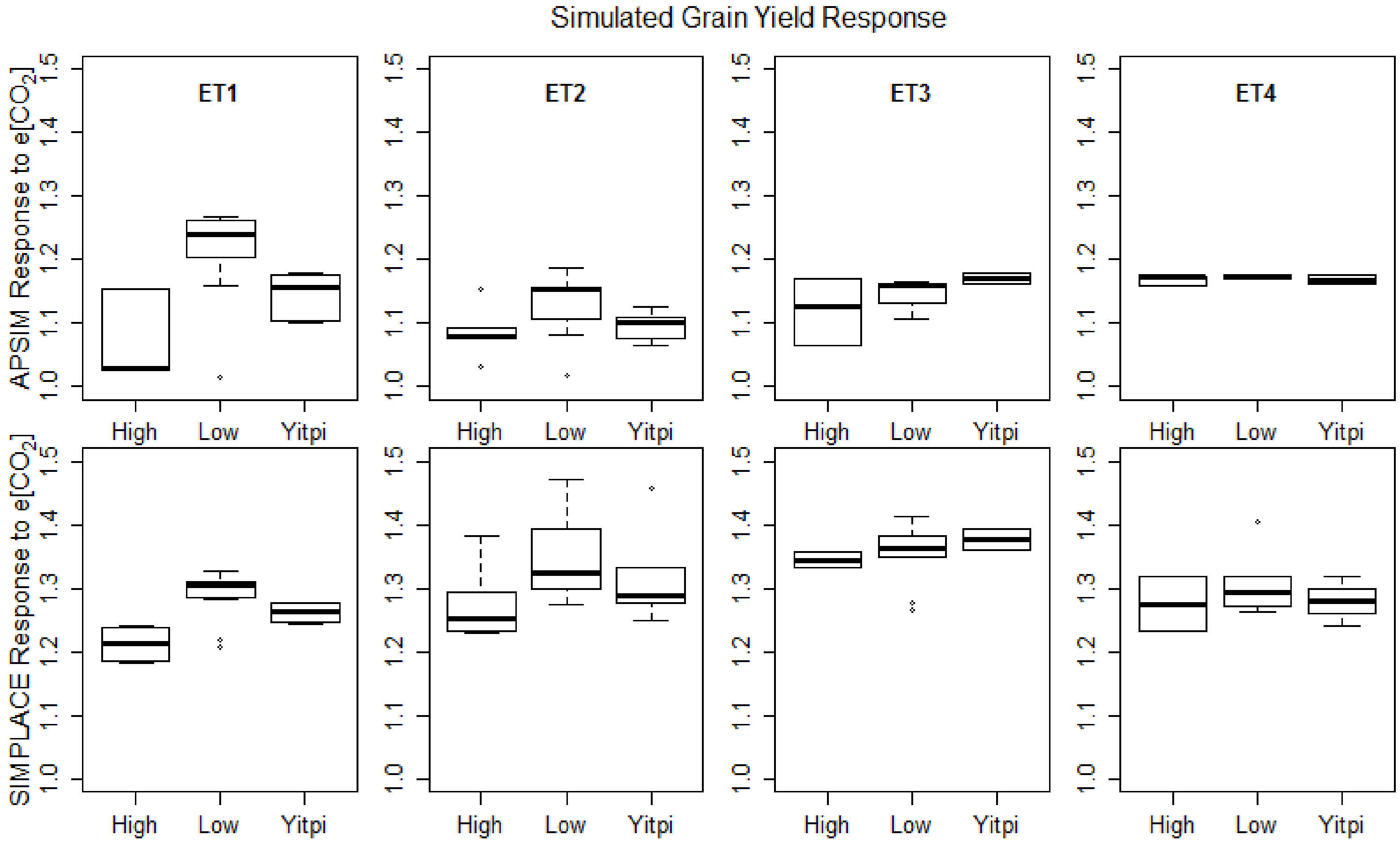


Figure S6: Simulated biomass response to elevated [CO_2] according to vigour groups in different environment types (with characterization done with respective model) for the Australian Grains Free Air CO2 Enrichment (AGFACE) facility in 2007 to 2013. Simulated data includes all 7 cultivars in all 7 years, plus/minus irrigation (therefore including combinations that did not occur experimentally).

