

Elevated CO₂ and temperature increase soil C losses from a soybean–maize ecosystem

CHRISTOPHER K. BLACK^{1,2}, SARAH C. DAVIS^{2,3}, TARA W. HUDIBURG^{2,4},
CARL J. BERNACCHI^{1,2,5} and EVAN H. DELUCIA^{1,2}

¹Department of Plant Biology, University of Illinois at Urbana-Champaign, Urbana, IL 61801, USA, ²Carl R. Woese Institute for Genomic Biology, University of Illinois, Urbana, IL 61801, USA, ³Voinovich School of Leadership and Public Affairs, Ohio University, Athens, OH 45701, USA, ⁴Department of Forest, Rangeland, and Fire Sciences, University of Idaho, Moscow, ID 83844, USA, ⁵Global Change and Photosynthesis Research Unit, USDA-ARS, Urbana, IL 61801, USA

Abstract

Warming temperatures and increasing CO₂ are likely to have large effects on the amount of carbon stored in soil, but predictions of these effects are poorly constrained. We elevated temperature (canopy: +2.8 °C; soil growing season: +1.8 °C; soil fallow: +2.3 °C) for 3 years within the 9th–11th years of an elevated CO₂ (+200 ppm) experiment on a maize–soybean agroecosystem, measured respiration by roots and soil microbes, and then used a process-based ecosystem model (DayCent) to simulate the decadal effects of warming and CO₂ enrichment on soil C. Both heating and elevated CO₂ increased respiration from soil microbes by ~20%, but heating reduced respiration from roots and rhizosphere by ~25%. The effects were additive, with no heat × CO₂ interactions. Particulate organic matter and total soil C declined over time in all treatments and were lower in elevated CO₂ plots than in ambient plots, but did not differ between heat treatments. We speculate that these declines indicate a priming effect, with increased C inputs under elevated CO₂ fueling a loss of old soil carbon. Model simulations of heated plots agreed with our observations and predicted loss of ~15% of soil organic C after 100 years of heating, but simulations of elevated CO₂ failed to predict the observed C losses and instead predicted a ~4% gain in soil organic C under any heating conditions. Despite model uncertainty, our empirical results suggest that combined, elevated CO₂ and temperature will lead to long-term declines in the amount of carbon stored in agricultural soils.

Keywords: climate change, DayCent, priming, soil respiration, soybean free-air concentration enrichment, warming

Received 26 October 2015 and accepted 4 May 2016

Introduction

Human activity, primarily fossil fuel burning, is increasing atmospheric [CO₂] and raising global mean temperature (Hartmann *et al.*, 2013). These changes are likely to have direct and indirect effects on storage of soil organic carbon (SOC), but estimates of the direction and magnitude of these effects are poorly constrained (Dieleman *et al.*, 2012; Lu *et al.*, 2013). Soils worldwide store over two orders of magnitude more C than annual anthropogenic emissions (~1500 Pg C in the top 1 m; Eswaran *et al.*, 1993), so even small changes in soil C storage in response to climate change could produce large feedbacks to the global C cycle. This may be especially true of the SOC-rich former prairie soils of the agriculturally managed Midwestern United States, where annual tillage, infrequent water limitation, regular fertilization, and frequent pulses of highly labile C from crop residues provide ideal conditions for temperature-controlled microbial activity (Tisdall & Oades, 1980).

Correspondence: Evan H. DeLucia, tel. +1 (217) 333 6177, fax +1 (217) 244 7246, e-mail: delucia@illinois.edu

Changes in soil C are difficult to detect on short timescales because some pools turn over slowly, with mean residence times of hundreds of years. Although it is conceptually useful to identify the faster-cycling subpools of soil C, we lack experimental methods to measure them directly (Schmidt *et al.*, 2011). Instead, changes in the rate of CO₂ fluxes from soil can be used as a proxy for changes in the soil C cycle by partitioning total CO₂ flux (R_{tot}) into components attributed to ‘autotrophic’ respiration (R_{aut}) from plant roots and rhizosphere organisms, or to ‘heterotrophic’ respiration (R_{het}) from soil microbes in the process of breaking down soil organic matter (SOM). Because R_{het} is the primary avenue for loss of soil C, any change in R_{het} indicates a change in the rate of soil C loss. R_{het} is strongly controlled by soil temperature and moisture and therefore expected to shift under future climate conditions (Davidson & Janssens, 2006; Subke & Bahn, 2010; Conant *et al.*, 2011; Bradford, 2013). In contrast, changes in R_{aut} are indirectly linked to the rate of C input from roots, so a unit change in R_{tot} could indicate either increasing or decreasing soil C. Therefore, correct partitioning of fluxes is essential to their use as a

proxy for changes in pool size (Kuzyakov & Larionova, 2005).

Previous soil heating experiments have generally shown short-term increases in R_{tot} (Rustad *et al.*, 2001; Wu *et al.*, 2011), except when heating exacerbated soil water limitations (Schindlbacher *et al.*, 2012; Pendall *et al.*, 2013; Suseela & Dukes, 2013; Wall *et al.*, 2013; Wang *et al.*, 2014). This heating effect often diminishes after a few years of treatment. Whether these responses will persist over the long term under climate change depends on whether a particular soil's R_{het} response is modulated by availability of nutrients or C substrates (Luo *et al.*, 2001; Chevallier *et al.*, 2015) or by physiological adaptation of the microbial community (Allison *et al.*, 2010; Bradford, 2013). In addition, few of these studies were able to separate soil respiration into its autotrophic and heterotrophic components. As R_{het} is strongly controlled by thermal kinetics while R_{aut} responds to a wide variety of nonthermal factors, it has been widely assumed that temperature-associated increases in R_{tot} are driven by increasing R_{het} , but support for this assumption is equivocal (Hartley *et al.*, 2007; Bond-Lamberty & Thomson, 2010; Suseela & Dukes, 2013; Wang *et al.*, 2014).

Previous CO₂ enrichment experiments have generally shown sustained increases in R_{tot} (King *et al.*, 2004; Bernhardt *et al.*, 2006; Pregitzer *et al.*, 2006; Peralta & Wander, 2008; Adair *et al.*, 2011; Carrillo *et al.*, 2011; Drake *et al.*, 2011; Keidel *et al.*, 2015), but there are few reported results from field experiments that manipulate both heat and CO₂ simultaneously. Of those that are reported (Carrillo *et al.*, 2011; Pendall *et al.*, 2011, 2013; Selsted *et al.*, 2012), the observed responses seem to be mostly mediated by water availability, with heat increasing R_{tot} when moisture is available and reducing it when heating produces drier soil. Elevated CO₂ mediates these effects by ameliorating soil water stress through increased plant water use efficiency, but the strength and predictability of this effect seem to vary widely both within and between experiments.

The objective of this study was to measure the root- and SOM-derived components of soil respiration in an intact maize–soybean ecosystem subjected to mid-21st century temperature (+3.5 °C) and CO₂ (585 ppm) conditions under fully open-air conditions at SoyFACE (Urbana IL, USA). We then used a process-based biogeochemical model (DayCent; Parton *et al.*, 1998) to predict the long-term effects of these respiratory responses on soil C storage. We predicted that elevated temperature would increase the activity of soil heterotrophs, leading to increased respiration in root-free soil and long-term losses of C from the most labile pools of SOM. We further predicted that elevated CO₂ would increase plant biomass above- and belowground,

leading to higher C inputs that would at least partially ameliorate the long-term effect of heat on soil C, and therefore that the long-term fate of soil C at our site would depend on the strength of the interaction between heat and CO₂ effects.

Materials and methods

Site description

Measurements were made at the soybean free-air concentration enrichment (SoyFACE) experiment (40.04N, 88.23W; elev. 215 m; soyface.igb.illinois.edu), a 32-Ha experimental site near Urbana IL, USA. The site is flat, tile-drained, and has been cultivated for over 100 years. Soils are deep and highly productive, mapped as Flanagan silt loam (fine, smectitic, mesic Aquic Argiudoll) and Drummer silty clay loam (fine-silty, mixed, superactive, mesic Typic Endoaquoll). The mean annual temperature is 11 °C, with monthly mean temperatures ranging from −3 °C in January to 24 °C in July, and annual precipitation is ~1 m, with approximately half falling during the May–September growing season (Angel, 2010a).

The site was managed in a 2-year rotation, with maize (*Zea mays* cv 34B43) and soybean (*Glycine max* cv 93B15) alternating between the eastern and western halves of the site. Maize was fertilized with 202 kg N Ha^{−1} yr^{−1} and soybean was not fertilized. The soil was typically chisel plowed each spring before planting, and in the fall after maize harvest but not after soybean. Measurements were taken from 2009 to 2011 in the west half of the site, where the crop rotation cycle for these years was soybean–maize–soybean. Management of these crops was consistent with previously reported practices at the site (Leakey *et al.*, 2004; Morgan *et al.*, 2005), with the exception that no fall tillage was carried out after the 2010 maize crop so that the heating equipment could be kept in operation overwinter.

Elevated CO₂ and temperature treatments

The field was divided into four experimental blocks, each containing two 20-m octagonal rings spaced 100 m apart to avoid cross-contamination by fumigation gases. One ring in each block was maintained at ambient atmospheric conditions (approximately 390 ppm CO₂), while the other was fumigated to a target of 585 ppm CO₂ using Free-Air Concentration Enrichment (FACE) technology (Miglietta *et al.*, 2001). Elevated CO₂ was maintained from dawn until dusk throughout the growing season. The high-CO₂ rings in the study area have been fumigated every growing season since 2001.

Starting in 2009, each ring was further split by imposing an elevated temperature treatment. One 3-m hexagonal subplot of each ring was equipped with overhead infrared heaters as in Kimball (2005). The heaters were adjusted throughout the growing season to stay 1.4 m above the canopy and were controlled by infrared radiometers to maintain a difference of 3.5 °C between the canopy-top temperature of heated and unheated plots. When rain was falling and when soil temperature was ≤5 °C, heating was ineffective and heater output was

therefore reduced to a minimum. The heating system operated continuously between June 2009 and September 2011 except during planting when all equipment was removed to allow field tillage, during harvest when power cables were removed to allow harvester traffic, and during a 2-week period in January 2010 when the equipment was rebuilt to repair damage from rodents. Further details on the heating treatment are reported in studies by Ruiz-Vera *et al.* (2013, 2015) and Rosenthal *et al.* (2014).

Measurement of soil properties and CO₂ efflux

CO₂ efflux from soil was measured at three locations in each plot using 20-cm-diameter collars made from PVC pipe. At each location, one collar was inserted 3 cm into the soil to capture total soil respiration (R_{tot}), and a second collar was inserted 25 cm to capture respiration by soil heterotrophs (R_{het}) by excluding roots and rhizosphere: The top 30 cm of soil contains at least 70% of soybean and 60% of maize root mass (Mayaki *et al.*, 1976; Anderson-Teixeira *et al.*, 2013), so this root-exclusion collar acts as a small trenched plot (Vogel & Valentine, 2005). Collars were installed at crop emergence time each spring and left in place all year then removed for field tillage just before the next year's planting. This annual reinstallation also eliminated several major limitations that apply to root-exclusion methods in untilled systems: It removed any accumulated difference in C or nutrient availability from previous years of root exclusion, and there was no need to correct for decomposition of roots severed during installation (Hanson *et al.*, 2000) because root biomass at installation was near zero. Respiration by roots and rhizosphere (R_{aut}) was calculated for each location as the difference between R_{tot} and R_{het} .

CO₂ efflux rates were measured using an infrared gas analyzer (LI-8100; Li-Cor, Lincoln, NE, USA) fitted with a 20-cm static chamber (Li-Cor 8100-103) that rested on top of the soil collar. For each observation, the chamber was closed for two minutes while [CO₂] was logged every second. Linear regressions on static chamber observations underestimate the initial flux rate (Healy *et al.*, 1996), so flux rates were computed in software by the LI-8100, which fit a saturating exponential curve of the form:

$$C_t = C_\infty + (C_0 - C_\infty)e^{-a(t-t_0)}$$

where C_0 is [CO₂] at the moment the chamber closed, t is time, and a and C_∞ are fitted parameters representing curvature and [CO₂] at the asymptote, respectively. Evaluating the derivative of C_t at $t = 0$ then gives the instantaneous initial slope $a(C_\infty - C_0)$, which was scaled by the volume of the soil chamber to give CO₂ flux rate at the moment the chamber closed.

Using exponential rather than linear fits is especially important in a FACE setting, because it allows a further correction for initial chamber conditions: Respiration was measured while fumigation was active. Pure CO₂ is released from the upwind side of the ring and mixes to the target concentration as it is blown across the plot (Miglietta *et al.*, 2001), meaning that in elevated-CO₂ plots, the flux chamber would sometimes close on a transient high-[CO₂] air mass (up to 2000 ppm). In

these cases, the CO₂ concentration gradient from soil to chamber air, and thus the rate of diffusion across the soil surface, was small. This meant that for these readings, the fitted flux rate 'at the moment the chamber closed' was much smaller than the true equilibrium rate. We corrected this bias using a method recommended by Li-Cor Inc. that re-evaluates the previously fit [CO₂] curve to find the equilibrium flux $a(C_\infty - C_{\text{target}})$, where a and C_∞ are taken from the previous curve fit and C_{target} is the daily average [CO₂] in that ring (585 ppm for fumigated rings, 370–400 ppm for unfumigated rings).

Soil temperatures were measured simultaneously with each respiration measurement using a thermocouple probe inserted to 5 cm depth. Soil volumetric water content was measured from 5 to 105 cm depth 2–3 times each week using a capacitance probe and is reported elsewhere (Rosenthal *et al.*, 2014; Ruiz-Vera *et al.*, 2015).

Particulate organic matter (POM), which consists of fragmented but undecomposed plant matter and is used as a proxy for the abundance of labile soil C, was measured using a procedure modified from Marriott & Wander (2006). Briefly, air-dried soil was sieved to 2 mm and a 10-g sample was weighed into a 30-mL plastic bottle. The mouth of the bottle was covered with a 53- μm nylon mesh to retain POM and sand while allowing silt and clay particles to escape. The bottle was submerged in 5% sodium hexametaphosphate (HMP) and shaken for one hour, then the HMP and suspended fines <53 μm were removed, replaced with deionized water, and shaking was repeated until no further fine material was extracted. The remaining POM + sand was transferred to a pouch of 53- μm mesh, rinsed with DI water, dried at 30 °C, and weighed. Samples were then ground in a ball mill (Geno Grinder 2010; BT&C, Lebanon, NJ, USA) and combusted to determine C content using an elemental analyzer (Costech ECS4010; Costech Analytical Technologies, Valencia, CA, USA).

Statistical analysis

Analysis of variance for soil respiration was performed in a complete-block design using CO₂ as a whole-plot fixed effect and heat as a split-plot fixed effect nested within CO₂. Blocks were treated as random, and autocorrelation within plots from repeated measurement through the season was estimated as a first-order autoregressive function. R_{tot} , R_{het} , and R_{aut} fluxes for each season were analyzed separately as mixed-effects linear models with repeated measures using the nlme and lsmeans packages in R 3.2.4 (Lenth, 2016; Pinheiro *et al.*, 2016; R Core Team, 2016). The date of each survey event was included as a categorical variable to account for within-season changes. Although most of the temporal variation is likely to be driven by weather and crop growth phase, the day effect was treated as a catchall term and no explicit temperature or moisture covariates were included in the model. Because repeated measurements within the same plot are pseudoreplicates, the three flux measurements from each plot were averaged, giving $n = 4$ observations per treatment in each day. Because experiments with few replicates have low power to

detect small differences, we set a significance threshold of $P \leq 0.1$ to minimize the chance of false-negative conclusions (Filion *et al.*, 2000). Full statistical output and data-processing scripts are available in the data package for this manuscript (Black *et al.*, 2016).

Modeling of soil respiration and soil organic carbon

Because a 3-year heating experiment is likely too short to detect changes in SOC, we performed an *in silico* experiment using a process-based ecosystem model (DayCent; Parton *et al.*, 1998) to simulate the effects of a 100-year global change manipulation and better understand the long-term effects of elevated CO₂ and temperature on soil carbon dynamics. DayCent has been widely used to model soil C, N, P, and S dynamics and trace gas fluxes. It has been particularly well validated for crop and grassland systems and is straightforward to modify for predicted future conditions, making it ideal for simulations of the future ecosystem effects of climate and/or land-use changes (Davis *et al.*, 2010, 2012; Hartman *et al.*, 2011). DayCent model development has been closely tied to previous global change experiments and its input parameters are designed for easy calibration against experimentally measured responses (Parton *et al.*, 2007; Frey *et al.*, 2013).