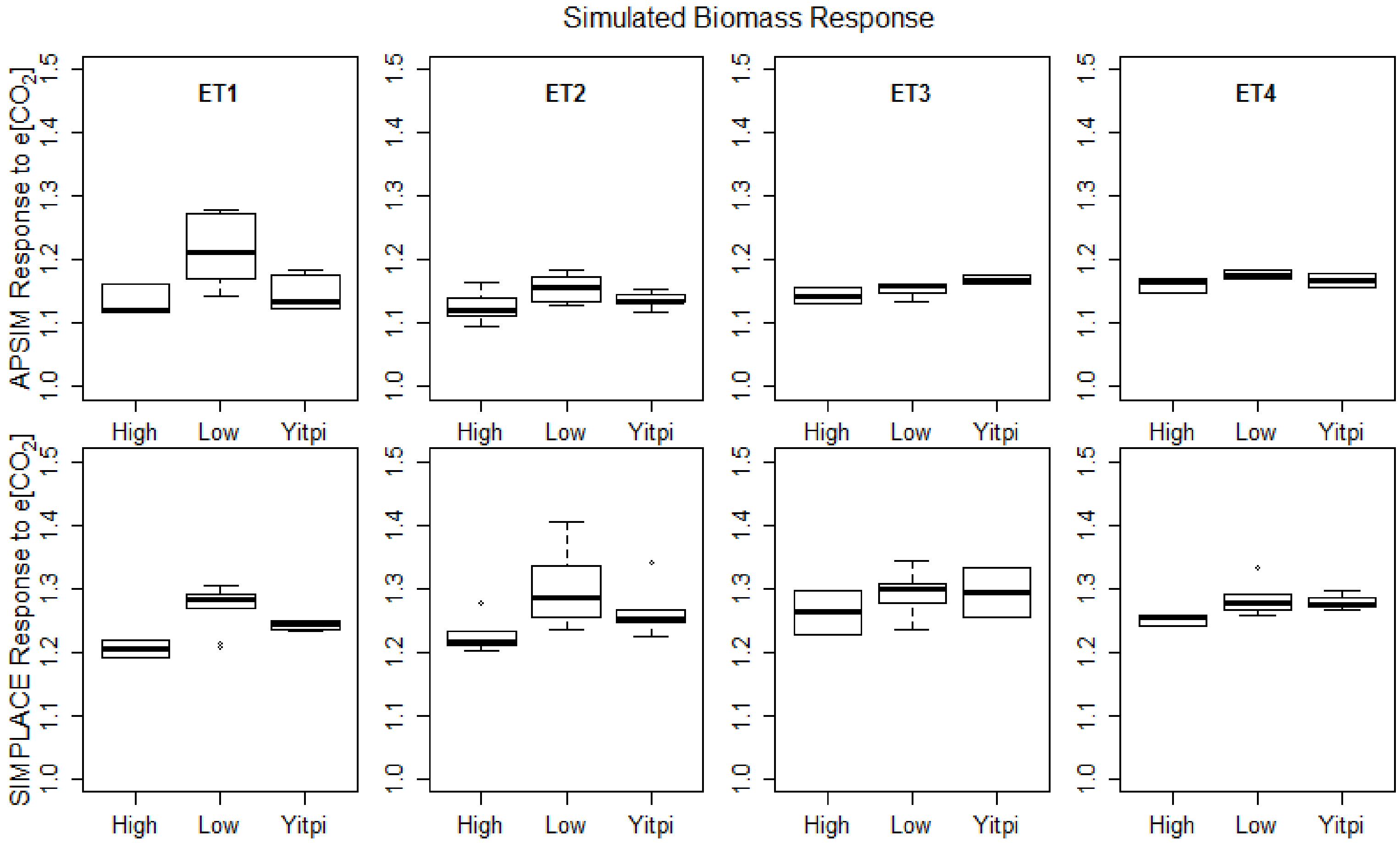


Figure S7: Correlations between relative simulated responses to e[CO_2] for pre-anthesis leaf area index and cumulative pre-anthesis water use (evaporation + transpiration), and cumulative pre-anthesis water use and average post-anthesis water stress index. For the water stress index, higher values refer to less stress with e[CO_2] compared to a[CO_2]. All high vigour lines are represented by the cultivar 'Hartog' as they did not differ in phenology or any other model parameters.