To predict the medium- and long-term effects of ongoing ecosystem warming and elevated CO₂ on soil carbon cycling, we performed a three-part set of DayCent simulations to simulate the historic development of the SoyFACE site from native prairie into a maize–soybean rotation, extended this simulation through the 21st century, then ran the model four times using all factorial combinations of elevated CO₂ and heat.

To calibrate the size and turnover rates of soil C pools, the model was first run to equilibrium by simulating a native tallgrass prairie at preindustrial [CO₂] of 294 ppm. Each simulation lasted 3867 years and looped over a weather file made by randomly ordering the years of an 1889–2009 temperature and precipitation record for Urbana, Illinois (Angel, 2010b). Vegetation for the spin-up period used prairie grass parameters provided by Hudiburg *et al.* (2015), with autumn burning every 5th year and low-intensity grazing by bison (10% of foliage removed three times per growing season). Soil parameters were based on bulk densities and organic matter contents of undisturbed Illinois prairie remnants (David *et al.*, 2009), and on physical properties of the Flanagan and Drummer soil series (NRCS, 2012). To match the high-moisture conditions predominant in central Illinois before the introduction of artificial drainage, a standing water table was simulated from January through May. Soil organic matter turnover times were adjusted to produce steady-state (<1% change per decade in last 100 years) SOM C and N of 10 450 and 760 g m⁻², respectively, in the top 20 cm (Fig. S1). These totals are comparable to those measured in tallgrass prairie remnants on deep, mesic soils throughout the Midwest (Aref & Wander, 1998; Kucharik *et al.*, 2006; Matamala *et al.*, 2008; Brye & Riley, 2009; David *et al.*, 2009; Jelinski & Kucharik, 2009) and were achieved using turnover rates for the active, intermediate, and slow soil

C pools of 11, 0.1, and 0.002 yr⁻¹, giving residence times of 33 days, 10 years, and 500 years, respectively.

Annual row crops were simulated beginning in 1868, the year our site was first recorded as occupied by European settlers. To simulate the change from an untilled, seasonally wet prairie to a tile-drained, annually-tilled crop system, we ceased simulating a standing water table, increased the maximum decomposition rate of intermediate- and slow-turnover organic matter, and reduced leaching rates for N and OM (Table S1). Additionally, we reduced the rate of nonsymbiotic soil N fixation and the fraction of mineralized N lost to nitrification to better match conditions observed in row crop systems (Table S1). Site-specific parameters were based on soil conditions measured at the site (Peralta & Wander, 2008; C. Black, unpublished data; J. Jastrow, unpublished data; Moran & Jastrow, 2010), soil moisture measured at the site (S.B. Gray, unpublished data; Rosenthal *et al.*, 2014; Ruiz-Vera *et al.*, 2015), and historical weather data from the Illinois State Water Survey (Angel, 2010b). Atmospheric [CO₂] was increased linearly to match the rise in industrial fossil fuel burning, from 294 ppm in 1868 to 370 ppm in 2000. Crop-specific parameters for maize and soybeans were developed by Hudiburg *et al.* (2015) to match the rate and physiological mechanisms of 20th-century crop genetic improvements: Maize yield gains have come mostly from increases in planting density and photosynthetic capacity (Duvick, 2005), so we simulated an increase in the maximum daily biomass production rate, with minor adjustments to other parameters (Table S3). In contrast, soybean yield increases have come mostly from improved yield partitioning at constant plant size (Koester *et al.*, 2014), so our soybean parameters were constant except that we increased the maximum harvest index in 1950 and 1980.

The management history of the site before 1980 was inferred from records of crop acreage and fertilizer usage for Champaign County retrieved from the National Agricultural Statistics Service (NASS, 2011). Site management since 1980 is well described (Moran & Jastrow, 2010) and was simulated accordingly (Table S2). Briefly, management progressed from low-yielding mixed maize/oat/pasture in 1869 through increasingly intensive cropping and fertilization to a maize–soybean–oat rotation by 1935 and a maize–soybean rotation by 1950, with fertilization rates and cultivar parameters adjusted each decade to match NASS records. Beginning in 1970, we changed cultivation from moldboard plow every spring and fall to chisel plowing each spring and in fall after maize only, and fertilization rates were held steady at 157 kg N Ha⁻¹. This management schedule was continued although 1999 then concluded in 2000 with 1 year of winter wheat (Moran & Jastrow, 2010).

To simulate the SoyFACE climate change manipulations, we extended the 20th-century simulation for the years 2001–2109 using the actual 2001–2011 planting and harvest dates of the SoyFACE experimental field. Weather data for 2001–2011 were retrieved from DAYMET (Thornton *et al.*, 2014) and the model was run four times: a control run with actual weather conditions and [CO₂] set to 370 ppm (ambient conditions at the initiation of the SoyFACE experiment), a CO₂-only run with [CO₂] increased by 200–570 ppm as a step

change in 2001, a heat-only run with daily maximum and minimum temperatures increased 3.5 °C as a step change in 2009, and a heat + CO₂ run with both temperature and CO₂ increased. Note that we did not simulate any further increase in [CO₂] after the step change, so for all model–data comparisons, we treated values modeled at 370 or 570 ppm as equivalent to field values observed in 2009–2011 at ~390 or 585 ppm.

Our model calibration strategy was to use the performance of our spin-up and historic hindcast scenarios as indicators of correct parameter calibration and then run the climate change scenarios with no further changes in model tuning. To the extent that model hindcasts do match known conditions, we gain confidence that model predictions for the future are reasonable. To evaluate model performance in more detail, we compared modeled soil temperature, moisture, and respiration rates against our 2009–2011 field observations. We also compared modeled aboveground biomass and grain yields for 2001–2008 against detailed phenological measurements from SoyFACE, using a database compiled by Twine *et al.* (2013). All model parameters and analysis scripts are available online (Appendix S1; Black *et al.*, 2016).

Results

Temperature and CO₂ manipulation

Infrared heating produced a mean temperature increase of approximately 3.1, 2.7, and 2.6 °C at the top of the canopy in 2009, 2010, and 2011, respectively, and CO₂ fumigation consistently maintained ~585 ppm CO₂ during daylight hours (Ruiz-Vera *et al.*, 2013, 2015). Soil temperature at 5 cm depth was increased by 1.8 ± 0.2 °C (mean ± standard error of daily differences) during the growing seasons (Fig. 1). During fallow seasons, heater output was intermittently reduced during extreme cold snaps (<16% of total time) but soil temperature for the whole season was still increased by 2.3 ± 0.1 °C (Fig. 1). There was no consistent difference

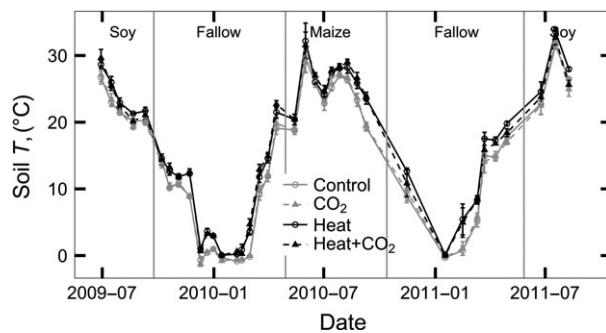


Fig. 1 Temperature measured 5 cm below the soil surface under heating and free-air CO₂ enrichment between June 2009 and October 2011. Error bars show treatment means ± 1 standard error for each day.

in soil temperature between eCO₂ and unfumigated plots given the same heat treatment.

Soil respiration

Overall, heating caused a consistent and large increase in R_{het} but reduced R_{aut} by a similar degree, producing no appreciable net effect of heating on R_{tot} , while eCO₂ increased R_{het} and affected R_{aut} differently each year, with the net effect of a small stimulation in R_{tot} from eCO₂. Averaged across the entire experiment, R_{het} was higher than control by 16%, 12%, and 48% in the eCO₂, heat, and heat + CO₂ treatments, respectively. R_{aut} was slightly (3%) higher in eCO₂ and lower in heated plots by 21% (heat) and 31% (heat + eCO₂). R_{tot} was higher in eCO₂ treatments by 11% (unheated eCO₂) and 13% (heated eCO₂) but 3% lower in the heated ambient CO₂ treatment (Fig. 2).

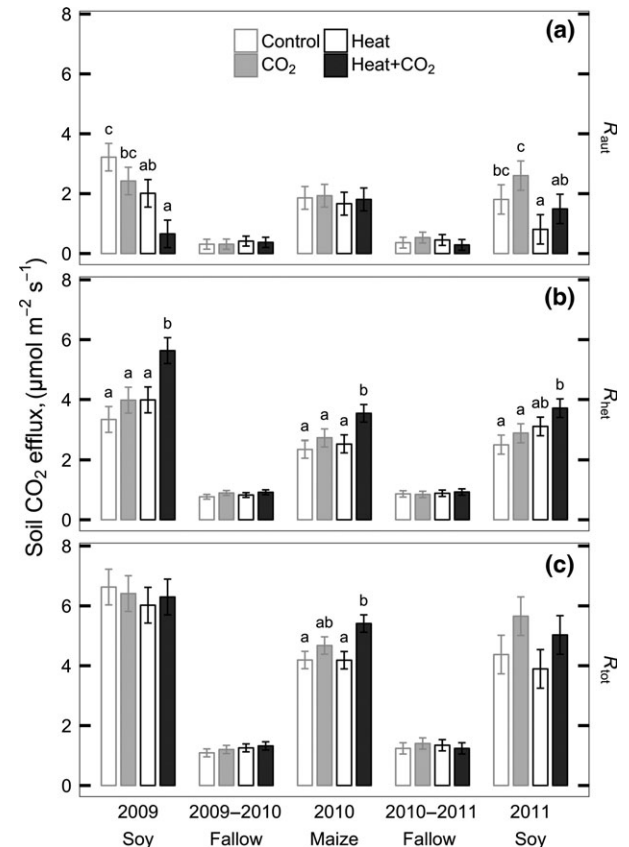


Fig. 2 Seasonal means of CO₂ flux measured from plant roots and rhizosphere (R_{aut} : a), soil heterotrophs (R_{het} : b), and whole soil (R_{tot} : c) under heating and free-air CO₂ enrichment between June 2009 and October 2011. Error bars show treatment LS means ± 1 standard error for each season. Within each season, treatments that share a letter are not statistically different ($P > 0.1$).

Separate mixed-model analyses of respiration from each season (Table 1) showed that under soybeans in 2009, R_{tot} was unchanged while R_{het} increased and R_{aut} decreased in both heat and eCO₂ treatments. Under maize in 2010, CO₂ increased R_{tot} , heat increased R_{het} and there were no differences in R_{aut} between treatments. Under soybeans in 2011, there were no differences in R_{tot} between treatments while R_{het} was higher and R_{aut} was lower in heated plots. R_{het} also showed a three-way interaction between heat, CO₂, and day, with higher R_{het} from heated eCO₂ plots on June 24 and July 18 but no statistical difference between treatments on the other days of the season (Fig. 3). During the fallow period following soybeans (winter 2009–2010), R_{aut} and R_{tot} did not differ between treatments while R_{het} was higher in eCO₂ plots and had an interactive effect with heat and day, with a trend ($P < 0.12$) for higher R_{het} from heated plots on October 7 and December 31, lower R_{het} from heated plots on December 10, and no statistical difference on the other days. During the fallow period following maize (winter 2010–2011), no component of soil respiration (R_{tot} , R_{het} , R_{aut}) differed between treatments. The main effect of day was significant in all treatments every season, while the heat by CO₂ and CO₂ by day interactions were never significant.

Particulate organic matter

Particulate organic matter carbon (POM-C) declined from the beginning to the end of the experiment (2009 > all other harvests; Tukey HSD $P < 0.01$; Fig. 4) and was approximately 14% lower in eCO₂ plots than in ambient plots (ANOVA $F = 7.69$, $P < 0.01$; Fig. 4), but showed no statistically resolvable difference between heated and unheated plots (ANOVA $F = 0.29$, $P > 0.5$). Averaged across all treatments, the top 30 cm of soil contained 588 ± 41 g POM-C m⁻² (mean \pm SE) in spring 2009, 439 ± 21 in spring 2010, 444 ± 25 in spring 2011, and 457 ± 22 in fall 2011.

DayCent model

DayCent simulations of 20th-century grain yields of maize and soybeans agreed well with historic crop yields from Champaign County and captured about half of the observed year-to-year variation in yield (Figs S2 and S3; root-mean-square error = 82.6 g C m⁻², RMSE/mean = 0.53). Modeled total C and N in soil organic matter at the end of the historic agriculture scenario were both very near the values measured at SoyFACE (Fig. S1). During the 2001–2008 CO₂ simulation,

Table 1 Mixed-effects model results for rates of soil CO₂ efflux attributed to soil heterotrophs (R_{het}), plant roots and rhizosphere organisms (R_{aut}), and whole soil (R_{tot})

Component	Heat	CO ₂	Heat \times CO ₂	Heat \times Day	CO ₂ \times Day	Heat \times CO ₂ \times Day	Heat % change	CO ₂ % change
Soy 2009								
R_{aut}	0.014	0.089	0.552	0.737	0.717	0.988	-53 \pm 15	-41 \pm 17
R_{het}	0.035	0.068	0.271	0.790	0.495	0.724	32 \pm 11	31 \pm 11
R_{tot}	0.475	0.920	0.636	0.859	0.389	0.932	-6 \pm 7	0 \pm 8
Fallow 2009–2010*								
R_{aut}	0.465	0.915	0.845	0.909	0.922	0.899	26 \pm 50	-6 \pm 43
R_{het}	0.875	0.097	0.879	0.053	0.775	0.934	5 \pm 7	14 \pm 7
R_{tot}	0.315	0.424	0.776	0.869	0.915	0.901	12 \pm 12	8 \pm 12
Maize 2010								
R_{aut}	0.716	0.806	0.917	0.561	0.317	0.277	-8 \pm 19	6 \pm 20
R_{het}	0.053	0.137	0.181	0.704	0.632	0.931	20 \pm 8	28 \pm 15
R_{tot}	0.263	0.059	0.226	0.687	0.642	0.605	8 \pm 7	20 \pm 7
Fallow 2010–2011								
R_{aut}	0.532	0.796	0.216	0.940	0.339	0.231	-17 \pm 30	1 \pm 43
R_{het}	0.740	0.800	0.687	0.636	0.310	0.752	6 \pm 12	1 \pm 12
R_{tot}	0.771	0.953	0.381	0.977	0.204	0.117	-2 \pm 11	2 \pm 17
Soy 2011								
R_{aut}	0.018	0.190	0.864	0.690	0.794	0.381	-48 \pm 15	57 \pm 33
R_{het}	0.047	0.224	0.700	0.801	0.739	0.091	27 \pm 11	18 \pm 12
R_{tot}	0.131	0.252	0.824	0.906	0.822	0.997	-11 \pm 6	29 \pm 21

The first six columns show P values for each effect. Boldface values are significant at a preselected threshold of 0.1. The main day effect was always significant (all $P < 0.02$) and is not shown here to save space. The last two columns show percent change from control for each treatment, presented as the estimated differences \pm 1 standard error of whole-season LS means.

*Data from March 1, 2010 were excluded from the model because they contained no usable observations from heated plots.

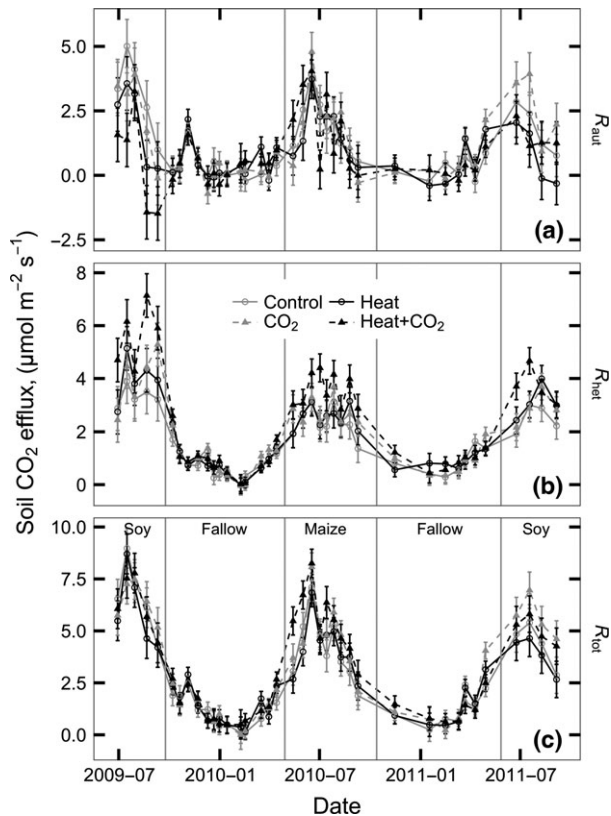


Fig. 3 CO₂ flux measured from plant roots and rhizosphere (R_{aut} ; a), soil heterotrophs (R_{het} ; b), and whole soil (R_{tot} ; c) under heating and free-air CO₂ enrichment between June 2009 and October 2011. Each season was analyzed separately; vertical gray lines indicate cutoffs between seasons. Error bars show treatment LS means ± 1 standard error for each day.