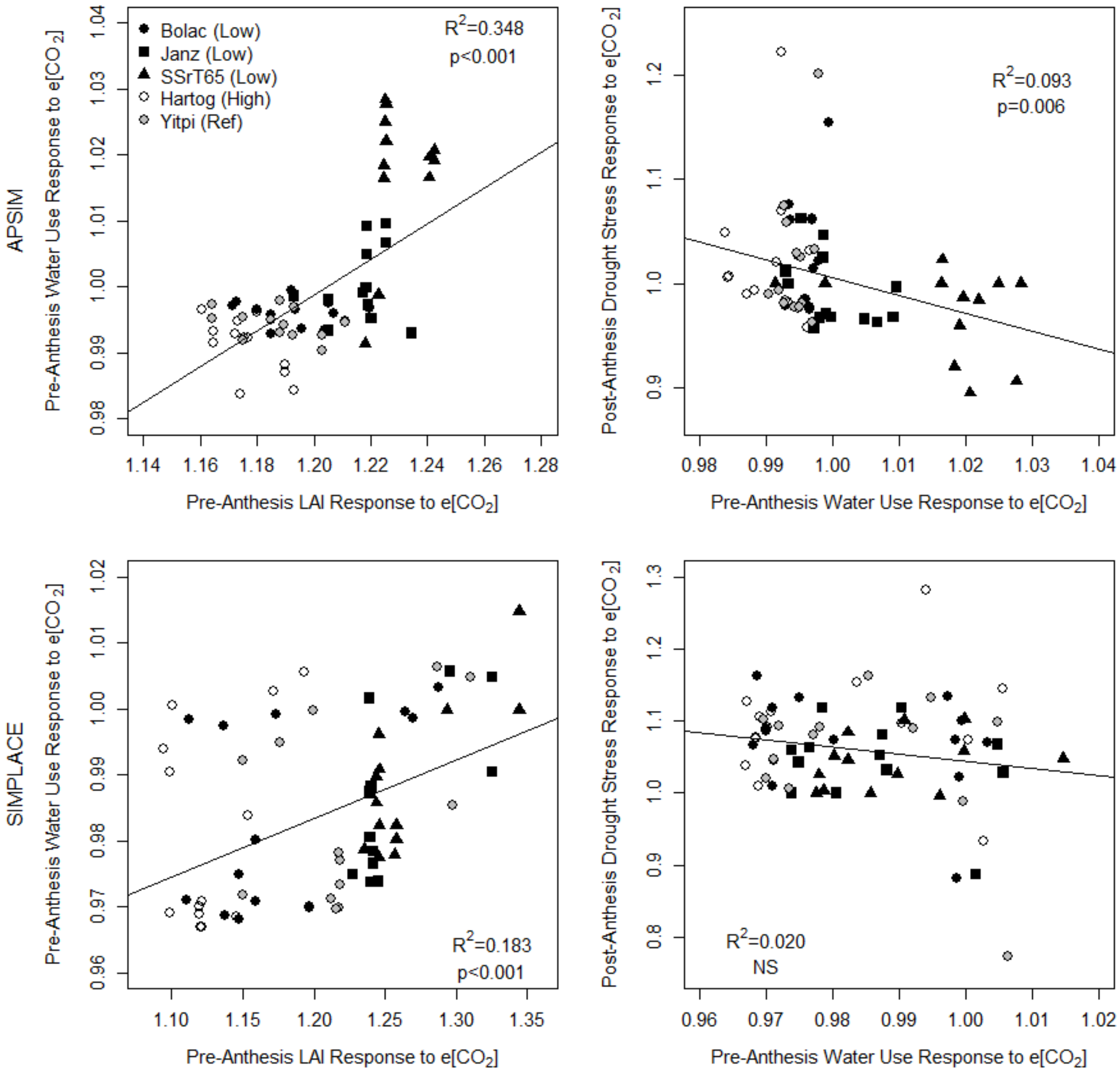


Figure S8: Correlations between relative simulated responses to e[CO_2] for average post-anthesis water stress index and cumulative pre-anthesis radiation intercepted with the simulated relative response in grain yield. For the water stress index, higher values refer to less stress with e[CO_2] compared to a[CO_2]. All high vigour lines are represented by the cultivar 'Hartog' as they did not differ in phenology or any other model parameters.