the temporal dynamics of modeled aboveground biomass within each season matched well with values observed at SoyFACE in those years (Fig. S4). Observed soybean grain yields at the site averaged $191 \pm 30 \text{ g C m}^{-2}$ in ambient plots and 212 ± 39 in eCO₂ plots (Twine *et al.*, 2013); modeled yields for the same years were 194 ± 65 and $254 \pm 75 \text{ g C m}^{-2}$, respectively. Observed maize yields averaged $423 \pm 15 \text{ g C m}^{-2}$ in ambient plots and 412 ± 44 in eCO₂ plots (Leakey *et al.*, 2006; Markelz *et al.*, 2011; Ruiz-Vera *et al.*, 2015); modeled yields for the same years were 432 ± 38 and $460 \pm 22 \text{ g C m}^{-2}$, respectively.

Modeled effects on soil temperatures were somewhat higher than the observed differences, with heated model runs $4.1 \pm 0.6 \text{ }^\circ\text{C}$ (mean \pm SD) warmer than unheated runs at 5 cm depth during the growing season, while observed differences were $<2 \text{ }^\circ\text{C}$. Additionally, modeled soil temperature differences dropped to $3.5 \text{ }^\circ\text{C}$ before planting and after harvest, while observed differences were larger than during the growing season

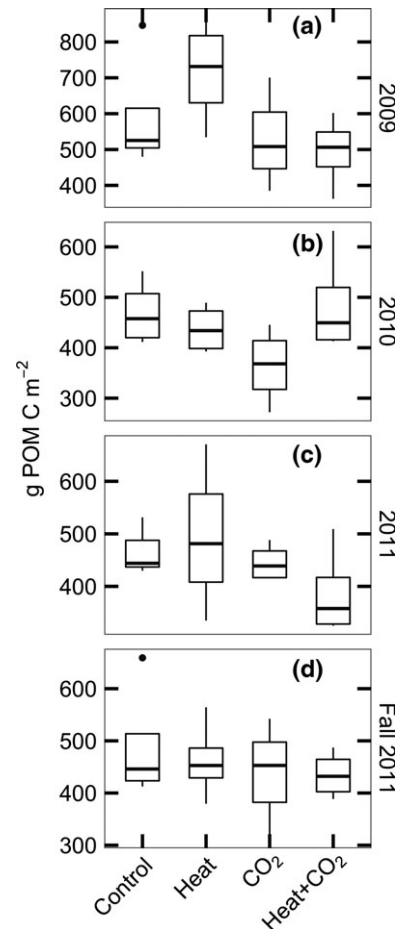


Fig. 4 Particulate organic matter carbon (POM-C) in the top 30 cm of soil under heating and free-air CO₂ enrichment, sampled in spring of 2009 (a), 2010 (b), 2011 (c), and at the end of the experiment in fall 2011 (d). Boxes cover the estimated interquartile range of each group, and whiskers extend to the smaller of max/min or 1.5 IQR.

($\sim 2.5 \text{ }^\circ\text{C}$; Fig. 1). Modeled temperatures in elevated CO₂ model runs were $0.5 \pm 0.4 \text{ }^\circ\text{C}$ lower than those in ambient runs, but no consistent differences were observed in the field (Fig. 1).

Compared to the values observed in the field, DayCent captured the seasonal variation and relative timing in all components of soil respiration, matched its magnitude well for R_{het} and consistently underpredicted R_{aut} producing a smaller overall under prediction of R_{tot} (Fig. S5).

When the simulation was extended to 100 years of heating and elevated [CO₂], DayCent predicted that CO₂ would increase all soil C pools, producing an increase of about $\sim 4\%$ in total SOM C (Fig. 5). In contrast, heating was predicted to produce a rapid drop in all pools that overwhelmed the increased C inputs from eCO₂, producing a loss from heated runs, relative to the

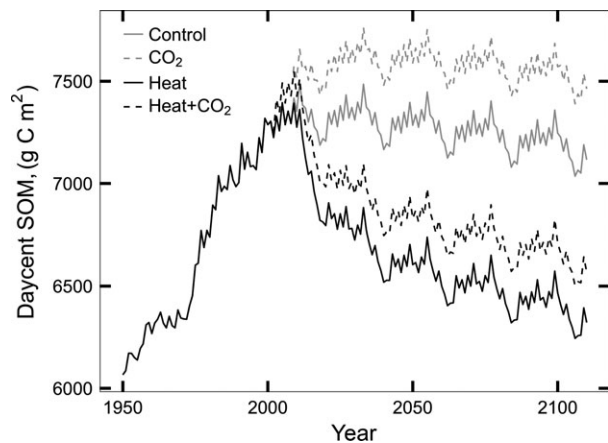


Fig. 5 Mean annual values for total soil organic matter (g C m^{-2}) predicted by DayCent in the top 20 cm of soil exposed to heating and CO_2 enrichment.

control scenario, of 15% of total soil C in the top 20 cm by 2109 (Fig. 5).

Discussion

Experimental manipulation of CO_2 and temperature conditions similar to those expected for the mid-21st century increased respiration from soil microbes, likely indicating a drawdown of both labile and protected soil C. This effect would not have been detectable without partitioning respiration into its root and microbial components, and it was not offset by the observed positive effect of CO_2 on plant productivity. Instead, CO_2 increased respiration and reduced POM-C, and we saw no evidence of interactions between heat and CO_2 responses, leading us to speculate that eCO_2 primed long-term losses of SOC. Our model results adequately captured the effects of temperature and the additive effect of the temperature and CO_2 responses but did not reproduce a CO_2 priming effect, so actual soil C losses may exceed our model-predicted value of 15% in 100 years.

Our results support the prediction that elevated temperature would increase the activity of soil heterotrophs and that this increased respiration would lead to long-term losses of soil C. Three years of heating produced strong and persistent increases in R_{het} (Fig. 2; Table 1) and our model results are consistent with these increases leading to losses of C from all soil pools (Fig. 5). Because heating reduced R_{aut} simultaneously with increasing R_{het} , there was no change in total soil respiration with heat (Fig. 2; Table 1). This result contrasts with many previous studies (reviewed in Rustad *et al.*, 2001; Wang *et al.*, 2014) where R_{tot} increased under heating except under water limitation, but this

discrepancy is explained by partitioning fluxes into their autotrophic and heterotrophic components.

Consistent with recent meta-analyses (Dieleman *et al.*, 2012; Wang *et al.*, 2014), we found that R_{het} responded reliably to increased temperature even when opposing changes in R_{aut} masked its effect on R_{tot} . This highlights the importance of separating soil CO_2 fluxes in global change experiments into their root and microbial components. We speculate that this masking may also occur at other sites where R_{tot} was measured without partitioning and found unresponsive to heat, especially those in grasslands and crops where root activity seems less responsive to eCO_2 than in forest systems (Wang *et al.*, 2014).

Our results did not support the prediction that elevated CO_2 would increase plant biomass above- and belowground, leading to higher C inputs that would at least partially ameliorate the long-term effect of heat on soil C. Although aboveground plant biomass was higher in eCO_2 plots during soybean years, heating largely negated this difference (Ruiz-Vera *et al.*, 2013) and there was little difference in root mass (S.B. Gray, in prep). R_{het} increased more in heat + eCO_2 plots than in plots given heat alone, indicating increased respiratory losses. Meanwhile R_{aut} , a probable correlate of C inputs from root exudation and turnover, was lower in eCO_2 plots in 2009 and showed no detectable change in other years, and the change in R_{aut} between heat and heat + eCO_2 plots was similar to that between unheated control and eCO_2 plots. One possible explanation for these findings is that the extra C inputs from eCO_2 were priming the breakdown of existing soil C, as seen previously at this and other FACE sites across widely differing ecosystem types (Peralta & Wander, 2008; Moran & Jastrow, 2010; Carrillo *et al.*, 2011; Drake *et al.*, 2011; Hopkins *et al.*, 2014; Fang *et al.*, 2015), rather than offsetting the effect of heat.

The priming hypothesis also is consistent with our observation that POM-C declined from 2009 to 2011 and was lower in eCO_2 plots than in unfumigated plots (Fig. 4). The lack of an increase in POM-C with eCO_2 at this site was noted previously and attributed to priming by Peralta & Wander (2008) after 3 years of fumigation, but it is worth noting that in year 3, the difference in POM-C between treatments was not yet significant. Given that the CO_2 priming effect required most of a decade to become statistically resolvable, it is perhaps not surprising that 3 years of heating did not produce a detectable change in POM-C.

Compared to other experiments that have examined the simultaneous effects of heating and eCO_2 on soil C dynamics, SoyFACE is notable for showing no obvious heat \times CO_2 interactions. Although unheated FACE experiments have commonly showed direct effects of

eCO₂ on soil respiration (Pendall *et al.*, 2003; Pregitzer *et al.*, 2008; Adair *et al.*, 2011; Drake *et al.*, 2011; Lam *et al.*, 2014), many heat × CO₂ experiments are dominated by indirect effects (Dieleman *et al.*, 2012), which seem to be mediated by the joint effects of CO₂ and heating on soil water availability (Wan *et al.*, 2007; Pendall *et al.*, 2011, 2013; Selsted *et al.*, 2012). Our site, by contrast, showed no significant heat × CO₂ interactions, perhaps because the site is only rarely dry enough to limit respiration. Water content was consistently higher in eCO₂ plots and was lower in heated plots in 2009 and on some days in 2010, but volumetric water content never dropped below 20% and the differences in soil water were not significant in 2011, the driest summer of the study (Rosenthal *et al.*, 2014; Ruiz-Vera *et al.*, 2015). Thus, the effects of water availability on R_{het} appear to be additive to the heat effect, not a driving mechanism.

The observed changes in R_{aut} may be caused by differences in root distribution. We have no evidence of changes in total root biomass; however, minirhizotron observations from maize in 2010 suggest that elevated CO₂ affected the depth distribution of roots, with greater root length in shallow soils and lower root length in deeper soils, but the effects depended on temperature treatment (S.B. Gray, in prep). In 2009, soybeans in heated plots appeared to use deeper soil water (Rosenthal *et al.*, 2014), possibly indicating a shift of roots toward deeper soil that would have reduced the amount of root-respired C reaching the surface, thus contributing to the reduction in R_{aut} from soybeans we observed that year.

The empirical results from this study are reinforced by forward extrapolations from a process-based model which indicates that heat, either singly or combined with increased CO₂, will drive long-term losses in SOC from agricultural soils, adding to losses in SOC caused by aggressive tillage practices. This result is consistent with previous models of CO₂ × warming experiments in predicting a net loss of soil C under global warming (Parton *et al.*, 2007).

Given the difficulty of inferring SOC changes from short-term direct measurements and the number of known processes that DayCent integrates, we posit that these model results provide our best available prediction of the *direction* of future SOC trajectories in a warming climate and that that they place a lower bound on the *magnitude* of future losses as CO₂ increases. However, our modeling approach was unable to test the hypothesis of a priming effect of eCO₂ on soil C breakdown, because DayCent's SOC model has an explicitly specified turnover time for each pool. Turnover can be manually increased to simulate priming (Cheng *et al.*, 2013), but this requires a known

degree of increase. Our observation that POM-C declined under elevated CO₂ gives an indirect indication that turnover rates have increased, but is not sufficient to constrain the magnitude of the increase, especially in slower-cycling C pools. Instead, increasing model C inputs through CO₂ fertilization lead to an increase in modeled SOM with no decrease in fast C pools that would match our observed drop in POM-C. Therefore, our model results probably understate the extent of soil C losses under elevated CO₂. To produce more accurate long-term predictions of SOC dynamics under systems with active priming, models with explicit microbial processes may be needed (Wieder *et al.*, 2013).

Elevated CO₂ and temperature, both singly and in combination, appear to accelerate the loss of soil C from agricultural ecosystems, through probably distinct and potentially additive pathways. Simple measurements of whole-soil respiration were not sufficient to detect these changes, so future experimental work should routinely include partitioning of soil respiration into plant-derived and SOM-derived components. Robust predictions of CO₂ priming effects will require updated ecosystem models that contain explicit microbial dynamics.

Acknowledgements

Thank you to Michael Masters, Nicholas DeLucia, Ahbisheik Pal, and Micah Sweeney for assistance with data collection; Michael Masters and Corey Mitchell for soil C analyses; David Drag and Kannan Puthuval for maintaining the experimental site; Sharon Gray, Matthew Siebers, Andrew Leakey, and Eva Joo for sharing calibration data; and Robert Paul, Benjamin Duval, and Cindy Keogh for valuable advice about DayCent. Portions of this research were funded by the US Department of Energy's National Institute for Climatic Change Research.

References

- Adair EC, Reich PB, Trost JJ, Hobbie SE (2011) Elevated CO₂ stimulates grassland soil respiration by increasing carbon inputs rather than by enhancing soil moisture. *Global Change Biology*, **17**, 3546–3563.
- Allison SD, Wallenstein MD, Bradford MA (2010) Soil-carbon response to warming dependent on microbial physiology. *Nature Geoscience*, **3**, 336–340.
- Anderson-Teixeira KJ, Masters MD, Black CK, Zeri M, Hussain MZ, Bernacchi CJ, DeLucia EH (2013) Altered belowground carbon cycling following land-use change to perennial bioenergy crops. *Ecosystems*, **16**, 508–520.
- Angel J (2010a) Official 1981–2010 Climate Normals. Illinois State Climatologist Office, Illinois State Water Survey. Available at: <http://www.isws.illinois.edu/atmos/statecli/newnormals/normals.USC00118740.txt> (accessed 18 January 2011).
- Angel J (2010b) Climate Observations for Champaign-Urbana, IL. Illinois State Climatologist Office, Illinois State Water Survey. Available at: <http://www.isws.illinois.edu/atmos/statecli/cuweather/index.htm> (accessed 18 January 2011).
- Aref S, Wander M (1998) Long-term trends of corn yield and soil organic matter in different crop sequences and soil fertility treatments on the morrow plots. *Advances in Agronomy*, **62**, 153–197.
- Bernhardt ES, Barber JJ, Phippen JS, Taneva L, Andrews JA, Schlesinger WH (2006) Long-term effects of free air CO₂ enrichment (FACE) on soil respiration. *Biogeochemistry*, **77**, 91–116.