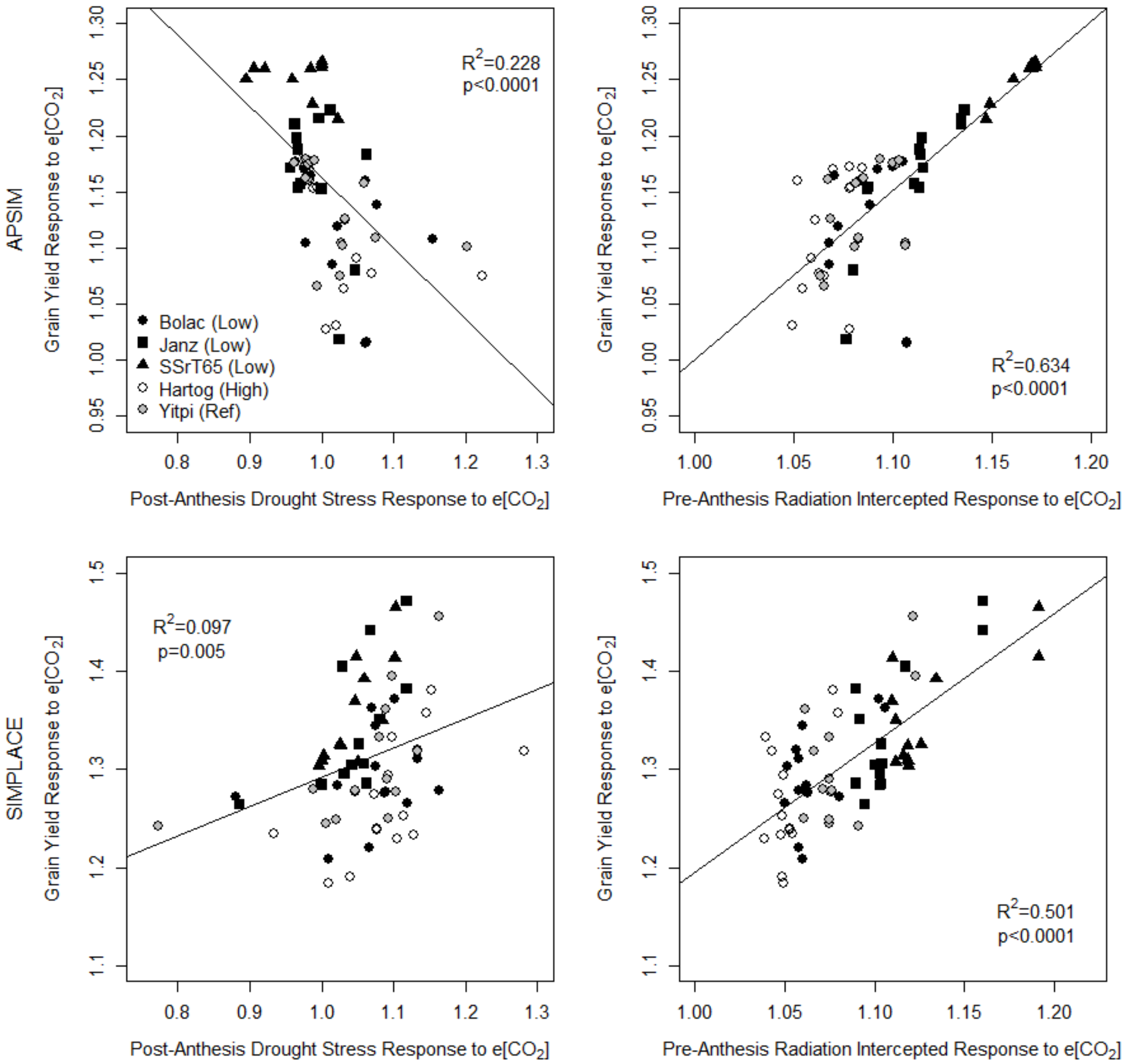


Table S1: Soil profile parameters describing air dry, crop lower limit (LL), drained upper limit (DUL), saturation (SAT), bulk density (BD), pH, and organic carbon concentration (OC) of the experimental site used by both models. The wheat module in APSIM also uses additional water extraction coefficients (KL and XF), and SIMPLACE uses van Genuchten parameters related to water retention in unsaturated soils (alfa and n).