- Black CK, Davis SC, Hudiburg TW, Bernacchi CJ, DeLucia EH (2016) Data from: elevated CO₂ and temperature increase soil C losses from a soy-maize ecosystem. *Dryad Digital Repository*. <http://dx.doi.org/10.5061/dryad.bn7j3> (accessed 24 May 2016).
- Bond-Lamberty B, Thomson A (2010) Temperature-associated increases in the global soil respiration record. *Nature*, **464**, 579–582.
- Bradford MA (2013) Thermal adaptation of decomposer communities in warming soils. *Frontiers in Microbiology*, **4**, 1–16.
- Brye KR, Riley TL (2009) Soil and plant property differences across a chronosequence of humid-temperate prairie restorations. *Soil Science*, **174**, 346–357.
- Carrillo Y, Pendall EG, Dijkstra FA, Morgan JA, Newcomb JM (2011) Response of soil organic matter pools to elevated CO₂ and warming in a semi-arid grassland. *Plant and Soil*, **347**, 339–350.
- Cheng WX, Parton WJ, Gonzalez-Meler MA *et al.* (2013) Synthesis and modeling perspectives of rhizosphere priming. *New Phytologist*, **201**, 31–44.
- Chevallier T, Hmadi K, Kouakoua E *et al.* (2015) Physical protection of soil carbon in macroaggregates does not reduce the temperature dependence of soil CO₂ emissions. *Journal of Plant Nutrition and Soil Science*, **178**, 592–600.
- Conant RT, Ryan MG, Ågren GI *et al.* (2011) Temperature and soil organic matter decomposition rates – synthesis of current knowledge and a way forward. *Global Change Biology*, **17**, 3392–3404.
- David MB, McIsaac GF, Darmody RG, Omonode RA (2009) Long-term changes in mollisol organic carbon and nitrogen. *Journal of Environment Quality*, **38**, 200–211.
- Davidson EA, Janssens IA (2006) Temperature sensitivity of soil carbon decomposition and feedbacks to climate change. *Nature*, **440**, 165–173.
- Davis SC, Parton WJ, Dohleman FG, Smith CM, Del Grosso SJ, Kent AD, DeLucia EH (2010) Comparative biogeochemical cycles of bioenergy crops reveal nitrogen-fixation and low greenhouse gas emissions in a *Miscanthus* × *giganteus* agro-ecosystem. *Ecosystems*, **13**, 144–156.
- Davis SC, Parton WJ, Del Grosso SJ, Keough C, Marx E, Adler PR, DeLucia EH (2012) Impact of second-generation biofuel agriculture on greenhouse-gas emissions in the corn-growing regions of the US. *Frontiers in Ecology and the Environment*, **10**, 69–74.
- Dieleman WIJ, Vicca S, Dijkstra FA *et al.* (2012) Simple additive effects are rare: a quantitative review of plant biomass and soil process responses to combined manipulations of CO₂ and temperature. *Global Change Biology*, **18**, 2681–2693.
- Drake JE, Gallet-Budynek A, Hofmockel KS *et al.* (2011) Increases in the flux of carbon belowground stimulate nitrogen uptake and sustain the long-term enhancement of forest productivity under elevated CO₂. *Ecology Letters*, **14**, 349–357.
- Duvick DN (2005) The contribution of breeding to yield advances in maize (*Zea mays* L.). *Advances in Agronomy*, **86**, 83–145.
- Eswaran H, Van Den Berg E, Reich P (1993) Organic carbon in soils of the world. *Soil Science Society of America Journal*, **57**, 192–194.
- Fang H, Cheng S, Lin E *et al.* (2015) Elevated atmospheric carbon dioxide concentration stimulates soil microbial activity and impacts water-extractable organic carbon in an agricultural soil. *Biogeochemistry*, **122**, 253–267.
- Filion M, Dutilleul P, Potvin C (2000) Optimum experimental design for Free-Air Carbon dioxide Enrichment (FACE) studies. *Global Change Biology*, **6**, 843–854.
- Frey SD, Lee J, Melillo JM, Six J (2013) The temperature response of soil microbial efficiency and its feedback to climate. *Nature Climate Change*, **3**, 395–398.
- Hanson PJ, Edwards NT, Garten CT, Andrews JA (2000) Separating root and soil microbial contributions to soil respiration: a review of methods and observations. *Biogeochemistry*, **48**, 115–146.
- Hartley IP, Heinemeyer A, Evans SP, Ineson P (2007) The effect of soil warming on bulk soil vs. rhizosphere respiration. *Global Change Biology*, **13**, 2654–2667.
- Hartman MD, Merchant ER, Parton WJ, Gutmann MP, Lutz SM, Williams SA (2011) Impact of historical land-use changes on greenhouse gas exchange in the U.S. Great Plains, 1883–2003. *Ecological Applications*, **21**, 1105–1119.
- Hartmann D, Klein Tank AMG, Rusticucci M *et al.* (2013) Observations: atmosphere and surface. In: *Climate Change 2013: The Physical Science Basis: Working Group I Contribution to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change* (ed. Intergovernmental Panel on Climate Change), pp. 159–254. Cambridge University Press, Cambridge.
- Healy RW, Striegl RG, Russell TF, Hutchinson GL, Livingston GP (1996) Numerical evaluation of static-chamber measurements of soil – atmosphere gas exchange: identification of physical processes. *Soil Science Society of America Journal*, **60**, 740–747.
- Hopkins FM, Filley TR, Gleixner G, Lange M, Top SM, Trumbore SE (2014) Increased belowground carbon inputs and warming promote loss of soil organic carbon through complementary microbial responses. *Soil Biology and Biochemistry*, **76**, 57–69.
- Hudiburg TW, Davis SC, Parton WJ, DeLucia EH (2015) Bioenergy crop greenhouse gas mitigation potential under a range of management practices. *Global Change Biology Bioenergy*, **7**, 366–374.
- Jelinski NA, Kucharik CJ (2009) Land-use effects on soil carbon and nitrogen on a U.S. Midwestern floodplain. *Soil Science Society of America Journal*, **73**, 217–225.
- Keidel L, Kammann C, Grünhage L, Moser G, Müller C (2015) Positive feedback of elevated CO₂ on soil respiration in late autumn and winter. *Biogeosciences*, **12**, 1257–1269.
- Kimball BA (2005) Theory and performance of an infrared heater for warming ecosystems. *Global Change Biology*, **11**, 2041–2056.
- King JS, Hanson PJ, Bernhardt E, deAngelis P, Norby RJ, Pregitzer KS (2004) A multi-year synthesis of soil respiration responses to elevated atmospheric CO₂ from four forest FACE experiments. *Global Change Biology*, **10**, 1027–1042.
- Koester RP, Skoneczka JA, Cary TR, Diers BW, Ainsworth EA (2014) Historical gains in soybean (*Glycine max* Merr.) seed yield are driven by linear increases in light interception, energy conversion, and partitioning efficiencies. *Journal of Experimental Botany*, **65**, 3311–3321.
- Kucharik CJ, Fayram NJ, Cahill KN (2006) A paired study of prairie carbon stocks, fluxes, and phenology: comparing the world's oldest prairie restoration with an adjacent remnant. *Global Change Biology*, **12**, 122–139.
- Kuzyakov Y, Larionova AA (2005) Root and rhizomicrobial respiration: a review of approaches to estimate respiration by autotrophic and heterotrophic organisms in soil. *Journal of Plant Nutrition and Soil Science*, **168**, 503–520.
- Lam SK, Norton R, Armstrong R, Chen D (2014) Increased microbial activity under elevated [CO₂] does not enhance residue decomposition in a semi-arid cropping system in Australia. *Soil Biology and Biochemistry*, **72**, 97–99.
- Leakey ADB, Bernacchi CJ, Dohleman FG, Ort D, Long SP (2004) Will photosynthesis of maize (*Zea mays*) in the US Corn Belt increase in future [CO₂] rich atmospheres? An analysis of diurnal courses of CO₂ uptake under free-air concentration enrichment (FACE). *Global Change Biology*, **10**, 951–962.
- Leakey ADB, Uribelarrea M, Ainsworth EA, Naidu SL, Rogers A, Ort DR, Long SP (2006) Photosynthesis, productivity, and yield of maize are not affected by open-air elevation of CO₂ concentration in the absence of drought. *Plant Physiology*, **140**, 779–790.
- Lenth RV (2016) Least-squares means: the R package lsmeans. *Journal of Statistical Software*, **69**, 1–33.
- Lu M, Zhou X, Yang Q *et al.* (2013) Responses of ecosystem carbon cycle to experimental warming: a meta-analysis. *Ecology*, **94**, 726–738.
- Luo Y, Wan S, Hui D, Wallace LL (2001) Acclimatization of soil respiration to warming in a tall grass prairie. *Nature*, **413**, 622–625.
- Markelz RJC, Strellner RS, Leakey ADB (2011) Impairment of C₄ photosynthesis by drought is exacerbated by limiting nitrogen and ameliorated by elevated [CO₂] in maize. *Journal of Experimental Botany*, **62**, 3235–3246.
- Marriott EE, Wander MM (2006) Total and labile soil organic matter in organic and conventional farming systems. *Soil Science Society of America Journal*, **70**, 950–959.
- Matamala RR, Jastrow JD, Miller RM, Garten CT (2008) Temporal changes in C and N stocks of restored prairie: implications for C sequestration strategies. *Ecological Applications*, **18**, 1470–1488.
- Mayaki J, Teare I, Stone L (1976) Top and root growth of irrigated and nonirrigated soybeans. *Crop Science*, **16**, 92–94.
- Miglietta F, Peressotti A, Vaccari FP, Zaldei A, deAngelis P, Scarascia-Mugnozza G (2001) Free-air CO₂ enrichment (FACE) of a poplar plantation: the POPFACE fumigation system. *New Phytologist*, **150**, 465–476.
- Moran KK, Jastrow JD (2010) Elevated carbon dioxide does not offset loss of soil carbon from a corn-soybean agroecosystem. *Environmental Pollution*, **158**, 1088–1094.
- Morgan PB, Bollero GA, Nelson RL, Dohleman FG, Long SP (2005) Smaller than predicted increase in aboveground net primary production and yield of field-grown soybean under fully open-air [CO₂] elevation. *Global Change Biology*, **11**, 1856–1865.
- NASS (2011) Census of Agriculture Quick Stats 2.0. National Agricultural Statistics Service, United States Department of Agriculture. Available at: http://www.nass.usda.gov/Quick_Stats/ (accessed 12 December 2012).
- NRCS (2012) Web Soil Survey. Soil Survey Staff, Natural Resources Conservation Service, United States Department of Agriculture. Available at: <http://websoilsurvey.nrcs.usda.gov/> (accessed 22 March 2012).
- Parton WJ, Hartman MD, Ojima DS, Schimel DS (1998) DAYCENT and its land surface submodel: description and testing. *Global and Planetary Change*, **19**, 35–48.
- Parton WJ, Morgan JA, Wang G, Del Grosso SJ (2007) Projected ecosystem impact of the Prairie Heating and CO₂ Enrichment experiment. *New Phytologist*, **174**, 823–834.

- Pendall EG, Del Grosso SJ, King J *et al.* (2003) Elevated atmospheric CO₂ effects and soil water feedbacks on soil respiration components in a Colorado grassland. *Global Biogeochemical Cycles*, **17**, 1046.
- Pendall EG, Osanai Y, Williams AL, Hovenden MJ (2011) Soil carbon storage under simulated climate change is mediated by plant functional type. *Global Change Biology*, **17**, 505–514.
- Pendall EG, Heisler-White JL, Williams DG, Dijkstra FA, Carrillo Y, Morgan JA, LeCain DR (2013) Warming reduces carbon losses from grassland exposed to elevated atmospheric carbon dioxide. *PLoS One*, **8**, e71921.
- Peralta AL, Wander MM (2008) Soil organic matter dynamics under soybean exposed to elevated [CO₂]. *Plant and Soil*, **303**, 69–81.
- Pinheiro J, Bates D, DebRoy S, Sarkar D, R Core Team (2016) nlme: Linear and nonlinear mixed effects models. R package version 3.1-127. Available at: <http://CRAN.R-project.org/package=nlme> (accessed 16 April 2016).
- Pregitzer K, Loya W, Kubiske M, Zak D (2006) Soil respiration in northern forests exposed to elevated atmospheric carbon dioxide and ozone. *Oecologia*, **148**, 503–516.
- Pregitzer KS, Burton AJ, King JS, Zak DR (2008) Soil respiration, root biomass, and root turnover following long-term exposure of northern forests to elevated atmospheric CO₂ and tropospheric O₃. *New Phytologist*, **180**, 153–161.
- R Core Team (2016) R: A language and environment for statistical computing. Version 3.2.4. R Foundation for Statistical Computing. Available at: <https://www.R-project.org/> (accessed 11 March 2016).
- Rosenthal DM, Ruiz-Vera UM, Siebers MH, Gray SB, Bernacchi CJ, Ort DR (2014) Biochemical acclimation, stomatal limitation and precipitation patterns underlie decreases in photosynthetic stimulation of soybean (*Glycine max*) at elevated [CO₂] and temperatures under fully open air field conditions. *Plant Science*, **226**, 136–146.
- Ruiz-Vera UM, Siebers M, Gray SB *et al.* (2013) Global warming can negate the expected CO₂ stimulation in photosynthesis and productivity for soybean grown in the Midwest United States. *Plant Physiology*, **162**, 410–423.
- Ruiz-Vera UM, Siebers MH, Drag DW, Ort DR, Bernacchi CJ (2015) Canopy warming caused photosynthetic acclimation and reduced seed yield in maize grown at ambient and elevated [CO₂]. *Global Change Biology*, **21**, 4237–4249.
- Rustad L, Campbell JL, Marion GM *et al.* (2001) A meta-analysis of the response of soil respiration, net nitrogen mineralization, and aboveground plant growth to experimental ecosystem warming. *Oecologia*, **126**, 543–562.
- Schindlbacher A, Wunderlich S, Borken W, Kitzler B, Zechmeister-Boltenstern S, Jandl R (2012) Soil respiration under climate change: prolonged summer drought offsets soil warming effects. *Global Change Biology*, **18**, 2270–2279.
- Schmidt MWI, Torn MS, Abiven S *et al.* (2011) Persistence of soil organic matter as an ecosystem property. *Nature*, **478**, 49–56.
- Selsted MB, Linden L, Ibrom A *et al.* (2012) Soil respiration is stimulated by elevated CO₂ and reduced by summer drought: three years of measurements in a multifactor ecosystem manipulation experiment in a temperate heathland (CLIMAITE). *Global Change Biology*, **18**, 1216–1230.
- Subke J-A, Bahn M (2010) On the ‘temperature sensitivity’ of soil respiration: can we use the immeasurable to predict the unknown? *Soil Biology and Biochemistry*, **42**, 1653–1656.
- Suseela V, Dukes JS (2013) The responses of soil and rhizosphere respiration to simulated climatic changes vary by season. *Ecology*, **94**, 403–413.
- Thornton PE, Thornton MM, Mayer BW, Wilhelmi N, Wei Y, Devarakonda R, Cook RB (2014) Daymet: Daily Surface Weather Data on a 1-km Grid for North America, Version 2. ORNL DAAC, Oak Ridge, Tennessee, USA. Available at: <http://daac.ornl.gov/> (accessed 14 March 2013).
- Tisdall JM, Oades JM (1980) The effect of crop rotation on aggregation in a red-brown earth. *Australian Journal of Soil Research*, **18**, 423–433.
- Twine TE, Bryant JJ, Richter K, Bernacchi CJ, McConaughay KD, Morris SJ, Leakey ADB (2013) Impacts of elevated CO₂ concentration on the productivity and surface energy budget of the soybean and maize agroecosystem in the Midwest USA. *Global Change Biology*, **19**, 2838–2852.
- Vogel J, Valentine D (2005) Small root exclusion collars provide reasonable estimates of root respiration when measured during the growing season of installation. *Canadian Journal of Forest Research*, **35**, 2112–2117.
- Wall GW, McLain JET, Kimball BA, White JW, Ottman MJ, Garcia RL (2013) Infrared warming affects intrarow soil carbon dioxide efflux during vegetative growth of spring wheat. *Agronomy Journal*, **105**, 607–618.
- Wan S, Norby RJ, Ledford J, Weltzin JF (2007) Responses of soil respiration to elevated CO₂, air warming, and changing soil water availability in a model old-field grassland. *Global Change Biology*, **13**, 2411–2424.
- Wang X, Liu L, Piao S *et al.* (2014) Soil respiration under climate warming: differential response of heterotrophic and autotrophic respiration. *Global Change Biology*, **20**, 3229–3237.
- Wieder WR, Bonan GB, Allison SD (2013) Global soil carbon projections are improved by modelling microbial processes. *Nature Climate Change*, **3**, 909–912.
- Wu Z, Dijkstra P, Koch GW, Peñuelas J, Hungate BA (2011) Responses of terrestrial ecosystems to temperature and precipitation change: a meta-analysis of experimental manipulation. *Global Change Biology*, **17**, 927–942.

Supporting Information

Additional Supporting Information may be found in the online version of this article:

Figure S1. Total organic C (a) and N (b) in soils under conversion from tallgrass prairie to agriculture.

Figure S2. Maize (a) and soybean (c) grain production simulated by DayCent (black lines) and Champaign County averages from NASS (grey lines). Linear regression of DayCent vs. NASS maize (b) and soybean (d) yields.

Figure S3. Linear regression of DayCent vs. NASS grain yields for all years combined.

Figure S4. Aboveground soybean biomass C observed at SoyFACE (dots) and simulated by DayCent in 2001 (a), 2003 (b), 2005 (c), and 2007 (d).

Figure S5. CO₂ flux from plant roots and rhizosphere (R_{aut} ; a), soil heterotrophs (R_{het} ; b), and whole soil (R_{tot} ; c) at SoyFACE between June 2009 and October 2011.

Appendix S1. DayCent model fit evaluation.

Table S1. Summary of model parameters changed between phases of the DayCent model run.

Table S2. Summary of management schedule for DayCent simulations.

Table S3. Summary of DayCent parameters changed between simulated maize cultivars.