Depth (cm)	Air dry (g/cm^3)	LL (g/cm^3)	DUL (g/cm^3)	SAT (g/cm^3)	BD (g/cm^3)	pH	OC (%)	APSIM KL	APSIM XF	SIMPLACE ALFA	SIMPLACE N
0-10	0.15	0.20	0.39	0.46	1.14	8.4	1.248	0.06	1.00	1	1.23
10-20	0.18	0.23	0.40	0.47	1.30	8.4	0.708	0.06	1.00	1	1.23
20-40	0.25	0.27	0.42	0.48	1.37	8.9	0.354	0.04	1.00	1	1.23
40-60	0.27	0.30	0.43	0.47	1.40	9.0	0.177	0.02	0.80	1	1.23
60-80	0.28	0.33	0.45	0.47	1.40	9.0	0.089	0.02	0.80	1	1.23
80-100	0.30	0.35	0.45	0.47	1.40	9.0	0.044	0.02	0.60	1	1.23
100-120	0.32	0.36	0.45	0.47	1.40	9.0	0.022	0.02	0.60	1	1.23
120-140	0.33	0.37	0.45	0.47	1.40	9.1	0.011	0.02	0.20	1	1.23
140-160	0.34	0.37	0.45	0.47	1.40	9.1	0.011	0.02	0.20	1	1.23
160-180	0.34	0.37	0.45	0.47	1.40	9.1	0.011	0.02	0.20	1	1.23

Table S2: Starting soil nitrate content used by both models for simulation of AGFACE experiments (2007-2013).

Depth (cm)	2007	2008	2009	2010	2011	2012	2013
0-10	42	34.8	13.3	38.4	47.2	25.7	19.3
10-20	25	26.6	26.1	23.8	23.3	14.6	8.4
20-40	15	21.4	27.5	24.3	12.7	11.2	11.4
40-60	13	21.4	27.5	22.8	12.7	11.2	11.4
60-80	6	11.8	10.5	22.8	20.4	20.1	15.2
80-100	2.5	11.8	10.5	11.7	20.4	20.1	15.2
100-120	2	2	2	11.7	2	2	2
120-140	1	1	1	1	1	1	1
140-160	1	1	1	1	1	1	1
160-180	0	0	0	0	0	0	0

Table S3: Starting soil water content used by both models for simulation of AGFACE experiments (2007-2013).

	2007	2008	2009	2010	2011		2012		2013	
Depth (cm)					Rain	Suppl	Rain	Suppl	Rain	Suppl
0-10	0.363	0.340	0.406	0.269	0.303	0.311	0.165	0.181	0.187	0.181
10-20	0.398	0.278	0.419	0.287	0.392	0.381	0.219	0.234	0.179	0.179
20-40	0.411	0.324	0.383	0.311	0.447	0.423	0.285	0.309	0.265	0.258
40-60	0.419	0.373	0.341	0.347	0.451	0.432	0.340	0.355	0.333	0.337
60-80	0.429	0.396	0.351	0.384	0.450	0.434	0.381	0.385	0.372	0.383
80-100	0.443	0.423	0.376	0.406	0.448	0.451	0.403	0.401	0.411	0.415
100-120	0.441	0.453	0.424	0.414	0.469	0.469	0.418	0.421	0.438	0.431
120-140	0.444	0.443	0.385	0.414	0.469	0.469	0.418	0.421	0.438	0.431
140-160	0.429	0.411	0.390	0.414	0.469	0.469	0.418	0.421	0.438	0.431
160-180	0.415	0.388	0.379	0.414	0.469	0.469	0.418	0.421	0.438	0.431

Table S4: Irrigation amounts and dates for the supplemental irrigation treatment used by both models for simulation of AGFACE experiments (2007-2013).

Year	Dates	Amount (mm)
2007	17 September	10
	24 September	10
	2 October	10
	8 October	10
	16 October	28
	14 November	28
2008	8 September	20
	25 September	20
2009	6 October	10
	22 October	30
	3 November	30
2010	6 October	30
	11 October	20
	22 October	30
2011	6 September	10
	12 September	20
	22 September	20
	10 October	20
	18 October	30
2012	11 September	30
	25 September	30
	10 October	30
	29 October	30
2013	No supplemental irrigation applied	

S5: Grain yield, above-ground biomass at maturity (DC90) and harvest index (HI) according to vigour group (high, low and the reference cultivar Yitpi) in the Australian Grains Free Air CO_2 Enrichment (AGFACE) facility in 2007-2013.

	High early vigour		Low early vigour		Yitpi	
	a[CO_2]	e[CO_2]	a[CO_2]	e[CO_2]	a[CO_2]	e[CO_2]
Grain yield (t ha-1)	4.30	5.25	4.12	5.23	4.66	5.72
Biomass (t ha-1)	11.7	14.6	11.1	14.0	12.5	15.9
HI (%)	36.2	35.0	37.0	36.9	36.8	35.8